



**SCIENTIFIC ASSOCIATION OF GENETICISTS AND BREEDERS OF THE
REPUBLIC OF MOLDOVA**

*University of the Academy of Sciences of Moldova (UnASM)
Institute of Genetics, Physiology and Plant Protection of the ASM (IGPPP)*

Xth International Congress of Geneticists and Breeders

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GENERAL AND MOLECULAR GENETICS

**The Xth International Congress
of Geneticists and Breeders**

ANALYSIS OF HEPATIC GENE EXPRESSION UNDER THE INFLUENCE OF NATURAL FLAVONOIDS

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Flavonoids are a large group of natural compounds (over 10,000 have been described up to now) with important physiological roles in plants, as well as potential health benefits in humans. They are classified in a number of structural groups, such as flavones, flavonols, flavanones, flavanonols, isoflavones, prenylflavones, aurones, chalcones, flavan-3-ols etc. A number of publications have claimed health benefits of flavones on lipid metabolism and insulin resistance, and in this context their use for the prevention or treatment of non-alcoholic fatty liver disease has been suggested. As flavonoids are chemically diverse, they are not equal in this respect. It is therefore of interest to understand the effects various flavonoids have on gene expression in liver cells (both for normal and fatty liver).

The Gene Expression Omnibus (GEO) has been interrogated to identify gene expression data in liver tissues under treatments with flavonoids. Differential gene expression was assessed with the help of limma (Bioconductor), the results ordered by the adjusted p value (Benjamini and Hochberg, i.e. FDR) and the top 250 genes were used in gene enrichment analysis with the help of several bioinformatics tools: DAVID EASE, GoMiner, FuncAssociate 2.1, FatiGO, GeneMerge and OntoExpress. Where necessary, data normalization was carried out with the print-tip, loess or robust spline methods. Conversions between different namespaces (if the appropriate namespace was not available in the platform annotation file) have been carried out using the bioDBnet and Synergizer online instruments. The different gene lists and metabolic pathways identified by the datasets analyzed were compared in an effort to understand apparent similarities and differences.

There is a considerable discrepancy in the number of flavonoids analyzed as of now with respect to gene expression in general and in particular in liver cells. By far most research has been carried out with quercetin, for which 5 datasets were analyzable ("quercetin" as a keyword returned 351 datasets, while limiting the search to "quercetin AND liver" returned 283 datasets, but that number is highly inaccurate, as for quercetin the first result was relating to its effect on Caco-2 cell differentiation, the second one to the quercetin effect on cardiomyocytes, the fourth to a fibrosarcoma cell line etc; the same holds true for all other searches). Although for genistein, daidzein and catechol the number of results returned by the GEO query is higher than for quercetin, an analysis of the results showed that this was not the case (two datasets were analyzed for genistein, while for catechol accessing the data was very difficult because of the large size of the series it was part of). One dataset could be analyzed for luteolin, while for kaempferol only data on fibroblasts are available (under the name "kaemferol"). For apigenin a single dataset was available with gene expression in liver cells (GSE12716), but it could not be analyzed properly, because the expression data for the control group have not been uploaded in the database; we have contacted the authors in this respect and are awaiting for their reply. For naringenin only expression data in *Salmonella sp.* are available in the database and minimal data regarding effects on gene expression in an animal tissue seem to be available for flavonoid glucosides.

In the case of flavonoids for which more than one datasets were available (quercetin, genistein), there was considerable variety in the genes expressed, which might partially be explained by different experimental conditions, but this is probably also related to the notoriously "noisy" character of microarrays. For quercetin, from five datasets analyzed, in four no gene was significantly expressed, while in one (GSE39140), the top 250 genes were significantly expressed. For genistein in the set GSE41420 no gene was significantly expressed under treatment with the isoflavonoid, while for the GSE23523 dataset a number of 14 genes did. In the only dataset for luteolin, all top 250 genes were significantly expressed. Gene enrichment analyses with different bioinformatics tools converged only partially in their results.

Conclusions. The data available currently in GEO database are insufficient to allow a deep understanding of the flavonoid effects on hepatic gene effects, but in the case of quercetin most data suggest that its influence on gene expression is minimal.

RESEARCH ON ACTIVITY OF PROTEIN INHIBITORS, EXTRACTED FROM SEEDS OF SOYBEANS, CHICKPEAS AND BEANS FROM EX SITU GENETIC COLLECTION RELATED TO PROTEINASES OF TRYPsin, CHYMOTRYPsin AND TRYPsin-LIKE PROTEINASES FROM PATHOGENE *FUSARIUM OXYSPORUM* (SCHLECHT.) SNYD. ET HANS

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Beans are between champions from the point of view of amount of protein inhibitors contained within the plant [Richardson, 1977], but long-term consumption of those plants leads to hypertrophy of pancreas and, as a result, slows the growth of consumers, thus decreasing the nutrition value of these cultures [Мосолов, 1993]. Study on dynamics of anti-trypsin activity in ontogenesis showed that it achieved its peak at the beginning of generative period [Черникова, 2004]. However, protein-inhibitors of trypsin and chymotrypsin, extracted from different plants, suppress activity of exogenous proteinases of stain fungus also [Mosolov et al., 1976; 1982; Wilson, 1997]. An important part of defensive answer of plant to different extreme exposures is dramatic increase of synthesis of protein-nature inhibitors within plant's organism. For selection purposes it's strongly needed to take into consideration the specific nature of proteins inhibitors, contained into different genotypes, and its conjugation and balance with plant's stress resistance.

It was performed a study of protein inhibitors activity in seeds of 24 specimen of soy: Alina, Amelina, Albishoara, Arcadia, Bucuria, Beltskaya-82, Gizo, Dorinta, Druzha, Zodiac, Clavera, Kishinevskaya-16, Nordic, Lanca, Lumina, Skynteia, Timpurie, Chaika, Bucuria x Kharkovskaya 150, Kishinevskaya 337 x K-004, Kishinevskaya 337 x K-004 - parallel selection, D.V. 2852 x Arcadia, Lanca x Druzha, Kishinevskaya-16 x Khabarovskaya-53; in 15 local forms of nut - MDI 02418, MDI 02403, MDI 02419, MDI 02420, MDI 02418, MDI 02405, MDI 02446, MDI 02410, MDI 02462, MDI 02444, MDI 02487, MDI 02497, MDI 02482, MDI 02404, MDI 02438; in 19 forms of fasoli from institution's ex situ collection in the year of their reproduction - D74, D69, D88, D43, D111, D74, D64, D72, D37, D87, D85, D114, D75, D86, D71, D9, D11, D100 и D119. Protein inhibitors were extracted from flour of finely ground seeds (without rind), previously degreased by sulfur ether, extraction was performed on temperature 5-6 °C during the night from loads (?) of 1000 mg in relation with solvent 1:10. Isolated of fungi *Fusarium oxysporum* (Schlecht.) Snyder et Hans. were extracted from soybeans and chickpeas (with signs of rot on roots) in the wort agar medium. Cultural filtrate of fungi was obtained on Chapek's liquid nutrient medium during 26 days (with $t = 22-24^{\circ}\text{C}$) with constant shaking in thermostated furnace. Exo-proteinases from cultural filtrate of microorganisms from chickpeas are proteinases I, from soybeans - proteinases II.

The degree of inhibition of fungus exo-proteinases *F. oxysporum* (Schlecht.) Snyder et Hans., trypsin and chymotrypsin (commercial preparations) activity inhibition by proteins-inhibitors from seeds was judged by residual proteolytic activity, studied by Kunitz method with casein as a substrate [Хортон и соавт., 1950]. The aim of research was to define the content of inhibitors of trypsin, chymotrypsin and exo-proteinases of microorganisms within seeds of different genotypes of three kinds of beans; Ranking genotypes of trypsin inhibitors in content, chymotrypsin and Exo-proteinases (I) and (II); Identification of accessions with the content of inhibitors of microbial proteinases and plant resistance to pathogens defeat.

In all three types of seeds from bean plants were identified inhibitors with a relatively high inhibitory activity of digestive tract proteinases trypsin and chymotrypsin, and proteinases I and II of the molds. Proteinase I inhibition ranged from 19 to 100%, proteinase II inhibition - from 40 to 100%, and the trypsin inhibition was varied in the range 55-100%, and chymotrypsin 55-100% and 41-80% for chymotrypsin chickpea genotypes. Regarding the exo-proteinase I, bean genotypes also present high inhibitory activity, and it ranges from 49 to 100%; it's lower for soybean - from 27 to 79%; inhibitors activity value of bean genotypes in relation to proteinase II varies in the range 23-100% and 76-97% for soybean. And anti-trypsin and anti-chymotrypsin activity in inhibitors seeds from various genotypes of beans consists 68-100%, in case of soybean they are at the same level - 70-100%. The greatest resistance to a fungal microorganisms had genotypes of chickpea, bean and soybean, in whose seeds the total activity of the inhibitory protein was higher.

PROBLEME ECOLOGO-GENETICE PROVOCATE DE ERBICIDE ȘI PESTICIDE APPLICATE ÎN AGROCENOZELE INTENSIVE

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Conform datelor oficiale¹, din cele mai bine de 1000 substanțe (preparate) chimice cu proprietăți de erbicide și pesticide, în agricultură se folosesc ceva mai mult de 150. Sunt destule date care confirmă faptul, că erbicidele și pesticidele constituie principalii poluanți ai ecosistemelor, manifestă pronunțate potenții mutagene, cancerigene, patogene etc. Denumirea de erbicid provine din latină (lat. herba - buruiiană și caedo - ucid) și pesticid (lat. pestis - infectare, contagiune). La al III Simpozion internațional „Теоретические и практические подходы к проблеме мутагенеза и канцерогенеза окружающей среды”, organizat de academicianul H. Дубинин (Dușambe, 1976), la care au făcut comunicări cunoscuți specialiști în domeniu (Дубинин, Пашин, Куринный, Бочков ș.a.) s-a atenționat despre marele pericol pe care îl tănuiesc erbicidele și pesticidele, nu în ultimul rînd pentru sănătatea populației și securitatea alimentară. Urmăresc aceste probleme din anii '70 (comunicarea noastră la simpozionul menționat: „Исследование мутагенного действия пестицидов на возделываемых растениях *Zea mays* L.”. Душанбе. Материалы симпозиума, 1976)². Au urmat în șir de publicații cu includerea în experiențele noastre a culturilor din agroecenoze: *Pisum sativum*, *Avena sativa*, *Beta vulgaris*, *Glicina max*. Aceasta ne-a permis să demonstrăm acțiunea mutagenă asupra cariotipului diferitor specii cultivate în agroecenozele din Moldova, în care (anual) se aplică așa numite măsuri de combatere (în lipsa forței de muncă) a buruienilor. În asemenea cazuri există date, care confirmă păstrarea unor rămășițe de substanțe dăunătoare, mai cu seamă a așa numitelor rămășițe de îngurășăminte minerale, pesticide (cum ar fi DDT). Vorba este și de rămășițele acumulate (așa numitele preparate de sistem): 2,5-D; 2m-4x ș.a. Se vorbește repetat că erbicide și pesticide sînt dăunătoare, chiar și pentru plante, chiar dacă după un scurt timp ele se dizolvă, acest fapt nu schimbă situația. Cercetările citogenetice și citoembriologice în experiențele de laborator și teren deschis, cu diferite specii de cultură, pe parcursul anilor au demonstrat evident că dozele minimale (ne mai vorbind de cele exagerate, aplicate fără control) provoacă aneuploidia, defectează crossingoverul. Diferiți agenți chimici, anual aplicați în agricultură (agroecenoze), dar și în teren protejat (sere) în scopul obținerii roadei profitabile, prezintă un pericol real pentru menținerea potențialului genetic – a nivelului de fertilitate, heterozisului, imunității, a rezistenței la diferiți factori exogeni. Este vorba de soiuri, hibrizi și cultivari noi, omologați în agroecenozele moldave – poligon unional de aplicare a așa numitelor metode agroindustriale (datorită cărui fapt, pînă astăzi nu ne putem elibera de depozitele de pesticide, în popor „otrăvă”). În confirmarea celor relatate vor fi aduse date concrete.

În rezolvarea problemei la prima etapă se propune:

1. Adaptarea legii de stat care va determina roda maximal admisă a diferitor culturi în agroecenoze, teren protejat etc., în concordanță cu fertilitatea solului, cu condițiile pedoclimatice etc.
2. Producătorul - realizatorul preparatelor chimice testează și obține un pașaport agroecologic, inclusiv citogenetic (incluzînd posibilitatea preparatului de a induce mutații cromosomice și genetice) obligatoriu pentru fiecare preparat pus în realizare. Comercializatorii obțin dreptul de realizare de la o instituție abilitată, iar consumatorul se conduce de regulamentul Comisiei de Stat (pentru testarea mutagenetică, elaborarea normelor cg/ha pentru pesticide, insecticide etc.
3. De elaborat cadastru pedologic la toate tipurile de soluri, punînd accentul pe evidențierea cantităților de rămășițe a compușilor chimici (DDT ș.a.). 4) de organizat un laborator specializat în cadrul AȘM, care ar elabora instrucțiuni normative de folosire a substanțelor chimice și pentru semnalizarea fonului mutagen în agroecenoze și teren protejat (sere legumicole).

¹ Сельскохозяйственный энциклопедический словарь. Москва. Советская энциклопедия, 1989, стр. 100.

² Временные методики рекомендации по оценке потенциальной мутагенной опасности пестицидов. Пилинская М.А., Куринный А.И. М., 1980.

Vezi art. “О хромосомных и ядерно-плазменных изменениях у кукурузы и пшеницы, вызванных действием гексахлорана, паров парафина и фенола”. А. Чеботарь, С. Коптарь, А. Суружуи, Б. Бухар. ДАН СССР, М., 223, №1, стр. 213-215.

AS REGARDS TO EMBRYONIC DIFFERENCE, MULTIPLICATION AND CONDUCTED CLONING

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The embryological researches at *Angiospermae* demonstrated that beginning with zygote formation, embryonic development traverse in accordance with certain laws, that repeats in the ontogenesis of each individual. The turning point in the appearance of biological diversity at cellular and embryonic level constitutes the first division of zygote, and the appearance of bicellular proembryo (*cb + cb*). Comparative and experimental embryology of the different species of plants (Sueges, 1924; Schnarf, 1927 / 29; Johansen, 1950; Поддубная-Арнольди, 1976, 1984; Яковлев, 1971; Чеботарь, 1972; Чеботарь and others 1987a, б; Johri and others 1990a, b; Батыгина, 1987, 1997; Банникова, 1991 and others) demonstrated that the biological variability appears at the embryonic differentiation stages.

About this, speak to us the biotechnological facts, double fecundation facts, and endosperm development „in vitro” (Kranz Lovz and others, 1978). From the standing point of embryology, the individual development begins with formation and the first division of zygote.

Sexual reproduction gives birth to an organism with a potential variability and parallel to a specialized tissue -endosperm with a trophic destination. Morphofunctional variability at *Z. mays* L. (Ciubotaru, 1972 - 1998) appears in the first division of zygote, unvaried division, asymmetric with the fragmoplaste under 45 ° to the longitudinal axis of egg cells. Concomitantly were evidenced and described the ultrastructural changes in egg cell organelles (P, M, AG, RE, R and others), until fecundation in zygotogenesis period and the formation of proembryonic neoplasm. About the specific changes speak to us the metamorphosis organelles, polarization, and unequal distribution of zygote cytoplasm between the first two cells («*ca* and *cb*).

So then, embryology as well as biotechnology remains to give an answer to the key question - the time and place of the differentiated expression of genes in early ontogenesis, knowing them will give the possibility to obtain some programmed results.

**THE PERSPECTIVE PRIMERS OF TRANSPOSABLE ELEMENT ACTIVATOR USED
TO HIGHLIGHT THE MOLECULAR POLYMORPHISM OF SWEET PEPPER
VARIETIES**

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Sweet pepper – *Capsicum annuum* - is an important food crop, a spice and medicinal plant, originating in Central America and cultivated since ancient times. This culture has been described in detail in terms of morphological, geographical and biochemical diversity, but these data are not always sufficient to resolve problems related to authenticity of varieties, copyright or testing of the purity of seed lots. Therefore, the development and application of new molecular markers for identity and/or DNA-polymorphism of *Capsicum* cultivars is very actual today.

As a result, specific polynucleotide fragments using primers E 17, E 18, D 3, N 51, N 52, N 53 and N 54 were amplified. Primers E 17, E 18, D 3, N 53, N 54 had determined the synthesis of identical fragments for all analyzed genotypes: E 17 – 1000, 805 and 610 bp, E 18 – 435, 320 and 215 bp, D 3 – 310 and 190 bp, N 53 – 770, 700, 610 and 450 bp, N 54 – 530, 410 and 330 bp. However, these primers may be considered perspective for taxonomic identification of *Capsicum annuum* because the obtained electrophoreses spectra data differ from those for *Asparagus officinalis* L., *Allium cepa* L., *Magnolia* sp., *Buxus sempervirens* L., *Anethum graveolens* L. and different species of g. *Helianthus*.

Primers N 51 and N 52 showed greater discriminatory power of the analyzed genotypes. The N 51 primer has determined the amplification of a fragment of 450 bp, common for the genotypes *Galant*, *Caolin*, *Fildes*, *Excelent*, two fragments of ~ 630 and 546 bp only for *Galant*, *Caolin*, *Excelent*, and the fragment of ~ 357 bp only for *Galant*, *Caolin*, *Fildes* genotypes. The primer N 52 has determined amplification on 1 to 3 fragments. The fragment of ~ 650 bp is identical for all analyzed genomes. The fragment of 467 bp is typical for *Caolin*, *Fildes*, *Excelent*. The fragment of ~ 803 bp is specific for *Excelent*. Thus, primer N 51 can be used for molecular identification of *Fildes* and *Excelent*, and primer N 52 for *Galant* and *Excelent* genotypes.

Obtained data confirmed the earlier hypothesis that primers based on sequences of transposable elements can be used as universal markers for highlighting inter- and intra-specific DNA-polymorphism of different plant genomes.

**MOLECULAR CLASSIFICATION AND COMPARATIVE ANALYSIS OF
ARTHROBACTER GENUS PLASMIDS**

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The genus *Arthrobacter* belongs to class Actino-bacteria and includes Gram-positive, obligate aerobes bacteria that are often isolated from contaminated or toxic soils. Because of their potential use in detoxification of xenobiotics, these microorganisms have received considerable attention. Albeit that more than 48 *Arthrobacter* genomes have been sequenced so far, little is known about their plasmids and the tools applicable for the genetic manipulation of this bacteria. In the current work, the available sequenced *Arthrobacter* plasmids have been analyzed comparatively in order to identify common genes encoding replication and partition functions. At least one *parA* homolog could be identified on every *Arthrobacter* plasmid. Using the Maximum Likelihood method, the evolutionary relationships of these homologs have been inferred and the *Arthrobacter* plasmids have been classified into 4 clades. A characteristic feature of *Arthrobacter* plasmids is that not all the plasmid presented a *parB* homolog, but when it was present, the *parB* gene was separated by more than 20kb from *parA*. Within the 1 kb region upstream from the putative *parA* gene, directly repeated sequences could be identified on all *Arthrobacter* specific plasmids, with the exception of clade I plasmids. It is highly probable that these sequences function as iterons and thereby mark the replication origin. The pairwise plasmid proteome comparisons showed that the conservation between the *Arthrobacter* specific plasmid proteoms is low. Beside the *parA* gene, the clade IV plasmids share only one more ORF, namely a putative CHAP -amidase, possibly implicated in plasmid conjugation.

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THE *ARTHROBACTER* GENUS *NIC*-GENE CLUSTERS SHARE A MODULAR DESIGN

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The pAO1 megaplasmid of *Arthrobacter nicotinovorans* shows low levels of sequence similarity with other *Arthrobacter* plasmids, but shares most of its *nic*-genes with three *Arthrobacter* draft genomes: M2012083 (GI:NZ_AKKK000000000), SJCon (GI: NZ_AOFD000000000) and AK-YN10 (GI: NZ_AVPD000000000). The objective of the current study is to make an evolutionary analysis of the *nic*-cluster taking into account the arrangement and collinearity of the *nic* ORF's.

The three draft genomes of the above mentioned *Arthrobacter* strains were assembled based on the existing final *Arthrobacter* genomes using MAUVE, annotated with RAST and further aligned with Artemis. *Arthrobacter* sp. AK-YN10 (a gift from Dr. Atya Kapley, CSIR-NEERI, Nagpur, India) and pAO1 strains were grown on citrate medium supplemented with nicotine. Nicotine consumption in the medium was followed by HPLC.

The *nic*-gene cluster can be divided into five modules, each module encoding a precise step in the nicotine-pathway. For each module, a general rule can be observed: the pAO1 modules are the most complex, with a large number of genes, including transposases and insertion elements. The SJCon modules are the most simple, with a small number of ORF's and large non-coding regions. The AK-YN10 strain is somewhere in the middle, but the five modules are spread through the genome. The test for the nicotine resistance of this bacteria has shown levels comparable with the pAO1 strain. Moreover AK-YN10 can grow on nicotine containing citrate media without forming the characteristic nicotine-blue pigment.

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**THE ABILITY TO BIOACCUMULATE ZINC (II) BY CYANOBACTERIA
*SPIRULINA PLATENSIS***

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Spirulina cultivation processes based on the ability to accumulate zinc cyanobacteria growing in the presence of organic sources of this bioelements and extraction and fractionation of bioactive principles zinccomponente can be included in the process industry for the production of Spirulina biomass and active principles - sources of raw materials and nutraceutical products zinccomponente immunomodulatory drug remedies. Accumulation of technological models, based on the ability of spirulina biosorbție the opportunity to obtain the nutraceutical and pharmaceutical products zinccomponente.

Scope of the work: Assessing the ability of zinc accumulation by cyanobacterium Spirulina platensis strain CNM-CB -11 cultivation in the presence of mineral and organic compounds of Zn (II) and / or contact of biomass with this new production processes and development of biomass and fractions of active zinccomponente - raw materials for us nutraceutical supplements and immunomodulatory remedies.

Objectives and directions of investigations: Study of zinc accumulation capacity of the cyanobacterium Spirulina platensis strain cultivation in the presence of mineral and organic compounds of Zn (II) and in direct contact with each them and develoment the integrated scheme of development and production of biomass fractions principles active zinccomponente: amino acids and peptides, proteins, carbohydrates - raw materials for us nutraceutical supplements and immunomodulatory remedies.

Results: A dynamic studied Spirulina biomass accumulation of zinc in the cultivation in the presence of various concentrations of the two sources of zinc: mineral salt - chloride (II) and the organic compound - zinc acetate. It was demonstrated the effectiveness of the use of organic sources of zinc, which induces an accumulation of up to about 190% of the normal level of zinc in the background of the biomass produced and the distribution of the predominant (about 70%) in extracts of free amino acids and peptides and proteins. The results obtained have been developed two processes: the cultivation of Spirulina biomass to obtain forecasted containing bioactive and zinc as part of effective and: extraction and fractionation of bioactive principles zinccomponente: amino acids and oligopeptides, polysaccharides and proteins zinccomponente.

Conclusion: Proces for cultivation of Spirulina in the presence of organic sources of this bioelement ensure getting about 1,16 g/l absolutely dry biomass spirulina within the limits af 96,85 % absolutely dry biomass containing 190mg% zinc distributed in around 76,4% in fractions of amino acids, oligopeptides and protein.

MECANISME DE CROSS-TALK DINTRE GIBERELINE ȘI ALȚI FITOHORMONI

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Fitohormonii au un rol major în creșterea și dezvoltarea plantelor. Aceștia reprezintă molecule mici derivate din metabolismul secundar și ocupă rolul central în controlul fiziologic și architectural ale plantelor. Semnalele generate de fitohormoni sunt integrate la nivelul rețelei de gene printr-un cross-talk în timpul transducerii semnalelor. Evidențe privind cross-talk-ul fitohormonilor au fost obținute prin analiza fenotipurilor mutante. Frecvent, mutații afectați în calea de semnalizare a unui fitohormon, prezintă modificări în răspunsul altora. Scopul acestui studiu a fost identificarea genelor cu expresie diferențiată la tratarea cu giberelină și caracterizarea proceselor biologice de bază asociate cu acestea utilizând metode bioinformatiche.

Setul de date brute de expresie microarray a fost extras din baza de date GEO (*Gene Expression Omnibus*), cu identificatorul GSE39384, care conține 72 seturi de probe efectuate pe Affymetrix GeneChip, ATH1. Rezultatele conțin datele de expresie a plantulelor de șapte zile de *Arabidopsis* (plante wild-type și mutații hormonali) tratate cu șapte fitohormoni (acidul 1-amino-ciclopropan-1-carboxilic – precursorul etilenei (ET); citokinină (CK); metil-jasmonatul – un derivat al acidului jasmonic (JA); acidul abscizic (ABA); acidul giberelic 3 (GA3); brassinolide – una din varietățile brasinosteroidilor (BR)). Citirea datelor microarray brute, evaluarea calității și normalizarea datelor a fost efectuată în mediul statistic și de programare R (versiunea 3.2.1). Normalizarea a fost efectuată cu utilizarea algoritmului RMA (Robust Multiarray Analysis) din librăria *affy* obținută de pe Bioconductor.

În această cercetare pentru prima dată s-a obținut setul de gene cu expresie diferențiată la semnalul GA (în total 107 gene) și care în același timp își modifică expresia la alți fitohormoni (75 gene), dintre care 52 cu ABA, 38 cu JA, 31 cu IAA și un număr mai mic de gene pentru ceilalți fitohormoni. Se reliefează o singură genă (*SAUR50*) care își modifică expresia diferențiat la tratamentele cu 5 fitohormoni (care), 4 gene (*XTH32*, *NAC047*, *AT3G07010*, *KCS20*, *AT5G45650*) răspund la 4 hormoni, și 17 gene răspund la trei fitohormoni. Se evidențiază și se descriu 3 grupe de gene cu răspuns la GA și asociate nemijlocit cu procesele de creștere la plante: (i) genele implicate în metabolismul și semnalizarea GA (10 gene); (ii) genele care răspund de creșterea și remodelarea peretelui celular (8 gene) și (iii) 5 gene cu rol în reglarea potențialului osmotic în celulă (reglarea turgescenței). În final pentru prima dată s-a arătat rolul GA în procesul de trecere de la mitoză spre endoreduplicare.

Rezultatele obținute indică că giberelinele în diferite etape de dezvoltare ontogenetică sau la diferite nivele tisulare, precum și sub influența stimulilor mediului intern sau extern pot interacționa atât sinergic cât și antagonistic cu ceilalți fitohormoni. Faptul că tratamentul cu gibereline a prezentat cel mai mic număr de gene cu expresie diferențiată, prezentând cei mai puțini efectori finali, indică asupra faptului că semnalizarea acestora se intersectează cu ceilalți fitohormoni în special la nivel de transducere a semnalelor (între recepția semnalului în celulă și efectorii finali), unde giberelinele acționează ca un mecanism fin și complex care întrunește și modulează căile de semnalizare ale celorlalți fitohormoni și pot influența nivelurile, precum și cantitatea efecturilor finale a celorlalți fitohormoni.

THE PRIMERS FROM THE TIR OF MUDR TRANSPOSON – MOLECULAR MARKERS FOR TOMATO GENOTYPES

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Cultivated tomato is one of the most consumed vegetable crops and a well-studied object in genetics, genomics, and breeding [Foolad et. all, 2007]. Despite the fact that many tomato varieties differ in productivity, shape, size, color, taste, flavor, stress tolerance, and numerous other traits, very few polymorphisms within the cultivated tomato gene pool have been identified, even using sensitive molecular markers [Bai and Lindhout, 2007]. Because of artificial selection realized for increasing the fruits' size and shape, tomato genomes are not so various [Flint-Garcia, 2013]. Thus, although the study of genetic markers for tomato has been conducted over 30 years through various approaches, the problem of identifying new types of molecular markers, allowing genome fingerprinting of different varieties of tomato, is actual [Yang et. all, 2014].

Recent data show that a very rich source of molecular polymorphism and an ideal tool for genome analysis are transposable elements [Amar et. all, 2013; Lee et. all 2012; Sveinsson et. all, 2013]. Transposable elements are DNA fragments that can move from one genomic location to another by a process called transposition, and they make up a fairly large portion of eukaryotic genomes [Han, 2013]. In this work, we show the possibility of using the single primer PCR, based on primers to terminal inverted repeats (*TIR*) of *MuDR* transposon of maize, as a tool for genotyping tomato varieties.

As a result, use of primer E 1 allowed for identification of the amplicons 506, 403 and 338 bp that are common for all analyzed genotypes. Also, the genotypes *Jubiliar*, *Mihaela* and *Elvira* have the additional, identical amplicon of 590 bp, *Jubiliar* and *Elvira* have an amplicon of 650 bp, but *Milenium*, *Tomiș* and *Mihaela* have two additional amplicons of 265 and 180 bp. The genotype *Prestij* was highlighted through the specific fragment of 518 bp, but *Milenium* had 235 bp.

Use of the primer E 2 for the *Jubiliar*, *Prestij*, *Milenium*, *Tomiș*, *Mihaela*, *Elvira* genotypes allowed for amplification of two common fragments of 680 and 490 bp; for *Jubiliar*, *Prestij* and *Tomiș*, the fragment was 258 bp. However, not identified was any genotype-specific fragment.

Use of the primer E 3 for the *Jubiliar*, *Milenium*, *Tomiș* genotypes allowed for amplification of the identical fragment of 610 bp, but for *Prestij*, *Milenium* and *Tomiș* the fragment was 327 bp. The *Jubiliar* genotype was highlighted through two specific fragments of 360 and 273 bp, *Prestij* through three specific fragments of 657, 550 and 390 bp, *Milenium* through the fragment of 489 bp, and *Tomiș* through the fragment of 517 bp. In this way, the E 3 primer assured the highlighting of molecular polymorphism of the analyzed genotypes.

Therefore, the primers E 1, E 2 and E 3, homologous to the region *TIR* of *MuDR* transposon, can be recommended as the perspective markers for genotyping of tomato varieties.

A POTENTIAL ROLE OF *DERMACENTOR RETICULATUS* TICKS (ACARI, IXODIDAE) AS VECTORS OF TICK-BORNE PATHOGENS IN THE CHERNOBYL EXCLUSION ZONE

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Parasitological studies were carried out in the Chernobyl exclusion zone (Polesky State Radio-Ecological Reserve). It is commonly accepted that radioactive contamination of the territory has an impact on the probability of infection of animals with various vector-borne pathogens. This is associated with the weakening of the immune system of the host, as well as changes in the immune system of the parasites themselves [1]. Favorable conditions are establishing for the development of various dangerous infections of humans and animals in the body of the parasite as well as for their distribution in existing foci.

Most interest for the study represented ectoparasitic ticks species *Dermacentor reticulatus*, which have been identified as dominant in the area. *D. reticulatus* ticks are known as reservoirs for many parasitic organisms with medical and veterinary importance. At the time of the accident at Chernobyl Nuclear Power Plant, *Ixodes ricinus* was the most common tick, while *D. reticulatus* was the second dominant species [2]. But at the present time, it is *D. reticulatus* is the dominant species in the area [3].

Ticks were collected at 2 sites of Polesky state radio-ecological reserve. The radioactive contamination of the tick foci was 0.76 and 1.91 mSv/h, respectively. Tick DNA was extracted using QIAamp DNA Mini Kit (Qiagen). DNA was screened for *Babesia* and *Rickettsia* species via PCR.

For the molecular genetic screening PCR was conducted with the following primers: Rp CS.877p/Rp CS.1258n, determining the 380 bp partial sequence of *Rickettsia spp.* gltA gene, as well as BJ1/BN2 primers determining the 470 bp sequence of *Babesia spp.* 18S rRNA gene.

Altogether 28 *D. reticulatus* ticks of 81 tested individuals were infected. We have found only the *Rickettsia spp.* The overall infection rate was 34.6%, which indicates a high level of pathogens circulating in the population of ticks.

We can assume that microorganisms of the genus *Rickettsia*, and *D. reticulatus* tick species were more adapted to the conditions of radioactive contamination of the environment. Especially since high infestation with *Rickettsia* pathogens for this tick species is not typical, it seems to be the consequence of exposure of the population of parasitic organisms to changed environmental conditions. Radioactive contamination affected not only the shift of the dominant species of ticks on the territory of Radiation Reserve, but also contributed to the increase in the number of the infected individuals. Our data of relevant research over the past 15 years which prove the increase in the number of different groups of parasitic organisms in areas with high radiation activity.

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CREATING PRIMERS FOR DETECTING PHYTOPLASMA INFECTIONS IN TOMATO PLANTS

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Phytoplasma is a specific group of phytopathogenic organisms occupying an intermediate position between bacteria and viruses. A special characteristic of these mycoplasma-like organisms is the lack of a cell wall - that's why they cannot be cultured *in vitro* in cell-free media. This fact predicts some difficulties in diagnosing diseases caused by phytoplasma. Moreover, these diseases have both specific and nonspecific symptoms that are similar to viral infections. Classical diagnostic techniques, such as observation of symptoms, do not yield precise results. Electron microscopy assay of phytoplasma-infected plants is more useful but also more complicated. Molecular diagnostic techniques, especially ones based on PCR, are more sensitive and allow the accurate identification of different strains and species of phytoplasma.

Primers were designed based on sequences of two *Phytoplasma* genes: 16S ribosomal RNA gene (plus 16S-23S ribosomal RNA intergenic spacer, and 23S ribosomal RNA gene, partial sequence), and chaperonin gene. Note that on one hand the sequence of ribosomal RNA gene is relatively conservative. On the other hand, the nucleotide sequence of chaperonin gene of different *Phytoplasma* species is very variable. Thus, we created a universal pair of primers for identification of both species of *Phytoplasma* (*C. ph. solani* and *C. ph. asteris*): 16Sr245F (GTAATGGCCTACCAAGACGATG) / 16Sr245R (TTAGCCGGGGCTTATTCAT). These primers are fit for nested-PCR in combination with the primers R16F2n / R16R2. Another pair of primers for nested-PCR was created for detecting tomato plants infected with the *Candidatus phytoplasma solani*: 16Sr396F (TAGGGAAGAGCTTGCGTCAC) / 16Sr396R (CGTTGAGCGTTGCACTTAGA). This pair is less suitable for *Candidatus phytoplasma asteris* identification, having few single nucleotide substitutions.

Being variable, chaperonin gene sequences may be used for species-specific identification of *Phytoplasma*. We created two pairs of primers for *Candidatus phytoplasma solani* detection which allow carrying out of nested-PCR assay. Primers for the first round are cpn421F (AGCGCAAAGTATGATTCATCGTGG) / cpn421R (AAGAGGTAAAATTTCTTGATCGTGC). Primers for the second round are cpn200F (TTAAAGAAGGGATCGAACTTGC) / cpn200R (AAAACCTTTGGACTCATCGACA). The pair of primers cpnA261F (ATGCAGGAGCTAATCCTGTTTT) / cpnA261R (CGTACTGCAATCCTTCAACAAC) was constructed for the identification of *Candidatus phytoplasma asteris* infection in plants.

The next stage of our studies in identification of the pathogen is selecting conditions for efficient and accurate PCR using created primers.



HUMAN AND MEDICAL GENETICS

**The Xth International Congress
of Geneticists and Breeders**

BLOOD CELL GENE EXPRESSION PROFILING

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The risk of developing CVD over time can be estimated with clinical factors and family history. Patients with suspicious clinical histories, may be diagnosed by indirect methods, including nuclear perfusion imaging, computed tomography angiography, coronary angiography etc. Although coronary angiography remains the “gold standard”, this test and other have drawbacks, including radiation exposure, contrast agent allergy, nephrotoxicity, and in some cases, procedures are invasive. Therefore, the development of a blood test that reliably identified patients with CVD would have diagnostic utility (Wingrove et al., 2008).

Studies on gene expression in humans are particularly difficult due to the limited availability of tissue from well phenotyped patients, and even more so from healthy subjects. Traditional gene expression analysis for cancer and cardiovascular biomarker discovery has comprised the profiling of *in vitro* or *in vivo* tissue from tumours, explanted hearts or biopsies. The use of blood as a “surrogate tissue” that can be obtained with a minimally invasive procedure is therefore an attractive alternative. More recently, the application of whole-blood or plasma based gene expression profiling has been proposed as a novel alternative to biomarker discovery (Francisco Azuaje, 2010).

Blood cells represent a novel and promising source of molecular information (Liew, 2005; Moore et al., 2005; Francisco Azuaje, 2010), that offer the possibilities for studying gene expression noninvasively (Tsui et al., 2002; Petra et al., 2004). Blood RNA expression profiling (Whitney et al., 2003; Petra et al., 2004) might have the potential to assist in diagnosis and therapy of cardiovascular diseases because of the wide range of genes expressed by white blood cells (Tang et al., 2001; Petra et al., 2004). It has been demonstrated the ability of blood RNA to reflect molecular and physiological states of solid tissues and organs in humans (Liew, 2005; Moore et al., 2005; Francisco Azuaje, 2010). The predictive potential and applicability of this resource is rooted in the fact that there is a continuous dynamic interaction between blood cells and the organs. This interaction may induce subtle changes in the gene expression patterns of the blood cells, which actually mirror physiological modifications or stimuli at the tissue or organ levels. This strongly indicates the relevance of biosignatures extracted from blood RNA as potential “biosensors” to estimate, for example, the presence or future onset of a disease. It has been estimated that blood cells can express approximately 80% of the genes encoded in the human genome (Liew et al., 2006). A similar proportion of genes expressed in different organs, including the heart, have also been detected in peripheral blood samples (Liew et al., 2006). Moreover, blood contains platelets, neutrophils and circulating leukocytes that are associated with processes in cardiovascular diseases (Aziz et al., 2007; Xingwang Jia et al., 2012). Thus, gene expression profiling in peripheral blood could provide information on early risk factors for CVDs (Whitney et al., 2003; Xingwang Jia et al., 2012).

Diagnostic or prognostic applications of blood cell gene expression profiling have been evaluated in a diverse range of diseases (Francisco Azuaje, 2010), such as coronary heart disease (Ma and Liew, 2003), hypertension (Bull et al., 2004), Kawasaki disease (Popper et al., 2007), development of collateral circulation in patients with coronary heart disease (Chittenden et al., 2006), different types of cancer (DePrimo et al., 2003; Montero-Conde et al., 2008), lupus (Bennett et al., 2003), hepatitis C virus infection (Huang et al., 2008) and neuronal injuries (Tang et al., 2003).

In the areas of cardiovascular diseases, advances in gene expression analysis have allowed the identification of a variety of potential biomarkers, useful to distinguish between ischaemic and non-ischemic heart failure, and between hypertrophic and dilated cardiomyopathies (Kittleson and Hare, 2005; Rajan et al., 2006; Francisco Azuaje, 2010). A recent study has demonstrated that specific peripheral blood transcripts play a role in the pathogenesis of coronary heart disease and its risk factors (Tabibiazar et al., 2005; Wingrove et al., 2008). Gene expression profiling has also allowed the identification of putative biomarkers of atherosclerosis, atherosclerotic lesions, plaque rupture, vascular stress and vascular remodelling.

In summary, blood RNA expression profiling is a promising tool to discover genes that determine cardiovascular phenotypes. Our work to date has focused on the examination of blood cells gene expression profiling in patients with different CVD. Our studies were initiated with the hope of identifying genes with different expression pattern in relation to cardiovascular diseases, in order to determine possible interferences in the evaluation of the diagnostic sensitivity and specificity, as well as novel therapeutic targets. Further clinical and functional studies may provide additional insights into role of the differentially expressed genes in the pathophysiology of CVD.

**PRIMARY IMMUNODEFICIENCY THE PARTICULARITIES AND PROSPECTS OF
DIAGNOSIS IN MOLDOVA**

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Immunodeficiency diseases (IDD) lead to increasing the incidence and prevalence of different acute and chronic pathologies. The variability of IDD, caused by gene mutations or chromosomal aberrations complicates the diagnosis and therapy. IDD is a concern for public health, due to clinical complexity, difficulties of diagnosis and growth of their prevalence from 1 to 8,5 per 10000 population. About 1 – 2% of the population suffering from IDD, and in 50% of cases IDD manifests over the age of 18. Today in Moldova there are no studies in this area of medicine. Adverse ecological and social issues are important in appearance of IDD.

We are starting the study to elucidate the epidemiological, clinical, immunological and molecular aspects of IDD. We are planning to develop an algorithm of diagnosis and rehabilitation with immune drugs, including of local origin. We planned to use the screening and innovative technologies such as blood cells count, appreciation the phenotype of subpopulation of lymphocytes by flow cytometry, indirect immunofluorescence with monoclonal antibodies of M, G, A, and E types, and IgAS by ELISA and nephelometry; appreciation of phagocytic function of PMN by photon detection, estimation of the complement activity by ELISA, the creation of DNA bank, the analysis of gene mutations Btk, CD40L, TACI, WAS, ATM, NBS, SH2DI, ELANE by SSCP and PCR.

The results will contribute to an understanding of the frequency and structure of IDD and will improve the diagnosis, therapy, rehabilitation and quality of life and will decrease the morbidity and mortality.

THE MOST COMMON MUTATIONS IN PAH GENE AND EFFECTIVENES OF THEIR SCREENING IN MOLDAVIAN POPULATION

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About 1 in 7000 to 10000 babies in Republic of Moldova is born with PKU (Phenylketonuria), caused by PAH (phenylalanine hydroxylase) gene mutations. From 1989 in our country was introduced the compulsory PKU screening of all newborns through a blood test based on a fluorometric method. As a result an early diagnoses and treatment is possible, because high or unstable Phe levels can cause serious health problems. By starting early the management of Phe levels throughout entire life is possible to treat or even prevent the most symptoms. Sometimes the regular screening is performed several times because of different physiological conditions of the newborns. That is why is important to have alternative molecular screening methods, especially for prenatal diagnosis.

The main goal of our research was to identify the most common mutation in the PAH gene in Moldavian population that will permit genotype identification in more than 95% of cases. Based on literature data regarding mutational spectrum of the PAH gene (more than 500 different mutations) in neighboring populations were developed specific primers (normal and mutagenic) for all 13 exons and several introns of the 10 PAH gene mutations (R408W, P281L, R158Q, R261Q, R252W, IVS12+1G>A, L48S, R261X, G272X, IVS10-11G>A). Using these primers, through PCR/RFLP method were screened 91 PKU patients from our data base. The most frequent mutation was R408W (50,6%) followed by P281L (5,5%) and the rest of analyzed mutations with a frequency bellow 5%. All 10 mutations allowed patient genotype identification only in 54% of cases. In 35% of cases was detected only one heterozygote mutation resulting in partial patient genotype identification and in 11% of cases we did not identified any mutation – unidentified genotype. Taking into account the relative low genotype identification rate of PKU patients from Moldavian population (only 54%) we established collaboration with Prof. Thony Beat (Division of Metabolism, Department of Pediatrics, University of Zurich, Switzerland) in order to sequence the PAH gene (all 13 exons and exon/introns jonctions) in those 11% of patients with unidentified genotype and selectively in patients with partial genotype. After mutations identification in those cases will be possible to elaborate specific primers and implement the PCR/RFLP screening method.

After the screening of 10 (potentially) most common PAH gene mutations in Moldavian PKU patients we were able to identify the complete genotype in 54% of cases, partial genotype in 35% of cases and no genotype was detected in 11% of cases. Was established collaboration with Prof. Thony Beat for PAH gene sequencing in case of patients with unidentified genotype. Obtained data will shed light on heterogenic mutational spectrum of the PAH gene in Moldavian PKU patients and will allow the elaboration of screening protocols with high genotype identification rate.

THE VALUE OF MEDICAL AND GENETIC COUNSELLING IN PREVENTION OF CYSTIC FIBROSIS

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Background. The presence of patients with cystic fibrosis (CF) in the patient's family is the most common cause of addressing for medical genetic counselling (MGC), which is one of the most important stages in the prevention of CF in families at high risk.

MGC is a specialized form of medical service aimed at preventing the cases of CF in affected families. The relevance of the present research can be showed by fact is that the MGC in high-risk families with CF in Moldova has a number of features related to the difficulty of clinical and molecular diagnostics, lack of neonatal screening and of reliable data on the prevalence of CF in Moldavian population.

Aim of the study is to evaluate the role of MGC in prevention of CF in high-risk families.

Materials and Methods. Was carried out an analysis of retrospective MGC in 59 families from the Republic of Moldova, in which were 68 children from aged from 1 month to 18 years (36 boys and 32 girls) with CF on the basis of the follow-up observation from 1992 to 2013. Evaluation of the effectiveness of MGC was carried out by analyzing the reproductive behavior of the couples. The reproductive decision was made by couples themselves, who received the comprehensive information about the possibility of prenatal diagnosis of CF with informativeness of their family, i. e., the possibility of identifying the mutations in *CFTR* gene.

Results and Discussions. The share of "informative" families by the analysis of the *CFTR* gene in the studied families was 64,4% (38 families). 12 families (20,3%) were completely "informative", i. e., two mutations were identified, and 26 families (44.1%) were partially "informative", which reduced the possibility of prenatal diagnosis up to 50%. "Non-informative", i. e., were not identified both mutations in *CFTR* gene, were 35,6% of the studied cases (21 families). The results of MGC revealed the following reproductive options: in 20 families (33,9%) was made decision to continue childbearing, but only in 11 of them (18,4%) was carried out prenatal DNA diagnosis. In 6 families were born healthy children. All the "non-informative" families refused to have children (35.6%) and 19 families decided to divorce (32,2%), the IVF procedures using the donor sperm was applied in 2 families (3,4%). In 6 families (10,2%) from 59 counseled were born children with CF. This means that the efficiency of the MGC in the studied families with CF was 89.8%. Given the fact that according to worldwide scientific information the efficiency of MGC in families ranged from 60% to 90% [S. I. Kozlova, N. S. Demikova, 2007], it was shown that the MGC in the studied families with CF was effective.

Conclusions. MGC is a highly effective method of preventing of CF in families with high genetic risk of severe hereditary disease. Application of prenatal DNA diagnosis in "informative" families can allow avoiding the birth of children with CF.

MONITORING OF CONGENITAL ANOMALIES IN THE POPULATION OF REPUBLIC OF MOLDOVA

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Background: Congenital anomalies (CA) occupies the second place in Republic of Moldova among the causes of infant mortality. Regular monitoring of CA is carrying out since 1991, from 2009 work was initiated to integrate the National Register of CA of Republic of Moldova to EUROCAT. Monitoring of CA in our country is carrying out to determine the prevalence's of CA in Moldavian population, to study the changes in their prevalence's and causes.

Aim of the study – to evaluate the prevalence and sharing of CA in Republic of Moldova during the period from 2009 to 2013.

Materials and methods. For the evaluation the epidemiological approach was used. Was studied prevalence and sharing CA in Republic of Moldova on the basis of genetic monitoring during the period from 2009 to 2013. The prevalence of CA in studied period was 17,5 per 1000 newborns. The maximal prevalence of CA was registered in 2010, i. e. 19,33 per 1000 newborns, and minimal prevalence in 2012, i. e. 16,0 per 1000 newborns.

Results. In general distribution of CA by prevalence the leading place occupies multiple CA (MCA) ($25,24 \pm 2,24\%$), anomalies of musculo-skeletal system ($19,54 \pm 3,10\%$) and CA of cardio-vascular system ($16,48 \pm 2,76\%$). By studying the sharing of CA during the studied period was noted trend to increasing the prevalence of MCA from 19,8% in 2009 to 31,0% in 2013 and steady trend to decreasing the prevalence of CA of musculo-skeletal system and cardio-vascular system. Thus, in 2009 CA of cardio-vascular system accounted for 23,6% from total number of CA, and in 2013 its share was 19,0%, and the share of CA of musculo-skeletal system decreased from 21,6% in 2009 to 11,3% in 2013. The share of CA of respiratory system was minimal – $1,16 \pm 0,10\%$.

The prevalence of individual types of CA in Moldova was: Down syndrome 1,24/1000 newborns, clefts of lips and palate – 0,87/1000 newborns, spina bifida – 0,29/1000 newborns, omphalocele – 0,11/1000 newborns, esophageal atresia – 0,08/1000 newborns, which corresponds to indexes of International Register EUROCAT.

Conclusion. The data obtained by monitoring allow planning and carrying out prophylactic measures aimed to decreasing the prevalence of CA in the Republic of Moldova.

DISTRIBUTION OF MUTATIONS IN *CFTR* GENE IN PATIENTS WITH CYSTIC FIBROSIS IN THE POPULATION FROM THE REPUBLIC OF MOLDOVA

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Background: *CFTR* gene (*cystic fibrosis transmembrane conductance regulator*) is mapped on the long arm of Chromosome 7 in the region q31, is about 250 kbp in length and comprises 27 exons. Today were described 1997 mutations in *CFTR* gene.

Determining the frequency of mutations and heterogeneity of mutations which are specific for individual population having a scientific as well as practical interest.

Methods: We are investigated 141 Moldavian patients with CF aged from 1 month to 28 years (75 male and 66 female), which were addressed to the Institute of Mother and Child during the period from 1992 to 2014. To determine 9 mutations (F508del, G542X, N1303K, W1282X, R117H, G551S, R347P, R334W, R553X) in *CFTR* gene we are used the method of PCR, in 15 patients was administered DNA diagnosis for 35 – 36 mutations jointly with professionals from Center of Cystic Fibrosis, Bordeaux, France, and Department of Genetics of University Clinic in Hannover, Germany.

Results: Mutations in *CFTR* gene were identified in 61,43 % of investigated chromosomes, totally were found 17 mutations. In 59 patients (41,8%) were determined both mutant alleles. In 57 of patients (40,4%) were determined only one mutant allele, and second remain unidentified. The most common were genotypes F508del/F508del – in 47 patients, F508del combined with unknown mutation in 55 cases. Mutation F508del was revealed in 103 patients (73,05%). The allele frequencies for determined mutations were: 53,21% (F508del), 1,79% (G542X), 1,07% (2789+5G>A), 1,07% (2184insA), 0,71% (N1303K), 0,71% (3849+10 kbC- >T), 0,71% (1898+G>A), 0,71% (R334W). The frequencies of other mutations were < 0,5%. 16 mutations (excluding F508del) were determined in 8,57% studied chromosomes. 38,57% mutations in *CFTR* gene were not identified.

Conclusion: The analysis of mutations in *CFTR* gene showed that in Moldavian patients prevails "severe" and rare mutations, more than one third of them remains unidentified. To improve the DNA diagnosis of CF in the Republic of Moldova has been proposed to wide the range of analyzing *CFTR* mutations.

MITOCHONDRIAL DISORDER: LEIGH SYNDROME - A CASE REPORT.

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Introduction: Mitochondrial diseases are a group of disorders caused by dysfunctional mitochondria. They appear to cause the most damage to cells of the brain, heart, liver, skeletal muscles, kidneys, the endocrine and respiratory systems. About 1 in 5000 people has mitochondrial disorder. One of them is the Leigh syndrome - a rare progressive neurodegenerative disease of childhood with variable clinical presentation, caused by mutations in more than 35 different genes of both nuclear and mitochondria origin, involving all five respiratory chain complexes. The incidence is 1:40000 newborns.

Materials, methods and results: We report on a case of a 20 months old boy, born at term in a no consanguineous healthy parents. He developed normal till 17 months old. He started to present generalized seizures responded to valproate initially and showing liver reaction on it. Two months later the convulsive crises reappeared partially with a tremor, ataxia, general weakness and fatigue, feeding difficulty, psychomotor delay (does not sit, not walk, not speak). At the moment of the evaluation he presented poor body weight (8 kg), hepatomegaly (+2cm), macroglossia (hypothyroidism was excluded), short stature [GH- 0.91, ref.val <20 ng/ml] and anemia'sings. No family history for any genetic or neurological disorder. According to these clinical manifestations he was suspected for an inborn error of metabolism and first line investigations showed highly elevated lactate in blood [2.2-2.4mmol/l], in amino acids - high level of Ala [594, ref < 450 µmol/L], Val, Ile, Cys and Ala/Lys ratio [3.44, abnormal if >3], and partial hyperaminoaciduria; high level of LDH [743UI, ref 135-225] and CK-MB [38UI, ref.<24]. Normal creatinine kinase and transaminases were found. The ECG reveals irregular sinus rhythm. The second line diagnosis including EEG showed dysfunction in cortical structures, low convulsive threshold and EMG – reduced widespread of muscle and peripheral nerves. MRI was very suggestive by bilateral hyperintense signal abnormalities in the caudate nucleus, putamen and globus pallidus on T2 weighted images. A muscle tissue biopsy was not performed. Evaluating the clinical criteria for mitochondrial diagnosis there were counted 9 points as scoring for definite mitochondrial disorder. The diagnosis should be confirmed by muscle biopsy.

Conclusion: In all cases of early infantile progressive encephalopathy of unknown origin we should think about subacute necrotizing encephalomyelopathy. We need to develop the field of clinical and lab diagnosis including the biochemistry, muscle biopsy, enzyme assays and molecular – genetic investigations in Moldova.

**SCREENING OF CONGENITAL HYPOTHYROIDISM (CH) FOR MOLDOVAN
INFANTS WITH A HIGH PERFORMANCE TSH NEONATAL SCREENING ELISA**

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Congenital hypothyroidism (CH) results from a partial or complete loss of thyroid function. It is the most common disorder identified by newborn screening. Analysis is done by assessing Thyroid Stimulating Hormone (TSH) levels in newborns; the incidence worldwide is approximately 1 in 4,000 infants. Without immediate treatment, irreversible mental retardation, growth failure and a variety of neuropsychological deficits are inevitable. IBL International introduces its new TSH neonatal screening ELISA for determination of Thyroid Stimulating Hormone (TSH) in dried blood spots. The test is a robust and sensitive “sandwich-type” immunoassay and features excellent diagnostic performance proven by the determination of >5000 samples under routine conditions in a clinical study. Moreover, direct method comparison studies with other commercially available methods showed similar Gaussian distribution, cut-off and measuring levels. A further study involving 10.000 Moldovan infants is currently in progress. This is an important implementations as the previous performed neonatal screening for TSH in Moldova in 1989-1994 (by DELFIA Neonatal TSH) showed a frequency of 1 to 3876 newborn of CH, having a screening rate 48,24% only.

In a such a way there is an intention to determine the true frequency of CH in Moldova and to prevent it's evolution, considering that Moldova is known as region with a medium risk of Iodine deficiency in Europe.

**CLINICAL AND PARACLINICAL MANAGEMENT IN EARLY ANATOMICAL-
TOPOGRAPHICAL DIAGNOSIS OF ANORECTAL ATRESIA IN CHILDREN**

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Early anatomical-topographical diagnosis of anorectal atresia in children is still a problem and its solving requires complex implementation of clinical and paraclinical methods of examination.

To evaluate differential diagnostic informativeness of different clinical and laboratory methods of examination of children born with anorectal atresia and to check their contributions in estimation of anatomical-topographical shape of the given defect.

The results obtained in this study allow us to conclude that anorectal atresia is accompanied by involvement of two or more organ systems in the pathological process: renal, urinary and genital systems (43,7%), osteo-neurogenic caudal system (22,9%), intestinal intramural innervation (11,1%), cardiovascular system (45,9%), central nervous system and neural axis (6,7%).

In 57,4% of cases anorectal atresia was accompanied by fistulas in uro-genital system and perineum, while in 42,6% anorectal atresia was complete. Depending on the position of the rectal pouch muscles to levator muscles, 38,5% of assessed children had high atresia (supraleatory), 16,2% - intermediate (at puborectal loop level) and 45,3% - low atresia (sublevatory).

The diagnosis of checking anatomical-topographical variant of the case evolved in three stages. The first stage evaluated general and local clinical data that contributed to the suspicion of anatomical-topographical nature of the defect in question. At the second stage examination included classical radiological methods and neurophysiological imaging, outlining the preventive schedule of medical and surgical treatment. The third diagnostic stage entailed computer tomography, nuclear magnetic resonance and histo-morphological examination, favoring selection of optimal surgical variant. The final strategy of medical and surgical treatment depended on both the anatomical and topographical nature of the defect in question and concomitant malformations as well as compensatory biological potential of child.

We believe that objectification of anatomical-topographical character along with development of an optimal medical-surgical strategy in treatment of anorectal atresia in children requires individual implementation adapted to a wide range of clinical and laboratory exams, including multimodal neurophysiological examination, computer tomography, nuclear magnetic resonance and histomorphological exam. Each of them has its specific informative contribution to argue the definitive diagnosis, especially in supraleatory and intermediate anorectal atresia.

PHENYLKETONURIA IN REPUBLIC OF MOLDOVA-25 YEARS OF RESEARCH

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Phenylketonuria (PKU) is an inherited metabolic disorder, autosomal recessive (OMIM 261600), caused by deficiency of phenylalanine hydroxylase (PAH, EC 1.14.16.1) that leads to severe mental retardation. Enzymatic block caused by mutations in the *PAH* gene increases blood *Phe* levels, that leads to severe mental retardation with clinical and biochemical polymorphism. *PAH* gene is located on chromosome 12q22-24 in which was described 564 mutations.

Aim of the study was to monitor the PKU detection based on neonatal screening and the effectiveness of molecular genetic diagnosis in PKU patients from Republic of Moldova.

Materials and Methods: Selection of the study group was made in 1989-2014 and it was consisting of patients with PKU identified by neonatal screening (flourimetric method), made obligatory for all newborns in Moldova. The diagnosis of PKU was based on the results of the screening test. For molecular genetic analysis was used DNA collected from 99 Moldavian PKU patients accumulated during 25 years. Identification of 10 *PAH* gene mutations (R408W, P281L, R158Q, R261Q, R252W, IVS12+1G>A, L48S, R261X, G272X, IVS10-11G>A) was performed by PCR/RFLP method.

The results: According to neonatal screening data the frequency of PKU in Republic of Moldova is 1:7866 newborns, the average screening rate being 76,5%. According to literature data regarding mutational spectrums of *PAH* gene in neighboring populations was decided to investigate in Moldavian PKU patients 4 new *PAH* gene mutations (L48S, R261X, G272X, IVS10-11G>A) due to their high prevalence in Serbia, Romania, Turkey etc. In PKU patients were identified following genotypes: R408W/R408W (27,6%), R408W/P281L (7,1%), R408W/R261Q (3,5%), R408W/L48S (3,5%), R408W/R158Q (2,3%), R408W/ R252W (1,1%), R408W/ IVS12+1G>A (1,1%), R158Q/R252W (1,1%), R408W/R261X (1,1%), R408W/ IVS10-11G>A (1,1%), L48S/L48S (1,1%), and IVS12/K363fsdelG (1,1%). The most frequent *PAH* gene mutation in Moldavian PKU patients is R408W (50,6%) followed by P281L (5,5%), L48S (4,9%), R261Q (3,1%), R158Q (3,1%), R252W (3,1%), IVS12+1G>A (2,4%), IVS10-11G>A (2,4%), R261X (1,2%) and G272X (0,6%). The mutation detection rate in PKU patients constituted 78,7%.

In the period from 2001 until now were performed 19 prenatal diagnoses in families with PKU and in 7 cases diagnosis of PKU was confirmed by molecular genetic analysis.

Conclusion: The obtained data demonstrate a good organization of neonatal screening for PKU and molecular diagnosis of *PAH* gene used in prenatal diagnosis and personalized treatment.

MORPHOFUNCTIONAL REGULARITIES OF HEART INTRAUTERINE DEVELOPMENT

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As a basis for the study of regularities of heart morphofunctional formation and the determination of stages served the principle of heterogeneity and heterochronia of the organs' structural components development and that of different morphofunctional degree of vulnerability of the heart in different periods of ontogenesis.

In conformity with the mentioned, have been established some specific features in the heart's organogenesis on the basis of which have been differentiated 6 stages in the heart's organogenesis and development in the early period of ontogenesis.

1. The stage of heart pair primordia initiation (day 19-21, 1,5-2,5 mm). The heart, like the vascular and lymphatic systems, is a derivative of mesoderm.

2. The stage of fusion of the pair primordia of endocardium and initiation of the non-pair primordium of the heart in the form of a single cardiac tube (day 21-22, 1,5-3 mm). On day 22, the pair cardiac primordia accrete into an aggregate cardiac tube and merge with the embryo's vessels, in particular, those of chorion and yolk sac forming the primitive cardiovascular system, which ensures integrity of the growing organism. Thus, from this moment until the formation of the nervous system, the cardiovascular system functions as the organism's principal organizing system.

3. The stage of two-chamber heart formation (week 4, 2,5-5 mm). At this time, the right tube is characterized by rapid growth increasing in length faster than the pericardial cavity. As a result, some curves and extensions similar to „S” are formed. In this period, blood circulation through the heart in one direction is established. In the middle of the fourth week, in the heart is the primordia of the valves and the septum. Concomitantly, the formation of the heart's conductivity system is initiated.

4. The stage of three-chamber heart formation (day 27-35, 7,5 mm). After the 27th day, on the inner surface of the common atrium appears a crimp which grows down and forms by the 29th day the septum, that separates the common atrium into two parts – the right atrium and the left one. However, an oval window remains in the septum, and from the auricular channel the 2 atrioventricular orifices are formed.

5. The stage of four-chamber heart formation initiation (day 36-49, from 7-9 to 13 mm). On day 31 of the development of the common ventricle, the septum is formed. It grows top down and separates the common ventricle into the right ventricle and the left one. Concomitantly, the formation of the semilunar valves of the aorta and the pulmonary trunk is initiated.

6. The stage of the heart's morphogenesis completion (from the 50th day until birth).

These stages are vulnerable as the impact of intrinsic- and extrinsic factors can cause diverse heart disorders.

The regularities and intrauterine stages of the heart's morphofunctional formation can serve as a vector in the elaboration of the concept and the algorithm of heart sanogenicity creation in intrauterine ontogenesis.

ACTIVITATEA ENZIMELOR LIZOSOMALE ÎN URINĂ LA COPII CU GLOMERULONEFRITĂ PRIMARĂ

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Obiective: Determinarea activității enzimelor lizosomale în urină la copii cu glomerulonefrită primară.

Material și metode: Studiul este axat pe un lot de 40 copii cu glomerulonefrită acută (GNA), cu vârsta cuprinsă între 3-15 ani, în stadiu funcțional compensatoriu, fiind examinați la diferite etape clinico-evolutive ale maladiei. Grupul de control a constituit 20 copii practic sănătoși.

Rezultate: Studiul arilsulfatazelor A și B în urină atestă o majorare concludentă a activității la pacienții cu GNA sindrom nefritic și sindrom nefrotic, în perioada manifestărilor clinice comparativ cu grupul de control. Activitatea enzimei date în perioada de remisie la pacienții cu GNA s-a redus statistic semnificativ cu excepția pacienților cu GNA sindrom nefrotic steroidrezistent, unde valorile enzimei continue să rămână ridicate.

Astfel, schimbări mai profunde s-au înregistrat în grupul pacienților cu GNA sindrom nefrotic, steroidsensibil, unde activitatea arilsulfatazei A și B în urină a sporit de aproape 6 ori ($543,55 \pm 52,77$ nM/s.l creatinină, $p < 0,001$), comparativ cu valorile grupului martor.

Activitatea arilsulfatazei C în urină a sporit de 2,4 ori ($299,14 \pm 34,36$ nM/s.l creatinină, $p < 0,001$), comparativ cu grupul martor. Cele mai profunde modificări ale activității enzimei date sau semnalat în grupul pacienților cu sindrom nefrotic steroidrezistent, unde activitatea enzimei date a constituit ($373,49 \pm 32,08$ nM/s.l creatinină, $p < 0,001$). În perioada remisiei activitatea arilsulfatazei C scade, cu excepția grupului sindrom nefrotic steroidrezistent, unde activitatea enzimei date în urină s-a redus până la $218,65 \pm 25,98$ nM/s.l creatinină, comparativ cu grupul martor.

Activitatea β -galactozidazei în urină a sporit notabil la pacienții cu GNA sindrom nefritic și sindrom nefrotic, în perioada manifestărilor clinice, comparativ cu grupul martor. Astfel activitatea β -galactozidazei în urină la pacienții cu GNA sindrom nefritic a sporit de 3,6 ori ($327,44 \pm 42,9$ nM/s.l creatinină, $p < 0,05$) comparativ cu grupul martor ($91,36 \pm 16,85$ nM/s.l creatinină). În perioada remisiei s-a constatat o discreștere a enzimuriei cu excepția GNA sindrom nefrotic steroidrezistent, unde ea a sporit de 4,2 ori ($387,01 \pm 45,38$ nM/s.l creatinină, $p < 0,05$) comparativ cu grupul martor.

Concluzii: Monitorizarea activității enzimelor lizosomale în urină reprezintă un criteriu de diagnostic care reflectă profunzimea lezării țesutului renal, gradul de lezare a canalelor distale cât și pronosticul maladiei.

**MOLECULAR GENETIC ANALYSIS OF CFTR GENE IN CYSTIC FIBROSIS
PATIENTS FROM REPUBLIC OF MOLDOVA**

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Cystic fibrosis (CF) is the most frequent autosomal recessive monogenic disease with chronic progressive evolution, potentially lethal caused by mutations in the *CFTR* gene. The disorder has a broad range of severity but has traditionally been manifested by chronic pulmonary disease, pancreatic exocrine insufficiency, elevated concentrations of chloride in sweat, and identified mutation.

Materials and Methods: The diagnosis of CF is based on clinical features with confirmatory evidence by sweat electrolyte analysis by the method of Gibson-Cook and molecular-genetic analysis of mutations in the *CFTR* gene using PCR-RFLP method. We evaluated 142 patients suspected of CF since 2014 – 2015. The diagnosis was established on the basis of positive test results (values > 60 mmol/l of Cl⁻ and Na⁺ ions in the sweat) and molecular-genetic analysis of the most common mutations in Moldova (Δ F508, G542X, G551D, R553X, 3849 + 10kbc->T, 2184insA).

Results: We determined the Δ F508 mutation frequency in 11 individuals from the 142 patients. In the CF patients we found 8 heterozygous individuals and 3 homozygous individuals (allele frequency of 9,86%) for the Δ F508 mutation. Mutation 2184insA was identified to a person. Other mutations were not identified during this period for children suspected to FC.

Conclusions: According to the results we can reveal that Δ F508 is the most frequent mutation in patients with CF from Moldova. Spectrum of mutations in the *CFTR* gene allowed the identification of 1/3 of pathological alleles and it is proposed therefore to implement a modern methods for detecting new mutations in the *CFTR* gene for CF patients from Moldova.

NMR SPECTROSCOPY – A VALUABLE RESEARCH TOOL FROM PLANTS TO HUMANS

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The Nuclear Magnetic Resonance (NMR) spectroscopy was independently discovered in 1946 by the groups of Bloch and Purcell. In 1961 the first commercial NMR spectrometer was produced and since mid 1960's NMR spectroscopy became an indispensable tool for chemists. Today this technique is still the most powerful technique for structure elucidation of chemical compounds in solution. It was only in late 1980's that some proof of concept for its use in medical applications were published, and only in late 1990's the technique started to be used for some real medical applications. In spite of several successful examples in various medical fields, the NMR spectroscopy remains less used in medicine than it should be. On the other hand, the Magnetic Resonance Imaging (MRI) techniques became very fast established diagnosis tools in medicine, in spite of the fact that the first MRI scanners have been produced only in 1980's. Although the MRI field is much younger than the NMR spectroscopy, MRI penetrated many fields of medicine and today one cannot imagine modern hospitals without such equipments.

Some progress has been made in recent years for introducing the NMR spectroscopy in clinical practice and the result is that in a limited number of hospitals, usually in major University Hospitals in developed countries, NMR spectrometers became available. Also, some other hospitals developed collaborations with research NMR laboratories which are not located in the hospitals. The main explanation for this situation is related to difficulties in automation and interpretation of the NMR results. Thus, in spite of the efforts made by the NMR manufacturers, the spectroscopy work cannot be handled to a technician as it is done with other clinical tests. A research team is still required for the operation of NMR spectroscopy equipment as well as for the interpretation of the results. These technical factors, together with both prospects and limitations of the method will be discussed in the talk.

The present paper will give a general overview on some of the successful medical applications of the NMR spectroscopy together with examples from our laboratories. The paper will also emphasize the collaboration with the group from the Centre of Reproductive Health and Medical Genetics, Institute of Mother and Child, Chisinau on diagnosing Inborn Errors of Metabolism.

In contrast to medical applications, the NMR applications in plant metabolism and genetics are currently undergoing a more balanced development. Thus, for some applications to plants and fruits, the technique may be automated to a level rivaling with chromatography or even biochemical kits, whereas for other plants and fruits applications, full human intervention is required in all steps of the analysis and data interpretation, in a way similar to applications in the medical field.

CLASSIFICATION BY TYPE OF CONGENITAL ANOMALIES IN THE REPUBLIC OF MOLDOVA, ACCORDING TO THE NATIONAL MONITORING FOR 2014 AND 2015, AND COMPARISON WITH THE DATA OF THE INSTITUTE OF MOTHER AND CHILD AND INFORMATION ABOUT ALCOHOL CONSUMPTION AND USE AND FOLIC ACID SUPPLEMENTATION

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Background: Congenital anomalies are an important factor affecting infant morbidity and mortality, leading to early disability and having a psychological impact as well as negative economical effect on families and on state economy.

Materials and methods: In accordance with the Order of the Ministry of Health, over the past 10 years in the Republic of Moldova is the National Registry of Congenital Anomalies. During the counseling of parents of children with congenital anomalies the relatives received comprehensive information on the harmful factors, which having impact in preconceptional, prenatal and perinatal periods.

Results: 651 questionnaire has been revised which were filled out by doctors advise parents who gave birth to children with congenital anomalies in 2014. At the same time genetics, who carried out counseling the children born in the Departments of the Institute of Mother and Child, or where genetic counseling was administered, collected information on 225 cases of congenital anomalies. Work at the Laboratory of Prevention of Hereditary Diseases (LPHD) was continued in the year 2015, during which was received the information about 40 children with congenital anomalies. According to the register from the Department of Statistics, the most prevalent were anomalies of the cardiovascular system (157 cases, 24,12%; according to LPHD, this pathology is ranked third, 36 cases, 16,00%). The second place occupied the multiple congenital anomalies (121 cases, 18,59%; according the LPHD, this anomalies were the first, 62 cases or 27,56%). The third place according to the Department of Statistics took anomalies of the nervous system (98 cases, 15,05%). According to LPHD, on the third place were anomalies of the cardiovascular system (36 cases, 16,00%). During the first three months of the year 2015, according to LPHD, on the first, second and third place respectively were multiple abnormalities, skeletal anomalies and anomalies of circulatory system (30,00%, 25,00% and 17,50% respectively). According to LPHD, most mothers during pregnancy used alcohol in small quantities (38,22% and 55,00% in 2014 and 2015 years, respectively). Most of the mothers had received supplement with folic acid (53,33% and 65,00%), and the number of mothers who received folic acid prior to conception was insignificant (5,33% and 5,00%, respectively).

Conclusion: In the structure of congenital anomalies of the greatest importance are multiple anomalies, malformations of the musculoskeletal, cardiovascular and nervous systems. The flaw of folic acid supplement in Moldova is its starting on advanced stages of pregnancy.

GENETIC FACTORS THAT CAN CAUSE MALE GAMETES DISSANOGENITY

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Gametogenesis represents the initial stage of the primordial genetic program of the human health. Simultaneously, creating of the directed health is impossible without knowing the genetic factors that may cause its dissanogenity. This motivated us to determine the factors that cause male gametes dissanogenity.

The etiology of sperm DNA damage, much like male infertility itself, is multifactorial and may be due to primary testicular or external factors. These factors may adversely affect male gametes sanogenity. In our view, sperm dissanogenity includes any negative changes in the normal functionality of spermatozoa under the action of various factors, regardless of their nature.

Previous studies have estimated that at least 2000 genes may be involved in normal spermatogenesis and fertility, a strikingly high percentage of the total complement of human genes (about 10%).

Sperm DNA damage is clearly associated with male infertility, but a small percentage of spermatozoa from fertile men also possess detectable levels of DNA damage.

Chromosomal abnormalities are reported in 2,1-8,9% of males attending a fertility clinic.

Recently employed proteomic techniques, such as 2D polyacrylamide gel electrophoresis (2D-PAGE), mass spectrometry, and differential in gel electrophoresis, have allowed for the identification of numerous sperm-specific proteins. These approaches have provided a greater understanding of protein function involved in sperm processes such as motility, capacitation, acrosome reaction, and fertilization.

Studies by Naaby-Hansen et al. have shown there to be at least 1400 distinct protein moieties of which approximately 100 are certainly localized to the cell surface. Proteins on the sperm surface are critical for cellular function and are responsible for cell regulation during sperm capacitation in the female reproductive tract, binding to the ZP and induction of the signalling cascades that trigger the sperm acrosome reaction.

Studies of the sperm proteome have demonstrated how post-translational modifications, such as phosphorylation, glycosylation, proteolytic cleavages and mutations, bring about the physiological changes in spermatozoa function.

In order to ensure male gametes sanogenity are required depth studies of sperm specific proteins, mechanisms induced by oxidative stress, proteins functionality and their proteolytic regulation, identification of molecular biomarkers, that would allow determination of genetic abnormalities and sperm dissanogenity.

GENETIC CONTRIBUTIONS IN DEPRESSION MANIFESTATION

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Depression is expected to be one of the leading causes of morbidity by 2020, according to the World Health Organization (2010). Nonetheless, the current methods of treatment for depression may not be effective in reducing the global burden of this disease.

Among pathogenetic factors with psychogenic nature that leading to the development of this disease, an important role belongs to the genetic factors which is confirmed by twin studies.

Twin studies have estimated that the heritability of depression is estimated at around 30 to 50% [Rice, 2009]. At the same time, the identification of vulnerability genes has not been as successful as it has been for many other complex disorders, such as obesity.

Due to its complex nature, a number of genes are likely to be involved in the pathophysiology of depression [Erin Bakshis Ware, 2014]. This may involve genes concerned with the function of neurotransmitters and receptors.

The combination of advances of understanding of human genomic variation and cost effective genotyping techniques have led to growth in molecular genetic studies of depression and other complex psychiatric phenotypes. These studies typically examine whether specific alleles or genotypes are associated with the phenotype of interest. Until recently, genetic studies of depression focused largely on candidate genes – that is, genes that are hypothesized to be implicated in the neurobiology of depression. Some of the most commonly studied candidate genes have been those regulating serotonin and dopamine neurotransmission, given the suspected involvement of these neurotransmitters in the pathophysiology of depression and their role as targets of antidepressant drugs [Dunlop and Nemeroff, 2007; Thase, 2009; Erin C. Dunn et al., 2014].

In the manifestation of depression, according to the results obtained by Plomin et al. (1977), Monroe and Simons (1991) and Belsky (1997) can play an important role gene-environment correlations, that may have *passive* character (referring to the association between the genotype a person inherits from their parents and the environment in which that person is raised), *evocative* character (also known as “reactive”, referring to the association of an individual’s genetically predisposed behaviors and other people’s reactions to those behaviors), and *active* character (also known as “selective”, referring to the relationship between an individual’s genetically influenced behaviors and the environmental exposures that an individual selects) [Plomin et al., 1977; Jaffee and Price, 2007].

Gene-environment correlation in the development of mental disorders has been demonstrated by Caspi et al. (2010), and Uher and McGuffin (2008), who argue that the experiences of maltreatment during childhood are associated with mental health disorders, possibly as a consequence of the moderating effect of specific genes on the risk conferred by such early traumatic events [Caspi et al., 2010]. Thus, maltreated children with the serotonin transporter (SERT) genotype conferring low levels of the SERT molecule (5-HTTLPR S allele carriers) seem to have an increased risk for depression compared to maltreated children with the 5-HTTLPR L/L genotype [Caspi et al., 2010; Uher and McGuffin, 2008].

Results obtained by Blanca Gutiérrez et. al. (2014) confirm that the increased risk for major depression disorder (MDD) conferred by childhood maltreatment is modified by variation at both SERT and BDNF genes. In particular, individuals who carry both the SERT S allele and the BDNF Met allele appear to be more vulnerable to the impact of childhood maltreatment on mental health.

However, we mention that the path forward to detect genetic risk loci for depression remains challenging, what is certain is that a deeper understanding of the etiology of depression is needed. Given the enormous burden of depression, identifying its genetic underpinnings may be essential to preventing the on-set of this disorder and improving the lives of those who already suffer [Erin C. Dunn et al., 2014].

Till now, it remains unclear whether genetic and environmental risk factors converge on a unitary molecular mechanism underlying MDD or MDD consists of a heterogeneous group of disorders with multiple causal factors and distinct molecular mechanisms. To answer these questions need further investigation at the genetic level involving bioinformatics tools.

GENETICA EPILEPSIEI

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Scopul. Testarea clinico-genetică a epilepsiei, prin analiza ADN-ului pentru mutații specifice cu scop de diagnostic sau ca testare predictivă.

Introducere. Cercetările genetice asupra epilepsiei au dezvăluit peste 20 gene cu efect major asupra susceptibilității la epilepsia idiopatică. Cel mai important potențial aplicativ al acestor descoperiri este testarea genetică: utilizarea informației genetice pentru confirmarea diagnosticului la pacienți deja cunoscuți sau suspecți de a avea epilepsie (*testare de diagnostic*), sau pentru a prezice debutul patologiei în cazul persoanelor cu risc de a dezvolta epilepsia din cauza istoricului familial (*testarea predictivă*).

Metodele moleculare pentru testarea genetică a epilepsiei.

- Reacția de polimerizare în lanț (PCR) prin care se realizează amplificarea selectivă și rapidă a secvențelor țintă ADN (fragmentelor de interes).

- În metoda Real Time PCR fragmentul amplificat (ampliconul) este vizualizat pe măsură ce procesul de amplificare înaintază. Aceasta urmărire în „timp real” a procesului de amplificare este posibilă prin marcarea cu molecule fluorogenice (fluorocromi) a primerilor, probelor sau ampliconului.

- Secvențierea, constă în determinarea secvenței nucleotidice a ADN-ului pentru întreaga genă sau o parte din ea.

- Hibridizarea fluorescentă „*in situ*” (FISH).

- Polimorfismul unei singure nucleotide (SNP), tehnică utilizată pentru determinarea variațiilor unei singure nucleotide la nivel de genom.

Rezultate. La ora actuală au fost descoperite mutații într-un număr mare de gene cu efect major asupra susceptibilității la variate forme de epilepsie cu transmitere mendeliană și cauză idiopatică.

Genele identificate în sindroamele epilepsiilor idiopatice: KCNQ2, KCNQ3, SCN2A, STXBP1, ARX, STK9/CDKL5, SCN1A, SCN1B, GABRG2, PCDH19, SLC2A1, GABRA1, EFHC1, CHRNA4, CHRNA2, LGI1, și altele.

Potențialele beneficii ale testării genetice sunt multiple. Un test pozitiv poate confirma diagnosticul, furniza un prognostic important și informații privind tratamentul, iar prin urmare posibilitatea de a feri pacienții și familia de la teste invazive și extensive. Unii pacienți pot obține o explicație din punct de vedere genetic a acceselor lor sau a membrilor familiei lor.

În privința diagnosticului predictiv, un rezultat negativ al testării genetice reduce anxietatea pacientului și necesitatea de a monitoriza accesele de boală.

Concluzii. Una dintre cele mai promițătoare direcții ale cercetărilor genetice în epilepsie este farmacogenomica: căutarea variantelor genetice asociate cu răspunsul la tratament (eficiența și toleranța).

Testele genetice pentru variantele asociate cu răspunsul la tratament ar trebui să aibă efecte benefice evidente și există probabilitate mare de a fi introduse în practica clinică odată identificate și confirmate.

Consilierea genetică pre- și post-testare este crucială pentru a ajuta pacientul să înțeleagă rezultatul testului și să-l perceapă în contextul circumstanțelor sale de viață.

CASE REPORT OF PRENATAL DIAGNOSIS OF DUCHENNE MUSCULAR DYSTROPHY USING DIFFERENT SETS OF PRIMERS

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Introduction. The monitoring and prevention of hereditary diseases are the current tasks of medical genetics and health. Prenatal diagnosis (PD) is one of the methods proposed. Muscular dystrophy Duchene/Becker (DMD/B) is neuromuscular hereditary disorder that its frequently encountered in Moldova and can be detected through prenatal diagnosis (PD). This disease affecting 1 in 3500 boys by mutations within DMD gene. The patients exhibit pathological deletions in approximately 60% of cases. Since 1988 existing reactions for the multiplex PCR amplification of exons in the dystrophin gene have been modified. We aimed to verify the reliability of the multiplex PCR amplification using two different primers sets in order to define the borders of the deletions in one family with one affected boy, during PD in this family.

Methods. After medico-genetic consultation pregnant women of this family with high risk of hereditary disease DMD/B undergo PD, by amniocentesis and then multiplex PCR for direct diagnosis and PCR/RFLP for indirect diagnosis using polymorphic sites (pERT 87-8 and 16 intron). Gender determination is performed by amplification of specific sectors of the Y chromosome (AZF). Three Multiplex PCR was performed on DNA from the patient sample previously identified as sick to amplify the 29 DMD gene exons and on fetal DNA using combined sets of primers designed by Chamberlain, Abbs and Ashton.

Results. The results of indirect prenatal diagnosis show that family is not informative by polymorphic sites. Gender determination test showed that the fetus is male. In this situation only through Multiplex PCR we can determine if the fetus is also sick or not. Deletions of 45 exon by Chamberlain and 75 exon by Ashton have been found in the affected boy, but deletion of 45 exon by Ashton primers is not confirmed. The length of 45ex (498 pb) amplified using Chamberlain's primers are more extended than 45ex (369 pb). Site of deletion is located on distance between start of 45ex primer designed by Chamberlain and start of 45ex primer designed by Ashton. Based on this we were able to do molecular analysis of fetal ADN which showed that he has no deletions under both sets of primers.

Conclusion. Molecular diagnosis, especially for prenatal diagnosis, is a procedure with individual character for each person. This must be done carefully and thoroughly. MPCR analysis is a reliable method for deletion detection but a noncontiguous and single-exon deletions within DMD gene should be interpreted with caution and confirmed with another technique or using alternative sets of primers to exactly mapping the end point of deletions. This aspect helped us to provide an exact diagnosis.

**MEDICO-GENETIC COUNSELLING IN THE MUNICIPAL CENTER OF
PERINATOLOGY OF CHISINAU**

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Currently, medical-genetic counseling, along with other genetic services, has a growing practical weight in healthcare system of the Republic of Moldova. This service ensures prevention prenatal of genetic disease with subsequent appropriate genetic counseling and postnatal diagnosis of genetic pathologies as well.

In the study there were used the data collected in the PMSI MCH, the Center of Perinatology, Consultative department, in the period June 2013 – February 2015. As indications for medical-genetic counseling were used:

a) Pregnant: age greater than 35 years; with aggravated obstetric history; after biochemical or ultrasound screening; with complicated somatic history; with aggravated heredocolateral history; after taking medicines and the use of other harmful substances during pregnancy; with indications for abortion;

b) couples for preconception genetic counseling;

c) children with suspicions for various genetic disorders: chromosomal aberrations; monogenic diseases; metabolic disorders; development disorders; embryo- / fetopathies.

Medico-genetic counseling was aimed on evaluation of anamnesis (general, personal, familial and gestational anamnesis), preparation of family trees, physical examination, and clinical diagnosis, indication of laboratory examinations and genetic tests, evaluation of the results of investigations, counseling and final etiologic diagnosis.

In the covered period, there were carried down 1342 medico-genetic consultations: for 1113 couples and 229 children.

In 71 pregnant was determined the risk for congenital malformations, as a result of influence of environmental factors, by detailed studying of personal somatic anamnesis, general examination of systems, assessment of exposure to various environmental harmful factors, and questionnaires. As the result of ultrasound screening there were recommended 9 abortions after detecting malformations incompatible with life.

In children included in the study were suspected: chromosomal aberrations - 46 cases, monogenic diseases - 40 cases, metabolic disorders - 51 cases, congenital developmental disorders - 37 cases, other – 55 cases, confirmed by biochemical, ultrasound, MRI tests and cytogenetic and molecular tests as well.

After medical-genetic consultation, appropriate genetic counseling to all applicants was given. Medical-genetic counseling represents the initial stage of screening of chromosomal aberrations in pregnant women and is the primary method of prevention of genetic diseases. To confirm suspicions for genetic abnormalities in children it is necessary to perform laboratory and genetic testing.

ASSESSMENT OF GSTM1, GSTT1, GSTP1 AND MTHFR POLYMORPHISMS IN PREGNANCY LOSSES IN MOLDOVA

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Introduction. Pathogenesis in pregnancy loss (PL) is complex and involves the interaction of multiple genetic and environmental factors. Genetic factors account for approximately 5% of pregnancy loss. It is supposed that genetic polymorphisms in glutathione S-transferases (GSTs)-major detoxifying enzymes and *MTHFR*- the most critical enzyme in the folate metabolism, catalyzing irreversible conversion of 5,10-methylenetetrahydrofolate (5,10-MTHF) to 5-methyltetrahydrofolate (5-MTHF), which is the predominant circulating form of folate and all together may be associated with the risk of pregnancy loss. In this study, we aimed to investigate a possible involvement of the polymorphism of *GSTM1*, *GSTT1*, *GSTP1*, *MTHFR A1298C* *MTHFR C677T* in the pathophysiology of PL.

Materials and Methods. The prevalence of these polymorphisms was compared in 163 women with two or more pregnancy losses (study) and 75 women without history of miscarriages and with 2 healthy births (control). Also have been investigated DNA of 100 healthy moldavian children aged up to 17 years, in order to make population study. Have been used molecular-genetics methods (PCR and PCR-RFLP methods). Statistical analysis-www.gen-exp.ru.

Results. The higher proportion of the *MTHFR 677TT* (OR=1.15), *MTHFR 677CT* (OR=1.30) and *GSTT1 null* genotype (OR=1.16) was found in the study group comparing with the control group, however, the difference was not statistically significant. The prevalence of *GSTT1 null/MTHFR 677CT* and compound *677CT/1298AC* was higher in the study group than in the control group (14 versus 8% and 26 versus 23%, respectively).

The comparative analysis performed in the healthy population of Moldova and other countries have been found that the frequency of *GSTM1* (50%-null genotype and 50% *GSTM1* positive) in Moldova is similar to there from Ukraine, Italy, Russia, Egypt and Brazil ($p>0.05$). *GSTT1* genotype through Moldavians (64% - *GSTT1* (+) and 36% - *GSTT1* 0/0) differs significantly from the population investigated previously ($p<0.05$). Frequency of the *GSTP1* polymorphism (*GSTP1 Ile/Ile*=37%; *GSTP1 Ile/Val*=53%; *GSTP1 Val/Val*=10%) differs from the same data of Ukraine, Russia and Turkey.

The frequency of genotypes of *677CC*, *677CT* and *677TT* of *MTHFR* gene among the moldavian healthy children is the following: 49%, 38%, 13% respectively, being similar with European countries data (Romania, Croatia) and different from Asia (China, India, Korea, $p<0.05$). *MTHFR A1298C* polymorphism differs from China ($p<0.05$), but is similar with Romania, Greece, India, Thailand's data.

Conclusion: The risk of PL is higher for women carrying *MTHFR 677TT*, *MTHFR 677CT* and *GSTT1 null*. The distribution of *MTHFR*, *GSTM1*, *GSTT1*, *GSTP1* gene polymorphisms in healthy population of Moldova resemble European countries.

PARTICULARITĂȚI CLINICO-EVOLUTIVE ÎN BOALA CRONICĂ DE RINICHI LA COPII

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Introducere. Boala cronică de rinichi (BCR) reprezintă o problemă majoră de sănătate publică la nivel mondial. Diagnosticul precoce al pacienților cu anomalii reno-urinare, glomerulonefrite, infecții urinare ar permite aplicarea unor strategii terapeutice preventive care să susțină atât creșterea și dezvoltarea psihică/intelectuală, cât și confortul social al pacientului, până în momentul inițierii tratamentului substitutiv renal.

Scopul studiului a fost analiza retrospectivă a etiologiei, aspectelor clinico-evolutive, unor complicații ale BCR pe un lot de pacienți cu vârste cuprinse între 6-18 ani.

Material și metodă. Studiul s-a bazat pe datele anamnestice, clinice și biologice culese din arhiva secției de Nefrologie, centrului de Hemodializă al IMSP IMȘC (foile de observație clinică, fișa de evidență a copiilor dializați). Au fost incluși în studiu 55 de pacienți cu boală cronică de rinichi, stadii I-V. Rata filtrării glomerulare (RFG) a fost calculată după formula lui Svart.

Rezultate și discuții. Dintre factorii demografici am luat în calcul sexul și vârsta copiilor, considerând că aceștia pot avea impact important asupra evoluției RFG. Se observă ponderea mai mare a băieților (58,8%), se constată că peste 85% dintre pacienți incluși în lotul studiat au vârsta între 11-18 ani. După mediul de proveniență s-a constatat o predominanță a pacienților din mediul rural (64,7% pacienți), față de cei din mediul urban (35,3% pacienți). După coroborarea rezultatelor anamnezei detaliate, explorărilor clinice și biochimice cu cele imagistice (ultrasonografie, scintigrafie, urografie intravenoasă sau pielografie ascendentă, TC, arteriografie) s-au depistat următoarele boli de bază, cauzatoare de boală renală cronică: uropatii malformative (35,5%), urmate de aplazie/hipoplazie renală (8,8%), rinichi solitar (6,5%), reflux vezico-ureteral (14,1%), glomerulonefrite primare (9,2%), nefropatii tubulointerstițiale (6,4%), și altele. Astfel s-a constatat, că boala renală cronică la copil este diferită față de cea de la adult. Ceea ce predomină la copilul de vârstă mică sunt malformațiile renale sau reno-urinare, unele boli ereditare, cu care acești copii se nasc - 45 (82,4%) de cazuri, la copilul mare crește rolul maladiilor dobândite ca glomerulopatiile, pielonefrita. În grupul de pacienți cu uropatii obstructive predomină băieții, în grupul cu malformații reno-urinare și nefrită predominau fetițele. La peste 1/2 copii cu BCR sa constatat prezența pielonefritei cronice secundare, care este indicator de agravare a bolii cauzale.

Manifestările clinice au fost foarte variate. Modalitățile principale de prezentare a BCR la copii din lotul de studiu: infecții de tract urinar recurente, enurezis, hematurie recurentă, retard staturo-ponderal, modificări la ecografia abdominală (modificări calice-bazinete, parenhim renal, displazie, hidronefroza). Semnele clinice cele mai frecvente: semne generale ca astenie, fatigabilitate, slăbiciune generală la toți pacienții; la 1/3 din cazuri au fost prezente paloarea, modificări de culoare ale unghiilor, gură uscată, gingivită, anorexie, greață; la 1/2 cazuri semne neurologice ca oboseală, insomii/somnolență, cefalee; sindrom hemoragipar – în 16,7% cazuri. La circa 1/3 copii cu BCR, în urma evaluării indicelui masă corporală, s-a înregistrat retard staturoponderal. Falimentul creșterii, anemia sunt complicațiile severe ale BCR și dependente de stadiul bolii: cu cât mai avansat este procesul patologic, cu atât mai semnificativ este falimentul creșterii și dezvoltării. Examenele de laborator au evidențiat: anemie, sindromul de retenție azotată. După monitorizarea biochimică a funcției renale pacienții din lotul de studiu au fost incluși în stadii diferite de evoluție ale BCR, în funcție de nivelul seric al creatininei. Majoritatea pacienților se aflau în stadiile inițiale ale bolii: 51% în stadiul I cu RFG >90 ml/min, dar cu un istoric și dovezi ale afectării renale cornice; 34% în stadiul II cu RFG 60-89 ml/min; 10% în stadiul III cu RFG 30-59 ml/min; 3% în stadiul IV cu RFG 15-29 ml/min, și 2% pacienți în stadiul V care se aflau la dializă.

Concluzii.

Malformațiile renale sau reno-urinare sunt cele mai frecvente cauze de BCR la copilul mic, în timp ce la copilul mare - maladiile dobândite ca glomerulopatiile și nefropatiile tubulointerstițiale.

Modalitățile cele mai frecvente de prezentare a BCR la copiii din lotul de studiu: infecții de tract urinar recurente, enurezis, hematurie recurentă, retard staturo-ponderal, modificări la ecografia abdominală (modificări calice-bazinete, parenhim renal, displazie, hidronefroza).

Copilul cu BCR prezintă o constelație de semne și simptome clinice subtile: oboseala, stări de rău neexplicate, apetitul diminuat, grețuri, vărsături recidivante, prurit, boli osoase mai puțin obișnuite, absența performanțelor școlare.

Boala renală cronică asociază frecvent tulburările de creștere, anemie, sindrom hipertensiv.

**ПЕРСОНАЛИЗИРОВАННЫЕ ПОДХОДОВЫ К ДИАГНОСТИКЕ, ПРОФИЛАКТИКИ
И ЛЕЧЕНИЮ ЗАБОЛЕВАНИЙ ЧЕЛОВЕКА**

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В настоящее время меняется парадигма общественного здравоохранения в сторону развития персонализированных подходов к диагностике, профилактики и лечению заболеваний человека.

В основе персонализированной медицины лежат достижения и открытия в области молекулярной биологии и генетики человека в течение последних 10-15 лет. Эпохальным событием в этой сфере – программа по секвенированию генома человека, которая завершилась в 2003 году. Одним из важных итогов программы ГЕНОМ ЧЕЛОВЕКА явилось установление роли однонуклеотидных полиморфизмов, лежащих в основе генетического разнообразия людей, исследования которых открывают уникальные возможности для предиктивной и персонализированной медицины.

Основными молекулярно-биологическими методами, используемым в практике для детекции однонуклеотидных полиморфизмов (SNP) являются:

1. Полимеразная цепная реакция с детекцией в режиме реального времени.
 - В медицинской лаборатории INVITRO DIAGNOSTICS используется генетический анализатор 7500 Real-Time PCR Systems, Applied Biosystems (США).
 - Выделение нуклеиновых кислот производится на автоматическом экстракторе InnuPure C16 Automated nucleic acid extraction, Analytik-jena (Германия).
2. Секвенирование
3. Биочипы

**A NATURAL HISTORY OF BOY WITH DOUBLE MICRODELETIONS
AT 16P11.2-P12.1**

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The LCR-rich proximal chromosome 16p has been shown recently to be associated with the genomic disorders: the 7.1-8.7 Mb microdeletion 16p11.2-p12.1, and the proximally adjacent, recurrent 593 kb microdeletion and microduplication in 16p11.2 found in ~1% of patients with autism. We present a patient with developmental delay, short stature, and dysmorphic features observed by us during 27 years. Using metaphase HR-CGH, we identified a deletion in 16p11.2-p12.1 and verified it by FISH with BAC clones. Whole genome array CGH with ~385,000 oligonucleotide probes (NimbleGen) defined the deletion between ~22.482.580-29.342.610 bp (6.8 Mb). Unexpectedly, we identified a second deletion ~600 kb in size, mapping ~750 kb distal to the first one. The breakpoints of both deletions map within nonhomologous sequences and we propose that the deletions arose through Fork Stalling and Template Switching (FoSTeS) replication-based mechanism. Detailed evaluation of morphological phenotype revealed : dolichocephaly, long face with low midface*, high and diffuse frontal hairline, narrow and flat forehead, medially diffuse eyebrows, hypertelorism*, narrow low palpebral fissures, ptosis, short and sparse upper and lower eyelashes strabismus, broad, protruding* nasal root, long back and pear-shaped tip of the nose, thick nasal wings, broad nasal septum, narrow mouth fissure, everted, upper mucous lip inverted lower lip, downturned mouth angles, flat and broad palate. Malocclusion. Posteriorly inclined ear insertion line, prominent helix root, broad descending helix edge were seen. Angulated corpus anthelicis with broad, deep incisura intertragica, flat tragus, prominent antitragus thick and long ear lobes. Short and broad neck. Narrow and long thorax with scoliosis. Broad metacarpus, short fingers, clinodactyly 5th and campodactyly 5th of the left hand. Short legs with asymmetry (L<P), genua valga, left pes equinovarus adductus, very small. hands and feets short toes with tibial deviation at left feet. Behavioral phenotype showed his verbal communication in the form of single words and an ability to distinguish his favorite clothes and meals. He had good emotional relation with his parents, as well as with several people of his age. His behavior is pleasant and corresponds to the current situation. His interest in swimming sport was effective giving him vice master prize in 2014 whorld championship for disabeled peoples.

ANALYSIS OF CLINICAL ASPECTS OF CONGENITAL MALFORMATION OF THE BRAIN

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Cerebral malformations are a great problem in neurogenetics. More than 2000 different congenital malformations of the brain have been described in the literature, and their incidence is reported to be about 1 percent of all live births.

The importance of this article emerges from the fact that, both in the world and in the Republic of Moldova, morbidity and mortality of children with congenital cerebral malformation is higher in the first year of life, with an estimated frequency of 10-25% of the all of malformations, and 40% of deaths.

The congenital brain malformation is an outcome of chromosome or gene abnormalities, unfavorable environmental factors, including infections, and teratogens, such as drugs and nutritional deficiencies. In the majority of cases, the precise etiology is unknown and is likely to be multifactorial, involving genetic and environmental factors.

The underlying etiology is unclear in over 60% of structural anomalies of congenital cerebral nervous system (CNS), but inherited factors account for 20%; chromosomal anomalies - 10%; and environmental factors - 10%. Magnetic resonance has become the imaging procedure of choice in the evaluation of congenital central nervous system.

The objective of this study was to determine the prevalence, the nature and clinical manifestations of congenital malformations of the brain. The study was conducted on a sample of 55 patients, aged between 2 week – 12 month, diagnosed with congenital brain malformation. All the children involved in the study were examined by complete neurological examination and supplementary investigation: neurosonography, computed tomography and magnetic resonance imaging.

Results: It was concluded that agenesis is the most common congenital brain malformation in children aged less than 1 year (42%). The agenesis of corpus callosum is often associated with agenesis of the septum pellucidum (11%), in which some clinical manifestation were vision impairments, low muscle tone (hypotonia), poor motor coordination. Microcephaly was met in 22% of cases being expressed clinically by mental retardation and epilepsy.

The most rare congenital brain malformations were hydranencephaly (5%), anencephaly (5%), hemimegalencephaly (2%) and microgiria (2%).

All the patients was examined by neuroimaging (computed tomography and magnetic resonance imaging), used to confirm the diagnosis of congenital malformations to the brain.

The most common clinical manifestations congenital malformation of the brain were: neuropsychic retardation of varying degrees (91%), disorders of muscle tone (50%), partial seizures/infantile spasms (36%), hemianopsia (5%), nistagmus (5%), ataxia (4%).

Congenital malformations affect neurological development and reduce the ability of the children leaving a severe prognosis, in many cases. The prognosis of children with cerebral brain malformation depends on the type and severity of the defect. Many congenital defects cause minor neurological impairment. Others are so severe that they are fatal before or soon after birth. Most congenital brain defects carry out a very poor prognosis.

Some cases of congenital brain malformations can be prevented with good maternal nutrition and, including folic acid supplements (800 micrograms of folic acid daily). Folic acid is a vitamin that has been showed to reduce the incidence of neural tube defects. This should begin at least one month before getting pregnant as well as during pregnancy.

Pregnant women should avoid exposure to infection, especially during the first trimester of pregnancy. Abstinence from drugs and alcohol during pregnancy, quitting to the smoking before getting pregnant, reach a healthy weight before getting pregnant, keeping under control the level of blood sugar before and during pregnancy, all may reduce the risk of congenital brain malformations.

OUR FIRST RESULTS OF TARGETED ARRAY CGH IN PRENATAL DIAGNOSIS

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Microarray-based comparative genomic hybridization (array CGH) techniques have revolutionized cytogenetic diagnostic since they can detect chromosome imbalances below the resolution of conventional karyotyping with high precision. Furthermore, the expanding and improving of available array platforms, as well as, the continuously improving of the decision algorithms for classifying copy number variants (CNVs) to clinically significant, benign or of unknown significance, has led to the implementation of array analysis also in prenatal diagnosis.

Since 2012, 138 prenatal cases were referred to our lab in order to be analysed by array CGH. In parallel we also performed conventional karyotype for all the samples and the results were compared. BAC array CGH was applied on DNA extracted from chorionic villus samples or amniotic fluid cells and the data were analysed by BlueFuse Multi software. Referral reasons included abnormal ultrasound findings, or high risk combined first trimester screening. Two cases had a previous abnormal family history.

Out of the 138 cases tested, 17 (~12%) had abnormal results (CNVs alterations) that we classified as: a) abnormal results confirmed by both techniques, b) normal karyotype but abnormal array CGH, c) *de novo* apparent balanced translocation by karyotype but abnormal array CGH.

Array CGH is a valuable tool in prenatal diagnosis, increasing the detection rate of structural abnormalities not detected by conventional cytogenetic analysis and providing rapid and accurate results for targeted genomic regions. Considering the implications and the complexity of syndrome-associated diseases, genetic counseling is important in the interpretation of the results. The result may have implications for the future health of the baby and/or the health of other family members.

THE INSERTION/DELETION POLYMORPHISM IN THE ACE GENE AND CHRONIC OBSTRUCTIVE PULMONARY DISEASE IN THE POPULATION OF MOLDOVA

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The relevance of the study. Chronic obstructive pulmonary disease (COPD) is a chronic inflammatory disease characterized by the gradual progression of irreversible airflow obstruction and increased inflammation in the airways and lung parenchyma. It is a major and an increasingly prevalent health problem worldwide. Genetic factors are believed to play a role in the development of COPD. The ACE gene encodes the amino acid sequence of angiotensin-converting enzyme (ACE) that regulates arterial pressure, as well as salt and water metabolism. In addition, it mediates biosynthesis of growth factors. Intron 16 of the ACE gene harbors an insertion-deletion (ID) polymorphism. This polymorphism is known to affect the plasma ACE levels and might, thus, modulate susceptibility to COPD and COPD-associated pulmonary hypertension.

The purpose of this study was to investigate the association of the I/D polymorphism of the ACE gene with COPD in the population of Moldova.

Materials and research methods. 87 patients with COPD (84 male and 3 female; 63.8±8.0 years old) and 90 volunteer participants (controls) from general population (87 male and 3 female; 51.8±8.1 years old) were placed in this study. COPD diagnostics was performed according to the criteria GOLD. DNA was extracted from blood samples and genotyping of ACE I/D was performed using polymerase chain reaction. Genotypic model-based associations of the I/D polymorphism with COPD were tested using multivariate logistic regression analysis adjusted for sex and age.

Results. The frequencies of ACE genotypes were found to be 29,9% for DD, 43,7% for ID, and 26,4% for II in the COPD group and 18,9% for DD, 56,7% for ID, and 24,4% for II in the control group. The allele frequencies were found to be 51,7% for the D allele and 48,3% for the I allele in the COPD group and 47,2% for the D allele and 52,8% for the I allele in the control group. These frequencies are similar with those from other European populations. All genotype distributions in cases and controls did not differ significantly ($p>0,05$) from those expected by the Hardy–Weinberg equilibrium.

The results from the association tests demonstrated absence of any significant association of the I/D polymorphism with COPD across different genetic models of inheritance ($p>0,05$), although a tendency of inverse association was found under the recessive model (II *versus* ID + DD; OR: 0.46; 95% CI: 0.19-1.07; $p=0,069$).

Conclusion. This pilot study suggests that ACE polymorphism does not play a major role in genetic susceptibility to COPD in the population of the Republic of Moldova. Further studies with larger numbers of patients considering interaction of ACE with other genetic and environment (i.e. smoking, living conditions) factors are needed to explore this relationship.

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MONITORING OF CONGENITAL ANOMALIES IN CEADIR-LUNGA REGION OF THE REPUBLIC OF MOLDOVA

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Background: Relevance of the study is reasoned by the higher prevalence of congenital anomalies (CA) in the South of Moldova, due to unfavorable ecological situation (i. e., high level of mineralization of drinking water, soil contamination by pesticides, etc.).

Aim of the study is to evaluate the prevalence and sharing of CA in Ceadir-Lunga region (Southern area of the Republic of Moldova) for the period from 2010 to 2014.

Materials and methods. Was examined the prevalence and sharing of the CA in the Republic of Moldova on the basis of genetic monitoring for the period from 2010 to 2014 years. We are used epidemiological approach for the study. Cytogenetic study was performed in all patients with multiple congenital anomalies (MCA).

Results. The prevalence of CA during the analyzed period was 19,33 per 1000 newborns and exceeded the frequency of CA in Moldova in overall, i. e., 17,55 per 1000 newborns. The maximal prevalence of CA was noted in 2011 – 24,25 per 1000 newborns, the minimal – in 2010 – 12,91 per 1000 newborns. In sharing of CA the leading place is occupied by the anomalies of the muscular skeletal system (37%), in overall in the Republic of Moldova this value was lower – 19,5%. MCA were registered in 11% of CA in overall, the share of CA of that type in Moldova in overall was 25,2%. CA of the cardio-vascular system amounted to 9%, in Moldova in overall they accounted for 16% of the total number of CA. We are note the following changes in prevalence of CA: from 2010 to 2011, increasing from 12,91 per 1000 newborns to 24,25 per 1000 newborns, in overall in Moldova this vale decreased from 19,33 in 2010 to 17,69 per 1000 newborns in 2011. From 2011 to 2013 we are mentioned a steady trend to decreasing in the prevalence of CA from 24,25 per 1000 newborns to 16,33 per 1000 newborns as in the region, as well as in overall in the Republic. In 2014, the prevalence of CA increased again to 23,19 per 1000 newborns. The prevalence of Down syndrome in Ceadir-Lunga region was 1,93 per 1000 newborns, which is considerably higher compared to the mean population prevalence, i. e., 1,24 per 1000 births, the prevalence of spina bifida (0,77 per 1000 newborns) is also higher than the overall prevalence in Moldova (0,29 per 1000 newborns).

Conclusions. The data obtained from monitoring have shown that the prevalence of CA in Ceadir-Lunga region is significantly higher than the mean population prevalence, which indicates a lack of preventive measures to reduce the prevalence of CA.

IMPORTANCE OF IMMUNOHISTOCHEMICAL INVESTIGATION IN DIAGNOSIS OF COLON DYSPLASTIC MALFORMATIONS IN CHILDREN

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Colon dysplastic malformations in children of diverse age result in chronic colostasis and are caused by anatomic abnormalities of development, such as dolicosigma, anorectal atresia etc., but by the majority of disorders of intestinal innervation. It is undeniable the affection of the nervous system manifested through agenesis of myenteric nervous ganglia in Hirshsprung disease established in 1886 in neonates with severe constipation and megacolon. But there are many clinical cases exhibiting chronic colostasis and megacolon in the presence of intestinal nervous plexus. However, the diversity of dysplastic malformations certified by classical and histochemical methods requires choosing new investigative tactics to objectify differentiation of lesion features in postoperative colonic pieces.

To elucidate specific features of immunohistochemical marking expression (IHCh) relevant to differential diagnosis of dysplastic colon malformations in children.

We established various deviations of monoclonal and polyclonal antibodies expression compared to expression of antibodies in visually healthy sectors. An important aspect detected by immunohistochemical researches is revealing a different expression and various combinations of expression of monoclonal antibodies in the same intestinal fragment. This leads to the assumption that the features detected are based on protein disorders, participating in nervous impulse transmission to the peripheral system both intraganglionic and intracellular ones, including at synapses level in neuromuscular junctions. Another feature detected by immunohistochemical research is revealing a different expression of primary antibodies in the same intramural nervous ganglion in consecutive histological sections.

The statistical analysis of the intensity of the monoclonal antibodies expression established that in rectal biopsies the positive reaction was distributed as follows: NSE $2,13 \pm 0,18$; NFP $1,9 \pm 0,18$; CGA $1,4 \pm 0,17$; SYP $1,63 \pm 0,23$ ($p < 0,05$). Immunomarking at visually healthy level determined an increase in the intensity of antibody expression: NSE $2,5 \pm 0,14$; NFP $2,4 \pm 0,14$; CGA $1,7 \pm 0,2$; SYP $2,0 \pm 0,2$. It was also revealed the low level of Synaptophysin antibody (SYP) ($1,5 \pm 0,1$) ($p < 0,05$), certified in myenteric neural ganglia and/or neuromuscular junctions. There are also strong associative links between disorders of expression of monoclonal antibodies at investigated colonic levels, established through high dependencies and direct in correlational analysis of the expression of NSE and NFP antibodies ($r = 0,72$), NSE and CGA ($r = 0,53$) NSE and SYP ($r = 0,53$), NFP and CGA ($r = 0,51$), CGA and SYP ($r = 0,60$).

Conclusion. Peculiarities of expression of monoclonal antibodies assessed by IHCh method in colonic biopsies revealed that in differential diagnosis of dysplastic colon malformations in children of diverse age, especially in aganglionosis objectification, Neuron Specific Enolase monoclonal antibody $2,13 \pm 0,18$ ($p < 0,05$) is the most relevant. It has been established presence of close associative links between disorders of expression of different monoclonal antibodies, which argues the fact that deregulation of some neurotropic enzymes cause disruption to others, and late detection of these patients influences the amount and quality of surgery.

THE ROLE OF CYTOGENETIC PRENATAL DIAGNOSIS IN PREVENTION OF CHROMOSOMAL ABNORMALITIES

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Introduction: The diagnosis of foetal chromosomal abnormalities is carried out at the population level by cytogenetic prenatal diagnostic technologies (CPD). The CPD solve a significant amount of pressing problems and critical issues related to the prophylaxis of genetic pathologies and to the prevention of birth of children with numerical and structural chromosomal abnormalities. Therefore, CPD contribute to the detection of genetic pathologies and especially of chromosomal abnormalities in foetuses before their birth. The core and value of cytogenetic prenatal diagnosis is determined first of all by the information on foetal genotype and by prevention and avoidance of childbirth with genetic diseases. Among the CPD methods most commonly is indicated the amniocentesis, including the study of foetal karyotype from 16 to 18 w.g. The CPD methods are considered widely applied and safe tests, during which the geneticist, specialized in genetic counselling, informs the proband correctly, completely and understandably on the role, benefits, risk level, indications and contraindications of these investigations.

The scope of the work is to highlight the role of prenatal cytogenetic diagnosis for identification of chromosomal abnormalities in foetuses at early stages of intrauterine development and decrease the incidence of chromosomal abnormalities in newborns.

Materials and methods: The process of investigation included the prospective medical – genetic counselling with the purpose of identification of the target group consisting in 12 938 pregnant women of the risk-group, being prescribed to be examined in CRHMG from the Institute of Mother and Child, in the period 2005-2014. 4731 ($36.6 \pm 0.4\%$) of total amount of women had cytogenetic prenatal diagnosis (CPD): amniocentesis and chorionic villus biopsy. Patients were divided into two clinical batches: a) group I: 4731 ($36.6 \pm 0.4\%$, $p < 0.001$) – pregnant women from medium and high risk group; b) group II: 8207 ($63.4 \pm 0.4\%$, $p < 0.001$) – pregnant women from low risk group.

Results: Those two groups were comparable in age, gestational period, degree of genetic risk. The age of women in high and medium genetic risk group was between 17 to 44 years old (average age 26.1 ± 5.3 years). The pregnancy term, when addressing to the geneticist, was between 6 to 22 w.g. (mean 14 ± 5.2 w.g.).

In 4731 pregnant women who performed cytogenetic prenatal diagnosis, most frequent chromosomal abnormality were aneuploidies, including autosomal trisomies, the most common: Down syndrome accounted for 75 cases ($0.18 \pm 1.6\%$), Patau syndrome - 9 cases ($0.06 \pm 0.2\%$) and Edwards syndrome - 20 cases ($0.09 \pm 0.4\%$). Among the diagnosed gonosomale abnormalities, monosomy X was found in 8 cases ($0.05 \pm 0.16\%$) and Klinefelter syndrome - 10 cases ($0.07 \pm 0.21\%$). During 2008 - 2012 five fetuses ($0.05 \pm 0.1\%$) were diagnosed prenatally with Triple X syndrome, 6 fetuses ($0.05 \pm 0.13\%$) with triploidy and 31 fetuses ($0.7 \pm 0.12\%$) with other structural chromosomal syndromes.

The research and evaluation of results of cytogenetic prenatal diagnosis – karyotyping, allowed prenatal diagnosis of chromosomal abnormalities in 164 cases, which was $0.3 \pm 3.5\%$ of total amount of pregnant women who carried out DPC in concerned period.

In the situations when numerical or structural chromosomal abnormality is diagnosed with a prognosis poor for life or incompatible with life, a therapeutic abortion may be an option, which is legally justified but controversial ethically and morally. During the medical-genetic counseling these aspects were analyzed from all points of view, taking into account the vital prognosis and quality of life. A medical-genetic advice was provided for patients. The decision to keep or not the pregnancy, depended on the couple, on parents and/or the mother.

Conclusions:

The methods of cytogenetic prenatal diagnosis (fetal karyotyping) and medical-genetic counseling contribute to reducing the frequency of chromosomal abnormalities in newborns.

Due to CPD methods was possible to prevent the birth in 164 ($0.3 \pm 3.5\%$) cases with chromosomal abnormalities, prenatally diagnosed until 21 weeks of gestation.

In the structure of fetal chromosomal abnormalities, diagnosed in fetus until 22 weeks of gestation, the most common is Down syndrome - 75 cases ($0.18 \pm 1.6\%$), Edwards syndrome - 20 cases ($0.09 \pm 0.4\%$) and structural chromosomal abnormalities - 31 cases ($0.12 \pm 0.7\%$).

GENETIC RENAL PATHOLOGICAL ISSUES IN CHILDREN

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Introduction: Renal diseases are a group of heterogeneous pathological failures determined genetically. The aetiology of many kidney diseases has been revealed as single-gene defects. Knowledge of the primary cause of a disease is essential for understanding its mechanisms and for adequate classification, prognosis, and treatment.

Purpose: To identify children with genetic renal anomalies and to evaluate all possible mutations triggering the production of diseases.

Materials and methods: The study is based on 40 children (12 girls (30%) and 28 boys (70%)), aged between 1 and 15 years old. They were clinical examined (physical examination), paraclinical investigated (determining renal excretion functioning – plasma urea, serum creatinine, urine testing, examining and testing of proteinuria and hematuria; biochemical investigations – potassium, calcium, sodium, phosphates; immunological investigations; imagistic investigations – intravenous urography, simple renal radiography, renal echography, renal scintigraphy, MRI, CT) and genetics (family anamnesis).

Results: A total of 40 patients were examined, out of which 15 patients (37.5%) were with positive family anamnesis and 25 patients (62.5%) were with negative family anamnesis 32 (80%) presented suggestive signs for a certain renal pathology, 8 (20%) were with uncertain clinical manifestations and 20 (50%) presented infections.

Within children with positive family anamnesis: 1 case with ADPKD, determined by gene mutation PKD1 (16p13.3), 1 case – Sdr. Barrter case type I (mutations in the SLC12A1 gene), 1 case with renal agenesis, 1 case with renal medullary spongiosis (Sdr. Cacchi–Ricci as a result of GDNF gene mutation), 15 cases –vesicoureteral reflux, 6 cases with familial glomerulonephritis and 6 cases with other renal diseases.

Conclusion: Renal genetic diseases are chronic and constantly progressive but a genetically new approach allows determining the correct symptoms that our body suffers from, therefore, a correct approach will help avoid future complications in this area.

**GENOMIC TECHNOLOGIES – INVESTIGATIVE TOOLS IN
NEURODEVELOPMENTAL DISORDERS**

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Neurodevelopmental disorders, including - but not limited to - intellectual disabilities and autism spectrum disorders, are life-long impairing conditions of the central nervous system growth and development, with severe impact on the quality of life of both the patient and patient's family; ultimately, considering the developmental disorder patient's health care, as well as corresponding social integration costs, these diseases impose a significant burden to the society.

Despite the decades-long research focusing on their causes, with constant and frequent additions to the list of genetic entities contributing to neurodevelopmental pathologic phenotypes, the identification of the causative factors remains a major challenge in the clinical practice; additionally, genetic and environmental factors may contribute in various degrees on a person's development and behavior. The identification of the genetic abnormality causative for patient's phenotype is instrumental for diagnosis, prognostic and for the genetic counseling of the patient's family. Currently, the clinical practice guidelines and recommendations suggest that early diagnosis is an advantage in the clinical management of the patient.

Here, we describe the impact and advantages of genomic technologies, particularly of array-based comparative genomic hybridization (array-CGH) in the investigation of developmental disorders and associated congenital malformations. Thus, genomic DNA samples of pediatric patients referred to our laboratory for array-CGH and, respectively, from commercially-available reference sources, were labeled with distinct fluorochromes and processed on Agilent Technologies platforms, according to the manufacturer's recommendations, aiming to identify submicroscopic defects potentially responsible for patient's phenotype.

Genetic defects causative for syndromes such as Mowat-Wilson, 15q26-qter deletion, 18q deletion, DiGeorge, MECP2 duplication etc, as well as other aberrations associated with idiopathic intellectual disability were identified and described. The results were validated through independent techniques, as recommended by the guidelines. Phenotype-genotype correlations were made.

In conclusion, genomic diagnostic technology is extremely useful for both clinicians and researchers in many fields focusing on pediatric disorders, as a first-tier diagnostic tool, ultimately leading to an increase in the efficiency of patient's management and quality of life.

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DESPRE FORMELE FAMILIALE ALE CANCERULUI TIROIDIAN DIFERENȚIAT

A. ȚÎBÎRNĂ

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Încă în anul 100 un medic roman a menționat ca o curiozitate frecvența înaltă a cancerului mamar în familia unei paciente.

Este cunoscută în istoria medicinei familia lui Napoleon, 11 membri ai căreia au suferit de cancer gastric. Astfel, s-a întărit rolul factorului genetic în etiologia cancerului.

Cancerului este un termen genetic, care definește o gamă largă de boli, ce se caracterizează prin alterarea proceselor de creștere și proliferare celulară.

Trăsătura de bază, care permite recunoașterea clinică a existenței unei predispoziții genetice este istoricul familial pozitiv. Predispoziția genetică poate îmbrăca un spectru larg, de la existența unor forme rare de cancer ereditare până la cancer cu predispoziție genetică, dar fără istoric familial.

Cancere familiale sunt o formă de predispoziție genetică, asociată cu un risc redus de dezvoltare a unor cancer și este întâlnită, în special, în formele comune de neoplazii. Spre deosebire de formele ereditare singura trăsătură, care poate fi recunoscută este agregarea familială, ceea ce impune o anamneză familială corectă.

Cauzele cancerelor familiale pot fi variante alelice particulare ale aceluiași gene implicate și în dezvoltarea cancerelor ereditare ori a altor gene, cercetarea cărora este într-un stadiu incipient.

Este necesară o catalogare a diverselor variante ale genelor, întâlnite în rândurile populației, în asociere cu identificarea corelațiilor existente între variantele specifice și riscul pentru anumite forme de cancer. Variantele cele mai studiate în prezent sunt polimorfismele mononucleotidice – SNPs (Single Nucleotide Polymorphism), care alcătuiesc câteva milioane în întreg genomul uman.

Prezentăm în continuare câteva forme de cancer tiroidian diferențiat, care se întâlnesc în unele sindroame ereditare.

Boala Cowden – se manifestă prin cancer/adenom al glandei mamare și tiroide, hamartom intestinal, mutații în gena PTEN/10q 22-23. În glanda tiroidă se dezvoltă tumori multiple benigne sau maligne, se afectează ambii lobi.

Sindromul Peutz-Jeghers – se manifestă prin cancer intestinal în asociere cu cancer ovariene/testiculare și cancer tiroidian diferențiat, mutații în gena STK – 11

Sindromul MEN 1 – adenome tiroidiene, hipofizare, pancreatice în asociere cu cancer papilar tiroidian în 10% cazuri, mutații în Menin/11q13.

Sindromul Gardner – polipoze ale intestinului gros și subțire în asociere cu lipoame, fibroame, osteome și cancer papilar tiroidiene în 1-2% cazuri mutații în gena APC/5q21.

Sindromul Werner – se manifestă prin hipogonadism, diabet zaharat, cataractă juvenilă, cancer tiroidian folicular sau papilar, mai rar anaplasice.

În prezent testarea genetică pentru identificarea persoanelor cu risc crescut din cadrul familiilor cu istoric familial pozitiv este parte a menegmentului standart pentru unele din aceste sindroame. Un interes deosebit îl prezintă genele modificate, care reduc penetranța genelor cauzatoare, atât pentru aprecierea corectă a riscului, cât și pentru elaborarea unor metode noi de prevenire și tratament.

THE STRATEGY OF DEVELOPMENT OF INBORN ERRORS OF METABOLISM IN MOLDOVA

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The inborn errors of metabolism (IEM) area is a very important domain determining the infant and child morbidity and mortality. There are more than 6000 recognized IEM. IEM are usually rare diseases which frequency is less than 1:2000 people. 75% of rare diseases affect children and 35% of rare disease patients will die before their 1st birthday and another 30% will die before their 5th birthday. Rare diseases are rare, but rare disease patients are numerous.

The IEM require very expensive equipment and a very good trained multidisciplinary team of professionals to be developed. In Moldova this domain is not evolved according to many standards due to very limited financial possibilities, but the diagnosis of IEM, beside Phenylketonuria (PKU) where there is the neonatal screening, is successfully initiated through a very fruitful collaboration with the foreign partners, supporting in such a way the creation of the National Register of Rare Diseases in Moldova.

Thus, the basic biochemistry as the ammonia, lactate, glucose, pH and amino acids of body fluids by liquid chromatography are performed in Moldova; the ¹H-NMR spectroscopy for organic acids of urine/CSF/amniotic fluid in Romania; Tandem MS with acylcarnitine profile - in Hungary and Romania; neurotransmitters in CSF – in Germany and IEF of Transferrin in USA.

Due to existing for about 25 years of neonatal screening for PKU in Moldova our National Register includes 102 PKU patients; the screening rate is 88-97% during last years, each year about 3-4 PKU newborns are found and PKU frequency is 1:7772 newborns. The genotype analysis is routinely performed in PKU patients only on 12 common mutations found in ~75% of them, a common work in sequencing of PAH gene in molecularly undefined PKU patients is initiated with Switzerland; the BH₄ loading test is not applied yet. We report on 3 cases of maternal PKU. The restarting of neonatal screening for Congenital Hypothyroidism is in progressing now.

Based on our collaboration, we created a regional network of diagnosis and could report on: Methylmalonic Aciduria - 5 cases, Glutaric Aciduria type I-1 case, Alkaptonuria-1 case, Galactosemia- 4 cases, Glycogen Storage Disorder - 4 children, suspected for Mitochondrial disorders – more than 10 cases, possible PDHC/KDHC deficiency -2 cases, GM1-Gangliosidosis-2 cases, Urea Cycle Disorder -2 cases, Hyperekplexia- 2 cases, about 10 cases suspected for Neurotransmitters disorders and one child for Congenital Disorder of Glycosylation.

In the absence of a system for IEM performing in Moldova the collaboration with other institutions is very important. The evaluation of clinical manifestations remains the most important to suspect an IEM patient. Among most accessible specific investigations for Moldova the ¹H-NMR spectroscopy of urine performed in Romania (the nearest Lab) seems to provide high analytic information improving the orientation in diagnosis of IEM in unclear patients, but all methods are very necessary and should be developed for the better IEM diagnosis. A new amino acids analyzer and other equipment (genetic analyzer, qPCR) will be used for diagnosis in the following months. Early diagnosis of IEM is very important for the specific therapy initiation and to prevent some of them by prenatal tests. Our further developmental strategy is to improve the field of IEM in Moldova through the common European projects.

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CHEMOKINE GENES POLYMORPHISMS AND PULMONARY TUBERCULOSIS IN THE POPULATION OF MOLDOVA

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Background. With approximately 9 million new cases and more than 1 million deaths tuberculosis (TB), a disease caused by *Mycobacterium tuberculosis* (*Mtb*), remains a global health problem. Among the exposed population, only 5–10% develop clinical disease while the remaining mounts a protective immune response. Host genetic polymorphisms have been suggested to explain this differential susceptibility to TB.

A primary pathologic feature of *Mtb* infection is the formation of a granuloma - collections of immune cells protecting against dissemination of mycobacteria. Chemokines govern cell (leukocytes) influx to *Mtb*-infected lungs and are critical for granuloma formation, and the control of infection. Polymorphisms in chemokine genes can alter the expression of these genes in the inflammatory cells, which, in turn, can affect the clinical phenotype of the disease.

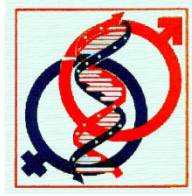
The aim of this study was to analyze polymorphisms in three chemokine genes (IL-8, MCP-1 and RANTES), exploring their associations with pulmonary tuberculosis in a Moldavian population.

Materials and Methods. 135 unrelated patients with bacteriologically confirmed pulmonary tuberculosis and 137 unrelated healthy individuals were placed in this study. Both groups came from the same communities living under the same conditions. DNA was extracted from blood samples and genotyping of IL-8 -251A/T (rs4073), MCP-1 -2518 A/G (rs1024611) and RANTES-403G/A (rs2107538) polymorphisms was performed using polymerase chain reaction-RFLP method. Multiple logistic regression models (co-dominant, dominant, recessive, and additive) adjusted for sex were used to test associations.

Results. The genotype and allele frequencies of the polymorphic loci tested in Moldavian population were found to be similar to those reported in other European populations. We found that all of the polymorphisms typed were in Hardy-Weinberg equilibrium in both control and TB subjects. No significant differences in the frequencies of the three polymorphic variants of the IL-8, MCP-1 and RANTES genes were observed between the healthy controls and TB cases across different inheritance models ($p > 0.05$).

Conclusion. These results suggest that IL-8 -251A/T (rs4073), MCP-1 -2518 A/G (rs1024611) and RANTES -403G/A (rs2107538) polymorphisms are not related with the risk of TB in the population of Moldova. However this does not exclude the possibility that other SNPs in these as well as other chemokine genes could be associated with the disease. Therefore, the role of chemokine gene variation in TB susceptibility should be further evaluated.

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PLANT GENETICS AND BREEDING

**The Xth International Congress
of Geneticists and Breeders**

CULTURILE COMPARATIVE DE PROVENIENȚĂ ECOLOGICĂ DIFERITĂ - BAZA AMELIORĂRII GENETICE A PĂDURILOR DE STEJAR PEDUNCULAT

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Prin ameliorarea genetică a speciilor de arbori forestieri se urmărește obținerea unor descendenți mult mai performanți din punct de vedere al productivității și calităților tehnologice, al capacității de adaptare la mediu în sensul creșterii rezistenței la acțiunile factorilor biotici și abiotici. De asemenea diversitatea genetică are o deosebită importanță în supraviețuirea speciilor de plante, deoarece capacitatea adaptivă a populațiilor față de condițiile mediului depinde de gradul de variație genetică. În cadrul populațiilor de arbori ale unei păduri naturale variația genetică se datorează proceselor care au loc sub influența condițiilor de mediu, rezultând pentru fiecare specie o anumită particularitate de manifestare a variabilității de-a lungul gradientului ecologic.

În ultimele decenii, cercetările și activitățile specifice domeniului ameliorării forestiere s-au perfecționat și extins extrem de mult, astfel încât în tot mai multe țări există și se derulează programe foarte complexe pe linia ameliorării forestiere. În acest mod sunt atinse tematici vaste ce pornesc de la studii proveniențelor și selecția de populații valoroase și se încheie cu abordarea celor mai noi și performante tehnici moderne specifice biologiei și geneticii moleculare.

La speciile de arbori forestieri ce însușesc calități economice și ecologice deosebite cum este și stejarul pedunculat cele mai sigure garanții privind eficacitatea unui program de ameliorare forestieră bazat pe selecție sunt oferite de alegerea și utilizarea celor mai bune proveniențe. În ciuda longevității mari a arborilor de stejar comparativ cu alte specii, selecția și efectele ei, în limita posibilităților, trebuie urmărită pe parcursul mai multor generații. Culturile comparative de proveniențe, în acest caz, au un rol extrem de important și determinant în ceea ce privește depistarea mai rapidă a efectelor selecției. Pe de altă parte, utilizarea micro-propagării în culturile in-vitro poate scurta semnificativ perioadele lungi de timp ce se scurg între ciclurile succesive de selecție.

Identificarea precisă și evaluarea eficientă a proveniențelor și a variabilității genetice a arborilor de stejar pedunculat s-a realizat în urma studiului asupra culturilor de diferită proveniență instalate în Rezervația „Plaiul Fagului”, în acest fel putându-se delimita cu precizie diverse populații și caracteriza proveniențele aparținând diverselor zone ecologice din țara noastră.

Crearea și studierea culturilor comparative de proveniențe contribuie la alegerea rațională a provenienței celei mai bine adaptate și mai stabile pentru zona în care se urmărește instalarea plantației speciale. Alegerea proveniențelor reprezintă prima etapă obligatorie a oricărui program de selecție, deoarece pe lângă estimarea capacității de adaptare, în alegerea proveniențelor se mai ține cont de vigoarea de creștere, dar și de calitatea lemnului, de asemenea, se ia în considerare și plasticitatea, opțional pe proveniențele mai puțin interactive și care sunt mai ușor adaptabile la situri experimentale diverse.

Excluderea acestui fapt face ca ameliorarea să pomească numai de la proveniențe locale, caz în care ameliorarea este mult mai lentă și mai costisitoare întrucât proveniențele locale nu sunt întotdeauna cele mai productive și nici chiar cele mai adaptate (Nanson, 1980; Kremer, 1986). Superioritatea genetică nu este un criteriu absolut pentru adaptabilitatea și productivitatea unei proveniențe, ceea ce înseamnă că aceeași proveniență poate da rezultate diferite de la un sit la altul, în funcție de mediu (datorită interacțiunii genotip x mediu). Acest lucru ne impune ca în viitor să creăm culturi experimentale în fiecare regiune de introducere și în special, în acele situri unde riscul unor interacțiuni climatice și pedologice este maxim.

Cel mai important criteriu care desemnează o proveniență ca fiind aptă de introducere într-o regiune diferită de cea de origine, este criteriul adaptării. Toate proveniențele care se dovedesc imperfect

adaptate, indiferent de alte performanțe ale lor, vor fi îndepărtate și nerecomandate de a fi folosite în programele de ameliorare.

În cadrul studiului asupra culturilor de diferită proveniență se atrage atenția la diferite caractere calitative și cantitative ale arborilor vizați, referindu-ne în special la calcularea unor parametri genetici importanți, în principal a coeficientului de eritabilitate. Eritabilitatea reprezintă capacitatea de transmitere a caracterelor de la genitori la descendenți (Stănescu, 1984) și se calculează statistic pe baza variației genotipice și fenotipice pornind de la diferite măsurători de teren.

În concluzie putem menționa că materialul de reproducere a speciilor de arbori și a hibrizilor artificiali, care sunt importanți pentru silvicultură, trebuie să fie adecvat din punct de vedere genetic diverselor condiții locale și să fie de calitate superioară; conservarea și sporirea biodiversității pădurilor, inclusiv diversitatea genetică a arborilor, sunt esențiale pentru o gestionare durabilă a pădurilor.

EXPRESSION OF SOME GENES IN BARLEY UNDER VIRAL INFECTION

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Viruses are the most damaging of plant pathogens, inducing a variety of responses in their hosts. It is known that virus infections dramatically affect plant physiology, decreasing photosynthesis, increasing respiration and altering carbohydrate levels. The contribution of viral agents to the enhancement of somatic and meiotic recombination, resulting in rearrangements that could, potentially, be transmitted to the next generation, has been recognized. According to the previous data, it was established that viral infection can contribute to chromosome breaks, modification of chromatin condensation, cell proliferation. The goal of this study is to evaluate the expression of some genes encoding antioxidant metabolism and stress related proteins in barley plants derived from susceptible cultivars to barley stripe mosaic virus (BSMV).

The experiments were performed on spring barley (*Hordeum vulgare* L.) cultivars Galactic, Sonor and Unirea. Seeds were obtained from healthy (mock-inoculated that served as control variant) plants and infected mechanically with BSMV (virus variant). Plants were sown in pots filled with a mixture of soil, peat and sand with six plants each (three pots for each experimental variant) and grown in a controlled conditions (Sanyo chamber). Samples (0.3–0.5 g from the middle part of the youngest, but fully developed leaf) at the 3th leaf stage were collected, frozen in liquid nitrogen and stored in -80°C. Five genes of interest were selected for study: superoxide dismutase (Sod), ascorbate peroxidase (Apx) and Pathogenesis-related (PR) proteins: PR3, PR5, PR10. Primers were designed in Primer Express Software v. 2.0 (Applied Biosystems, Foster City, CA, USA). Total RNA was isolated using *TRI Reagent* (AM9738, Applied Biosystems) according to the manufacturer's instruction. The RNA quantity and quality was assessed spectrophotometric (λ 260/280 nm) and by electrophoresis in MOPS-formaldehyde 1.4% agarose gel. PCR was performed using thermocycler DT-96 (DNA Technology, Russia) with the following cycling parameters: 10 min at 95°C, 5 cycles of 10s at 95°C, 20s at 64°C, 40 cycles of 15s at 95°C, 40s at 60°C. The specificity of PCR products was verified by dissociation curve analysis. The determination of Ct value and of the relative expression level of the studied genes (related to the target gene - α -tubulin) was achieved through the program Real Time PCR v7.3. The experience was carried out in triplicate independent cDNA synthesis (for each sample of RNA) and in double repetition qPCR (n = 6).

The expression of genes studied in the present experiment was clearly affected by viral infection. In barley progenies obtained from virus infected plants the activity of Apx and Sod was in decline in most variants. A no statistically deviation was established only for the expression of Apx in cv. Sonor. In the group of antioxidant metabolism genes, the expression of SOD mainly decreased (from 18% for cv. Galactic to 32% for cv. Sonor). The activity of both genes was higher in treatment with application of viral infection in cv. Unirea compared to control.

Genes related to pathogenesis proteins (PR) were significantly differentially expressed in evaluated barley cultivars ($P < 0.05$). The results of the present study showed a general up- or down-regulation of PR3 and PR10 in dependence of cultivars. The largest decrease of the PR3 activity was established for treatment variants of cv. Galactic and cv. Sonor (93% for cv. Sonor and % 76% for cv. Galactic, respectively less than in the control). A similar tendency was found and for PR10 activity.

The up-regulation of genes related to pathogenesis was reported for PR5 for all treatment variants. The highest increase (by 4.6 times) was detected for cv. Galactic. According previous cytogenetically studies this genotype present more plasticity. It is known, that the PR5 is related to stress mediated by jasmonate or ethylene signaling pathway. Their activity in young plant in normal condition is less, but in case of pathogenesis significantly increases. Also, PR10 exhibit ribonuclease, and PR3 - chitinase activity, which are more specific.

Evident modifications in genes transcriptional activity between control and virus variants included (1) suppression of genes involved in antioxidant metabolism; (2) activation or suppression of genes for jasmonate synthesis or chitinase activity in dependence of genotype specificity; (3) up-regulation of genes for stress-related transcription factors such as PR10.

**GENETIC DIVERSITY OF SOURCES FOR MALE STERILITY AND RESTORATION
OF FERTILITY IN SUNFLOWER COLLECTION**

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More than 130 sunflower inbred lines which differed by the origin and ability for the suppressing CMS phenotype were studied with the use of classical hybridological analysis, molecular sequencing, DNA and protein markers. The majority (73%) of restorer lines from VIR collection has been shown to possess sterile cytoplasm of PET1 type. It was assumed that these genotypes can be considered as the carriers of functional alleles of the fertility restoration (*Rf*) genes for CMS PET1. A heteroplasmy (presence of two or three haplotypes at the *orfH522* locus associated with PET1) was found in 7 fertile lines and the source of CMS RIG0. Nucleotide sequences of the mitochondrial genes *orfH522* and *atp9* were identical in different genotypes that indicated a high stability of CMS PET1.

Seven PCR-markers (STS, SCAR, SSR) linked to the *Rf1* locus clearly differentiated male sterile and restorer lines. The STS115, HRG01 and HRG02 markers have demonstrated the highest diagnostic value for the identification of the *Rf1* gene in a group of genetically diverse genotypes compared to the SSR markers ORS224, ORS511 and ORS799. A polymorphism at the examined SSR loci was firstly described. Five alleles were observed at the ORS511 locus, and four and two alleles were found at the ORS224 and ORS799 respectively. Lines with the unique alleles at the SSR loci also possessed unique alleles of the genes encoding helianthinin and 2S albumins seed storage proteins.

The majority of the known *Rf* genes encode proteins containing pentatricopeptide repeat motifs (PPR) and are classified as the members of the *RFL-PPR* (*Restorer-of-Fertility-Like-PPR*) family. To test the hypothesis on belonging the sunflower *Rf* genes to the *RFL-PPR* family we have studied the nucleotide polymorphism of 10 EST fragments homologous to the *Rf* genes of other plant species. All the fragments contained PPR motifs. Seven fragments were polymorphic, and the three others were nearly identical among the CMS and restorer lines. The highest frequency of nonsynonymous SNPs (>4%) was mentioned in the fragments QHL12D20 (1174 bp) and QHB20M13 (421 bp). Two fragments contained introns with the length of 80 bp and 635-628 bp. The intron of QHL12D20 was highly homologous to the intron of the *AHBP_1B* gene; the product of this gene has a similarity with the transcription factor of the bZIP family of *Arabidopsis*. Two allelic variants of the fragments QHL12D20 and B20M13 were detected. CAPS markers have been developed and the lines were differentiated depending on the presence of different alleles of the *RFL-PPR* genes homologs. All the CMS lines possessed identical variants of the CAPS markers whereas in a group of restorer lines the alternative variants were more common. A co-segregation of the developed CAPS markers with the *Rf1* common molecular markers in the F2 populations of interline crosses is currently under examination. Due to their relatively high polymorphism the polymorphic variants of the *RFL-PPR* genes homologs can serve as a source of markers for evolutionary and diversity studies in sunflower.

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EVALUATION AND SELECTION OF TOMATO FORMS RESISTANT TO GYDRIC INSUFFICIENCY

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A special role among strong negative factors affecting crop plants is drought. Drought conditioned by separate action of temperature or separate influence of temperature or associated influence with impairment of water and high temperature is the main limiting factor for agriculture. Avoiding and reducing the negative effects of drought requires a deep knowledge of resistance mechanisms and their genetic control at different stages of plant organization. It was found that the tomato pollen grains ability to maintain dimensional stability under conditions of water stress can be successfully used for testing and forecasting resistance genotypes at different stages of plant development. The above were taken as a basis for research regarding the evaluation of tomato genotypes resistant to water stress by emphasizing forms and precious characters. We have analyzed 14 genotypes of tomato pollen grains size variability in optimal conditions and of osmotic stress. At the stage of flowering was selected pollen of respective genotypes and was located on stress conditions artificial created that contains osmotic solution (sorbit) at three concentrations of 8, 50, and 85%. As control served dry pollen. At the microscope in the room specially designed were determined dimensions of pollen grains in units of microns. After evaluating the reaction of genotypes of tomato pollen on all osmotic conditions it was observed the size reduction of pollen grains, because they decrease due to water loss. In base of report sample-control it was established the capacity of moisture retention and water absorption power by studied tomato genotypes. On the basis of the results obtained was calculated perimeter of pollen grain. This indication varies depending on the osmotic pressure of pollen grain. Bifactorial analysis of variance determined that on the pollen grain perimeter exert considerable influence the sorbitol solution with a value of 76%, but the contribution of genotype is four times smaller - 19%. The results showed significant differences in absorption capacity and by this criterion genotypes were divided and distributed in groups of resistance. Thus was established that increasing concentrations of sorbitol-containing solutions leads to resize the pollen grains, they are to be reduced in all studied genotypes. From the analyzed genotypes were highlighted Delta, Victorina Rio-Grande and Apelsin varieties by moving the minimum pollen dimension, which is characterized by being resistant to water stress. For example at Delta variety grain of pollen area shrinks significantly by 14% and also at Moscovschie zvezdi, Apelsin, Rio-Grand, Hurma and other genotypes in dependence on the concentration of sorbit pollen size decreases from 26,5% to 13,5%. Character analysis of the absorption capacity at different tomato genotypes enabled their differentiation by water stress resistance. It is already known, the higher the value of this indicator is, and the more resistance is high. It was established that the osmotic pressure of 104, 6 atmospheres (85% of sorbit solution) allow authentic differentiation of genotypes by drought resistance (absorption capacity and water retention). Rio-Grande genotype showed the highest absorption capacity of 85%, follows L.cheesmanii – 68, 5%, and those genotypes were placed in the first group of resistance. In the second group with average capacity of absorption and retention of water were distributed five genotypes and the other with the lowest absorbency and water retention were placed in the third group.

Therefore it can be confirmed that the action of osmotic stress conditioned by sipped, allowed the differentiation and selection of genotypes with valuable characters of hydric insufficiency resistance. GP rating scale variability at tomatoes in hydric stress conditions can serve as an effective method of highlighting the differences between genotypes.

QTL ANALYSIS OF BIOCHEMICAL AND IMMUNOLOGICAL TRAITS IN *BRASSICA RAPA* SUBSP. *RAPA* L.

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The genus *Brassica* has a long history of world wide cultivation and comprises large and diverse groups of important vegetable, oil, fodder and condiment crops. One of the subspecies of the genus is *Brassica rapa* subsp. *rapa* L., which is a very diverse subspecies with the longest cultivation history, and encompasses different kinds of turnips. *B. rapa* subsp. *rapa* morphotypes, including leafy and rooted turnips, vegetable and fodder types, differ based on which organs are consumed as feed and food. At the same time cruciferous plants (Brassicaceae family) are affected by different kinds of phytopathogens, and one of most harmful is bacteria *Xanthomonas campestris* pv. *campestris* Pam. (Dow.), which affects crucifers to bring on black rot. Strains of *X. campestris* pv. *campestris* are dividing into physiological races, which one can reveal by reaction of varieties with race specific genes of virulence, but existence of 9 races of pathogen complicate the interpretation of published results of plant resistance evaluation. So, the question on quality and resistance of *B. rapa* subsp. *rapa* plants is one of the main questions of modern genetics and breeding, including technology of plant genetic resources valorification. The modern method to investigate the genetic basis of variation within the germplasm of a species is genetic mapping based on segregating populations or quantitative trait loci (QTL) mapping. QTL mapping is based on the principle that genes and markers, which are saturated linkage groups of mapping population, segregate via chromosome recombination (called crossing-over) during meiosis (i.e. sexual reproduction), thus allowing their analysis in the progeny and detecting an association between phenotype and the genotype of molecular-genetic markers.

Using mapping population of double haploid (DH) lines of *Brassica rapa* subsp. *rapa* L. obtained by crossing of vegetable turnip and yellow sarson, the QTL analysis of five biochemical traits of quality (dry matter, total protein, sum of sugars, carotenoids, and β -carotene) was done. The DH population of *B. rapa* were evaluated on resistance to strains of three *X. campestris* pv. *campestris* races (1279a, B-32, PHW231). QTL analysis have been carried out by means of MAPQTL®6.0 which was used for identification and localization (candidate) QTLs on linkage groups (mapping interval 5 cM), LOD-score (Logarithm of Odds) ($P=0.05$), and degree of resistance trait variations for different races for each QTL, each resistance index. Significance of each QTL was verified by permutation test (1000 replication). It was also established effects of action of each QTL, percent of phenotype variation determined by each QTL, and molecular markers genetically linked with revealed QTL. For example, for biochemical traits it was revealed QTLs, which are determined β -carotene, total protein, as well as chlorophyll a and b shown relative low LOD, which it is depended on environment. The QTLs of investigated biochemical traits are located mainly in R03, R05, R07, and R09 linkage groups. Some QTLs were detected in R06.

Carried out QTL analysis for *X. campestris* pv. *campestris* resistance allowed obtained for DH mapping population five markers linked to resistance for three races of pathogen in linkage groups R03, R07, and R09. For DH mapping lines molecular marker KS50200, localized in R03, was linked with genetic locus of resistance to two *Xanthomonas* races (1279a, B-32), and marker SSR89, localized in the bottom of R07 – with loci of resistance to one race (B-32). Level of variability explained by revealed QTL was from 2,7% till 28,3%. Gene effects at all revealed loci were additive. In R05 and R10 it was found one marker for each linkage group respectively.

For the first time, for each investigated trait was determined QTL, as well as the effects of action of revealed QTL, percent of phenotype variability for each QTL and molecular markers genetically linked with identified QTL. Revealed molecular markers can be used as effective tool upon mass screening of collection and breeding material for biochemical traits of quality as well as for resistance for black rot. Moreover by identifying molecular markers linked with QTL determining each component of studied biochemical and resistance traits we have been able to 'dissect' the complex trait of a plant morphology and resistance and will be able to provide plant breeders with tools to breed these agronomically important traits using marker-assisted selection.

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ASSOCIATION MAPPING OF MORPHOLOGICAL AND BIOCHEMICAL TRAITS IN
BRASSICA RAPA L. CORE COLLECTION OF VIR

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Association mapping in plants uses linkage disequilibrium (LD) that is presented in natural and breeding populations including collections of plant genetic resources in genebanks. Using this tool, the genetic diversity expressed as allelic polymorphisms can be related to the observed phenotypic variation. Results in various crops indicate association mapping can be successful in the identification of markers linked to genes and/or genomic regions associated to the desirable traits. Several candidate loci and genome wide markers have been associated with agromorphological traits in potato, barley, cotton. *Brassica rapa* L. is the most diverse species in genus *Brassica* with the longest cultivation history, and encompasses leafy vegetables, turnips and oilseeds. VIR *B. rapa* core collection of 96 accessions bases on full collections representing different morphotypes of different geographical origin: 380 acc. – Chinese cabbage, 97 acc. – pakchoi, 19 acc. – black cabbage, 16 – mizuna and mibuna, 37 – komatsuna and neep greens, 13 – broccoletto, 425 acc. of vegetable and fodder turnip and 327 – annual and biannual oilseeds.

In present study we have used VIR core collection for association mapping of morphological and biochemical valuable characters. We analyzed phenotypic variation of growth related traits (plant and leaf sizes), leaf morphological traits connected with quality and biochemical traits and evaluated genetic diversity with a set of 135 SSR and 123 S-SAP (CACTA) markers.

Cluster analysis revealed accurate positions of accessions in system of species: *B. rapa* crops were divided into two major clusters: East-Asian vegetables and Indo-European-Asian oilseed and turnip. The first cluster was divided into subclusters of Chinese cabbage and pak-choi, with accurate division of Chinese cabbage into four groups, including a separate group of headed cabbages; separate positions of hiroshimana, shirona, mana and mizuna forms were determined. Second cluster was divided on oilseeds according to their geographical origin and separate group of turnips in Nepal-Indian subcluster. Only few accessions of European turnip were dispersed among accessions of the other cluster, they are supposed to be the oldest forms closely related to the first domesticated Central-Asian *B. rapa* form.

For searching of association of markers with traits standardization of the data was made, when molecular matrix was used with “1” for marker presence and “0” for marker absence, and phenotypic traits have been taken values between 0 and 1 and divided by the range. Genetic distance and similarity analysis were calculated by NTSYSpc, cluster analysis by Neighbor joining. The traits were integrated into the tree and molecular markers localized nearby them were desirable markers for these traits. Signification levels of associations were checked by parametric and nonparametric tests.

Upon searching association of markers with morphological as well as biochemical traits in natural heterozygous and heterogenic populations of *B. rapa* core collection accessions genetic variability expressed as allele polymorphisms of SSR and S-SAP markers bring into correlation with phenotype variation of traits. As results of investigations upon analysis of association of morphological traits weight and diameter of plants, leaf type and lamina width on the basis of VIR *B. rapa* collection 6 S-SAP markers were found, for traits of lamina length and colour – 3 SSR markers, and for traits of lamina hairiness, character of surface and lamina edge – 2 SSR and 3 S-SAP markers.

We have found two markers for each follow traits: leaf length, width, edge, colour, hair, surface, leaf type, plant diameter, plant weight; SSRs were located more often in A03 and A09. Two markers were determined for β -carotene and chlorophylls a+b. SSRs were located in A04. Positions of S-SAP markers are unknown. It is necessary to note that for traits of plant weight and leaf type, lamina length, width, surface, edge, hairiness, colour association of markers with contrast manifestation of the traits were revealed. For example, marker BOT1_3_E_ACA_270 was associated with locus determined weight of plants $2,172.91 \pm 146.52$ g, and marker BOT1_2_M_CAG_145 – weight $1,561.19 \pm 96.51$ g. It means that chromosome locus associated with marker BOT1_3_E_ACA_270 determined relatively high weight of plants, and locus linked to the marker BOT1_2_M_CAG_145 – in contrast low plant weight. Our results should be useful for large screening of collection and breeding material. This work was partially supported by grant RFBR 13-04-00128-a.

ОНТОГЕНЕТИЧЕСКАЯ НАСЛЕДСТВЕННОСТЬ ЭТО ОБЪЕКТИВНАЯ РЕАЛЬНОСТЬ

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В практическом растениеводстве экспериментально доказано, что семена, репродуцированные в различных агроэкологических условиях, будучи посеянными в одних и тех же условиях дают количественно различные уровни урожая. В результатах этих исследований было доказано, что это явление, каким то, образом наследуется семенами и реализуется в первом последующем поколении, а в следующих поколениях элиминируется, подобно эффекту гетерозиса и некоторыми учеными даже было акцентировано как географический гетерозис. Но при этом оставалось неизвестным, какой же фактор, из всех известных агроэкологических условий, является главным для создания в репродуцированных семенах высоких урожайных качеств (УК). За счет каких элементов продуктивности реализуются УК семян. Как в лабораторных условиях различать партии семян по их УК.

Пока генетики, в основном, имеют дело с наследственностью, сформированной в процессе филогенеза в течение миллионов лет эволюционного процесса, как ими постулируется, за счет спонтанного мутагенеза и последующего отбора внешней средой в борьбе за существование, и не учитывают направленное влияние внешней среды на наследственный аппарат в процессе онтогенеза растений.

Вместе с тем, в многолетних исследованиях, как в полевых, так и в вегетационных опытах в сосудах с песчаной культурой, природы (УК) семян на яровой твердой пшенице Харьковская 46 нами был выделен главный фактор из комплекса условий агроэкологической среды, влияющий на УК семян. Им оказался уровень влажности почвы. Дефицит почвенной влаги снижает УК семян. Путем анализа элементов продуктивности материнских растений пшеницы, выращенных из модифицированных семян на песчаном субстрате, нами был выявлен главный элемент продуктивности, за счет которого реализуются высокие УК семян. Это количество зерен в колосе. Это совпадает с реализацией эффекта гетерозиса, который также осуществляется за счет увеличения числа зерен в початке. Таким образом, нами доказано, что условия внешней среды в течение одного поколения ведут к групповой направленной изменчивости наследственного аппарата семян. Эта часть наследственности отделена нами от филогенетической наследственности и названа онтогенетической наследственностью.

Исследования модифицированных семян показало, что условия дефицита почвенной влаги формируют более крупные семена с низкими УК и индуцируют в них избыточный синтез глиаина и, в целом, белка. Дефицит почвенной влаги усиливает накопление фосфора в семенах. Этим нами опровергнута гипотеза, что чем крупнее семена и больше в них белка, и фосфора тем они урожайнее. Итак, высокоурожайные семена мельче и имеют пониженное содержание белка и фосфора. Таким образом, нами доказано, что высокие УК семян не обусловлены повышенным содержанием резервных питательных веществ в семенах, а имеют наследственную природу.

Изучение активности ферментов РНКазы и кислой фосфатазы показало более высокую активность в расчете на общее содержание белка у семян с высокими УК. Таким образом, синтеза запасного белка и функционально активных белков ферментов при формировании семян являются не зависимым. По высокой активности этих ферментов в расчете на общее содержание белка в семенах можно прогнозировать высокие УК семян.

Что касается природы онтогенетической наследственности, то еще в 1975 году нами при изучении молекулярных процессов фотосинтеза на кукурузе была высказана гипотеза, что гетерозис вызван гетерозиготностью по коротким хромосомным инверсиям, то же сейчас можно сказать и об УК семян. Механизм агроэкологического влияния на геном состоит в формировании инверсий в блоках генов, кодирующих водный обмен растений.

Наличие групповой экологически направленной изменчивости наследственности растений снимает главное обвинение в ошибочности теории Ч. Дарвина, основанной на спонтанной индивидуальной изменчивости, которое сам Ч. Дарвин назвал «кошмаром Дженкина» по неизбежному поглощению и элиминации индивидуально измененных генотипов внутри популяции. Наличие групповой изменчивости является гарантией сохранения генетических изменений в популяции в процессе образования половых гамет в мейозе и при скрещивании.

РАЗБАВЛЯЕТСЯ ЛИ БЕЛОК В ЗЕРНОВКЕ ПШЕНИЦЫ ПРИ ПОВЫШЕНИИ УРОЖАЯ

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В настоящее время существует мнение, что селекция на продуктивность вызывает падение содержания белка в зерне, которое якобы неизбежно связано разбавлением азота и фосфора в зерне при увеличении его биомассы. При этом есть и данные, что отрицательная коррелятивная зависимость может быть устранена применением азотных удобрений в определенный период вегетации, т. е. после цветения пшеницы. Иногда отрицательная связь проявляется не всегда, иногда может быть нулевой и даже положительной. По нашим многолетним экспериментальным данным, по влиянию минеральных удобрений и влажности почвы на урожай и белок, важнейшим фактором, влияющим на содержание белка в зерне пшеницы, является уровень влажности почвы. Поэтому для получения четкой функциональной зависимости содержания белка от урожая зерна, белок необходимо определять не по неорганическому азоту методом Кельдаля, а по числу пептидных связей биуретовым методом и все экспериментальные данные должны быть нанесены в соответствии с уровнями влажности почвы. Из рисунка видно, что наиболее сильное влияние на содержание белка имеет уровень влажности почвы. При дефиците почвенной влаги наблюдается

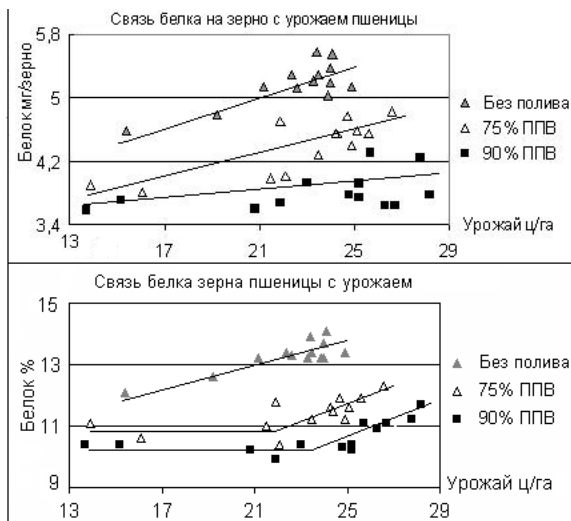


Рис. 1. Взаимосвязь урожая зерна твердой яровой пшеницы

Харьковская 46 с содержанием белка в одной зерновке в мг и в валовом зерне в % в зависимости от уровней и сочетаний минеральных удобрений и режимов влажности почвы прямая пропорциональная зависимость между урожаем и содержанием белка в зерне и зерновке. При повышении влажности почвы эта зависимость исчезает и содержание белка в зерне находится на одном уровне, слабо увеличиваясь в одной зерновке. При избыточном увлажнении до 90% ППВ эта связь исчезает и наблюдается эффект разбавления белка в зерновке, хотя в валовом зерне еще слабо возрастает.

Эту закономерность надо учитывать при индивидуальном отборе колосьев на продуктивность и биохимическом анализе на содержание белка в отдельных зерновках пшеницы. Ответ на важный для селекции вопрос, поставленный в заголовке, зависит от влажности почвы.

THE BREEDING STRATEGY AND HYBRIDIZATION OF TOMATO PLANTS WITH *d*-GENES IN THE SPECIAL BREEDING PROGRAM

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The special breeding program has been developed for the obtaining new tomato forms could be cultivated under special hydroponic conditions - on narrow benches. The first step of this program was the breeding of dwarf plants (30-35cm) with good productivity and big fruits. Using of gamete's selection approaches and 2-factorial dispersion analysis we show, that "the dwarf"-trait ($h^2=0,83$) and "the early ripening"-trait ($h^2=0,60$) could be inherited by father's line. But main traits of productivity - "the mass of one fruit" ($h^2=0,99$) and "the number of fruits on the plant" ($h^2=0,96$) - could be inherited by mother's line. We bred 9 mother and 7 father forms with needed parameters in the pre-breeding studies and developed strategies of selection and hybridization. We used two traditional breeding strategies:

- Selection from populations
- Hybridization

Selection from populations. We applied of gamete selection approaches for the acceleration of traditional breeding process. Gamete selection proposes the existence of sporophyte and microgametophyte phases in *Telomophytæ* plants. Division of gametophyte and sporophyte cycles in *Telomophytæ* evolution ensured significant advantages for adaptation of higher plant in the environment. And separate selection of gametes can be used in the breeding of needed forms – in gametes selection technologies. Morphologic markers of *d*-genes can to appear at seedling stage, so, the selection of dwarf forms from populations on the sporophyte level is easy. We bred 11 "dwarf" forms from about of 2000 samples with help of sporophyte selection during 3 years of breeding, and then we bred 2 forms with good productivity from 11 dwarf forms.

Hybridization. Obtaining of dwarf hybrids with good productivity and big fruits is connected with many problems. *d*-genes have some negative characteristics. Its are recessive and control the little mass of the fruit. But *d*-genes are located on the chromosome 2 near the group of genes, which are controlled "the early ripening" trait in *Solanum lycopersicum* L.. So, we can to expect, that its will be inherited with "early-ripening" genes during hybridization. *d*-genes are recessive and its will to appear in F_2 –progeny. We crossed mother forms with good productivity and big fruits with "dwarf" father forms, obtained F_1 – progenies and F_2 –progenies from self-pollinating of F_1 – progenies. F_2 –progenies were divided in the expected proportion: 3 parts of "high" plants and 1 part of "dwarf" plants. We bred some of early ripening plants with good productivity and big fruits from "dwarf" plants and studied F_3 –progenies of this "dwarf" plants. It was confirmed, that the strategy of hybridization was right: the height of the plant was lowered to the level of the "dwarf" father. The mass of one fruit and the productivity of plant were increased in 2 times. We obtained 1 hybrid form with needed parameters.

SOIURI DE SALVIA SCLAREA L. CREATE ÎN REPUBLICA MOLDOVA

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Soiurile de șerlai cultivate prin anii 60-80 ai secolului XX în Republica Moldova au fost create peste hotarele țării noastre și ne fiind adaptate la condițiile pedoclimatice locale. Investigațiile efectuate cu utilizarea metodelor moderne, s-au creat linii consangvinizate și androsterile, a început în Republica Moldova în 1978. Unul din obiectivele de bază în ameliorarea șerlaiului este crearea hibridilor și soiurilor noi cu productivitate sporită de inflorescențe și ulei esențial de calitate, cu termeni de maturizare tehnică diferiți, adaptate la condițiile pedoclimatice ale Moldovei.

Crearea și includerea liniilor androsterile și consangvinizate de diferită proveniență în schemele de hibridare au contribuit la elaborarea hibridilor simpli. Soiurile de proveniență hibridă create sunt rezultatul multiplexelor cercetării pe parcursul a câtorva generații de hibridi, care au servit ca bază în elaborarea soiurilor: timpuriu - Dacia 50, mediu - Dacia 99 și tardiv - Victor. Soiul Dacia 50, este un hibrid simplu, soi omologat din 1994 care a fost obținut în rezultatul încrucișării liniei 0-14 și cu linia 0-20. Soiul Dacia 99, hibrid simplu, ce rezultă din hibridările dintre linia S-1122 N2 și linia 0-30 care a fost omologat din 2004. Soiul Victor, hibrid simplu, este obținut în rezultatul încrucișării liniei M-69 5S₄ ce provine de la soiul autohton Moldovenesc 69 cu soiul Voznesensc 24. Soiul Victor a fost omologat în 2004. Datele experimentale demonstrează, că soiurile noi create depășesc soiul martor M-404 după producția de ulei esențial. Spre exemplu, soiul Victor în medie pe trei ani a garantat 74,2 kg/ha producție de ulei esențial, depășind cu 14,2 kg/ha soiul martor M-404. În patru cicluri de testare productivitatea medie a soiurilor Dacia 99 și Victor a constituit 56,3kg/ha (de la 34,2 până la 74,2kg/ha) și 57,3kg/ha (de la 41,8 până la 73,7kg/ha).

Un rol important în crearea soiurilor performante de șerlai îl manifestă hibridii de diferită complexitate servind drept ca bază în elaborarea soiurilor cum ar fi: Nataly Clary, Ambra Plus și Balsam. *Soiurile înfloresc și realizează producție de materie primă și ulei esențial din primul an de vegetație, iar plantațiile pot fi exploatare trei ani. Studiind indicii caracterelor cantitative ale soiurilor noi s-a constatat, că plantele sunt bine dezvoltate, cu talia de peste 110,0 cm și au format inflorescențe lungi compacte.*

Soiul tardiv, Nataly Clary, reprezintă un hibrid în trepte F₅, obținut prin încrucișarea hibridului simplu (S-3 x Cr.p. 8S₂) F₂ cu linia consangvinizată S-1122 4S₂ ce provine de la soiul Crâmskii rannii. Soiul Nataly Clary a fost omologat din 2006 și pe parcursul anilor s-a dovedit a fi cel mai rezistent la ger și iernare. Testat în culturi comparative de concurs, asigură o producția de inflorescențe medie în trei ani de cultivare de peste 2,8t/ha. Productivitatea medie a soiului în trei ani de exploatare a plantației a constituit 57,9kg/ha ulei esențial (de la 39,1 până la 72,5kg/ha).

Soiul timpuriu, Ambra Plus reprezintă un hibrid backcross F₆ la crearea căruia au participat hibridul triplu (K-36x0-41)F₂ încrucișat cu linia consangvinizată timpurie 0-19S₇. Backcrossarea efectuându-se cu linia consangvinizată 0-19S₇. Soiul Ambra Plus este omologat din 2009 și se caracterizează prin înflorire abundentă în primul an de vegetație. Testat în culturi comparative de concurs, în trei ani de exploatare a plantației asigură cea mai înaltă producție de materie primă în comparație cu soiurile omologate. În patru cicluri de testare (anii 2005-2013) a înregistrat cea mai ridicată producție de ulei esențial în anul întâi de vegetație 2005 - 26,8 kg/ha și anul 2011 - 22,3 kg/ha.

În 2014 a fost omologat soiul Balsam, cel mai timpuriu soi, care reprezintă un hibrid în trepte complex în generația F₁₀. Spre exemplu, soiul Balsam, în sumă pe trei ani de vegetație, asigură obținerea a 21,2 t/ha de inflorescențe și 79,5 kg/ha de ulei esențial, depășind cu 15,7 kg/ha soiul standard timpuriu Dacia 50. E cazul să menționăm, că soiurile testate în condițiile climatice de secetă și arșiță din aprilie până în septembrie ale anului 2012, în anul al doilea de vegetație au format producții înalte de inflorescențe de la 10,4 t/ha până la 11,7t/ha, cu conținut sporit de ulei esențial: de la 1,143% (s.u.) la soiul timpuriu Dacia-50 până la 1,494% (s.u.) la soiul Balsam.

Astfel, în rezultatul a peste 35 ani de cercetare la specia *Salvia sclarea* L. au fost create și omologate 7 soiuri de proveniență hibridă cu productivitate sporită, calitate superioară a uleiului, cu termeni diferiți de maturizare ce înfloresc și realizează producții de materie primă și ulei esențial din primul an de vegetație. *Soiurile noi pot fi exploatare trei ani în loc de doi și sunt rezistente la condițiile specifice de iernare ale țării noastre. Cultivate concomitent soiurile timpurii, intermediare și tardive formează în timpul recoltării un conveier, asigurând recoltarea fiecărui soi în termeni optimi, excluzându-se pierderile de materie primă și ulei esențial.*

UTILIZAREA POPULAȚIILOR SINTETICE LA CREAREA LINIILOR CONSANGVINIZATE DE PORUMB INDURATA

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Crearea cu succes a liniilor consangvinizate de porumb în mare măsură depinde de diversitatea genetică a materialului biologic inițial. Programul de ameliorare a demarat cu dezvoltarea a 6 populații sintetice HK169-3, Pi187, MKP18, F7, 4091/94 și F564 cu participarea a mai mult de 6 linii consangvinizate de tip indurata și predominarea în genotip a 50% genomului unei linii de bază. În scopul concentrării genotipurilor cu caractere agronomice valoroase populațiile au fost îmbunătățite prin metoda selecției recurente fenotipice. Populațiile inițiale s-au caracterizat prin diversitatea largă a plantelor după perioada până la înfloritul organelor reproductive și a caracterelor fenotipice. Anual populațiile s-au semănat pe 50 parcele cu un volum de 1500 plante, dintre care jumătate serveau ca forme materne cu înlăturarea paniculelor și polenizare cu amestec de polen colectat de pe plantele paterne. Plantele materne au fost supuse unei selecții riguroase, selectându-se până la 100 știuleți elită pentru următorul ciclu de selecție recurentă. Urmează să accentuăm că pe parcursul a mai multor ani o cotă semnificativă a materialului biologic a fost rebutată din cauza frângerii tulpinilor, sensibilității la secetă, productivității scăzute și taliei joase a plantelor. Familiile selectate fenotipic în generațiile următoare au manifestat rezistență înaltă la condițiile stresante de mediu și capacitate generală de combinare înaltă în test-încrucișări. Descendențele cu performanțe au constituit baza materialului inițial pentru următoarele cicluri de selecție cumulativă. Liniile consangvinizate obținute în ultimii ani din materialul respectiv posedă un șir de caractere ameliorative superioare celor evidențiate la etapele precedente. Liniile noi se deosebesc prin multiplele caractere fenotipice și însușiri agronomice apreciate vizual, posedă capacitate generală și specifică de combinare. În rezultatul experimentării liniilor respective în culturi comparative de orientare s-au depistat 26 familii fenotipic omogenizate. După o triere mai riguroasă în următorii ani pe diferite fundaluri stresante s-au evidențiat 19 linii consangvinizate cu bob indurata transferate în colecția operațională a laboratorului. În procesul de creare a hibridizilor, efectuat în încrucișări sistematice de tip topcross, liniile indurata s-au utilizat ca forme paterne, iar ca forme materne au fost selectate linii consangvinizate și hibridi simpli înrudiți din grupele de germoplasma Reid Iodent, BSSS-B37 și Lancaster. În baza datelor experimentale obținute în culturi comparative de preconcurs a fost estimată capacitatea de combinare a liniilor indurata. Analiza arată că valori combinative înalte după producția de boabe au manifestat liniile consangvinizate MKP19A și MKP22, care au realizat cele mai superioare performanțe în hibridi. Pe parcursul a mai multor ani liniile respective au fost studiate per se în culturi comparative de orientare pe parcele cu suprafața de 10m² în două repetiții. Din cadrul acestora interes ameliorativ reprezintă liniile timpurii MKP19A și MKP20 cu participarea pedigreeului a donatorilor Pi187 și respectiv HK169-3. Ambele linii manifestă rezistență înaltă la cădere și frângere a tulpinilor, se deosebesc distinct prin calitate înaltă a boabelor, toleranță înaltă la temperaturi scăzute și germinație bună, apreciată în termeni timpurii de semănat. Linia MKP19A a manifestat ritm intens de creștere a plantulelor la faza de 5-7 frunze și producție de boabe înaltă. În condiții climatice favorabile ale anului 2010 linia respectivă a format la plantă câte doi știuleți bine dezvoltati la recoltare. Menționăm că liniile respective servesc ca forme materne în hibridul ultratimpuriu Rosmold 159CRF, omologat în Federația Rusă. Următoarele linii cu caractere ameliorative importante MKP21/182 și MKP22 au fost create cu participarea germoplasmei donatorilor F564 și HK169-3. Rezultatele cercetărilor obținute ne permit să constatăm, că liniile respective se deosebesc prin producție relativ înaltă și umiditate a boabelor la nivelul matorului. Linia MKP21/182 posedă talie înaltă a plantelor iar MKP22CRF pierde rapid umiditatea din boabe după maturitatea fiziologică, caracter mai rar întâlnit la liniile din convاریetatea Indurata. Este necesar de accentuat că MKP21/182 este forma paternă a hibridului Rosmold 202MRf, înregistrat din 2013 în Registrul soiurilor de plante pentru cultivare la boabe și siloz a Rusiei, iar MKP22 este forma paternă a hibridului Bemo203 omologat recent în Belarus. O deosebită atenție merită liniile AN6560/02 și MKP27 extrase din populația sintetică LO3, care se evidențiază prin talie înaltă a plantelor cu multiple ramificații a paniculului și capacitate înaltă de polenizare, caractere moștenite de la genitorul LO3. O deosebire esențială prezintă rezistența plantelor la frângere și cădere, umiditatea scăzută în boabe, în mod deosebit linia MKP27. Datorită perioadei de vegetație mai mare și calității boabelor cu sticlozitate și culoare portocalie, liniile sunt utilizate la crearea hibridizilor cu destinație alimentară. Linia MKP27 este folosită în calitate de formă paternă în hibridul Alimentar 325 inclus în Registrul soiurilor de plante în R. Moldova pentru cultivare la boabe din anul 2014.

MODEL DE APRECIERE CORECTĂ A SEGREGĂRII CARACTERELOR PE CLASE FENOTIPICE LA HIBRIZI DIN GENERAȚIA A DOUA

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Această segregare este rezultatul repartiției a “**k**” clase genotipice în “**c**” clase fenotipice. Astfel, că din **Q** evenimente statistice (observări) în fiecare clasă genotipică revin câte **q=Q:k** evenimente. Fiecare clasa fenotipică este formată din $\sum m_i$ -genotipuri sau câte **F_i=q·m_i** evenimente în realitate și sub acțiunea aleatorie a factorului **X** frecvențele reale sunt **f_i**, care sunt legate coliniar cu **F_i**. Astfel că **f_i** și **F_i** nicidecum nu sunt frecvențe de origine aleatorie și deci nu pot fi nemijlocit apreciate statistic prin criteriile teoretice χ^2_{teor} , t_{teor} etc. Astfel algoritmul $\chi^2 = \sum (f_i - F_i)^2 / F_i$ nicidecum nu corespunde algoritmului real $\chi^2 = \sum (v_i - np_i)^2 / np_i$, unde **v_i** sunt variabile aleatorii empirice ; **p_i** sunt probabilitățile repartiției Poisson și $\sum v_i = n$; iar $\sum p_i = 1$ (1).

Anume diferențele **x_i=f_i-F_i** sunt de natura aleatorii și se supun repartiției normale . Deaceia această variabilitate și poate fi apreciată prin intermediul criteriului teoretic **F**(Fisher) cu condiția de a restabili și valorile ipotetic așteptate **x₀** pentru fiecare clasă fenotipică. Deoarece frecvențele **F_i** și variabilele **X_i** între ele nu există nici o corelație valoarea a variabilei ipotetic așteptată **x₀=q·a_{teor}** unde **a_{teor}** este nivelul de semnificație la care se efectuează aprecierea statistică **a_{teor}=1-P_{teor}**. De obicei **a_{teor}=0,05**. Această variabilă este una rudimentară deoarece este identică pentru toate clasele. Deaceia ea este concomitant și abaterea standard **δ=x₀**.

Volumul eșantionului ipotetic este **n=c x₀** și dispersia **δ²=x₀²**. Aprecierea statistică se efectuează prin criteriul **F_{teor}** (**Fisher**) pentru gradele de libertate **γ₁=γ₂=n** (pentru eșantioanele conjugate dispersiile sunt nedepasate . Unica restricție constă că valoarea minimală a dispersiei **δ²>5**. Valoarea **F=Σ (f_i - F_i)² / δ²_x =Σ x² / δ²_x** Ipoteza nulă se admite cînd **F_{teor}>f** și indică că variabilele **x_i** au distribuție normală și deci factorii genotipice au acțiune semnificativă.

Experimentatorul este cointerestat a evedenția în cazul respingerii ipotezei nule a evedenția datorită cărei variante se datorește această respingere . În acest caz ne folosim de criteriul **R_{teor}=√F_{teor}** pentru gradele de libertate **γ₁= x₀** și **γ₂=cx₀**. Variabilele **x_i** (ele și sunt valorile mediei pe variante se compară cu **DL_{teor} = R_{teor} S_{x0}**. Este îndeajuns ca o singură valoare **x_i** să fie mai mare ca **DL_{teor}** că ipoteza nulă se abandonează pentru eșantionul integru. Variabilele **x_i>DL_{teor}** indică și lipsa sau surplusul de evenimente din clasa dată ceea ce permite a formula noi ipoteze.

Prezentăm pe un model experimental citat din B.Dospheov 1979 pag.238 privitor la segregare dihibridă ab de tipul (1x2x1)x(1x2x1). În generația a doua 4x4=16 genotipuri formează 3x3 clase fenotipice în care se repartizat **q=304** evenimente statistice. Deci fiecare genotip e format din **q=304:16=19** evenimente statistice ale factorilor genetici și din **q=19·0,05=0,95≈1** evenimente statistice aleatorii. Aceia aprecierea segregării dihibride este imposibilă. Frecvențele **f_i** indică că se resimte lipsa evident anume a fenotipului **bb**. Deaceia se determină variabilele **x_i=f_i-F_i** pentru segregarea mo nohibridă **BBX2Bbx1bb**. Vezi tabela 1.1

Aprecierea segregării monohibride

Tipul segregării	1BB	2BB	1bb	1BB	1,8Bb	0,7bb
Variabile x_i=f_i-F_i	+12	+7	-19	+1,2	+2,6	-3,8
Valoarea DL_{teor}	=√F_{teor}·X₀=√3,351·3,8=±6,90			=√3,18x4,34=7,54		

Observăm că în prima variantă segregare de tipul 1x2x1 se respingedin cauză lipsei de evenimente **bb** și surplusului de evenimente **BB**. Ipoteza secundă precum că segregarea e de tipul **1x1,8x0,7**se confirmă. Deci segregarea dihibridă este nu de tipul **(1x2x1)x(1x2x1)** ci de tipul **(1x2x1)x(1x1,8x0,7)**. Metoda tradițională confirmă prima ipoteză și deci a doua ipoteză nici nu se preccută.

SEMNIIFICAȚIA APLICATIVĂ A COEFICIENTULUI DE EREDITARE

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Acest coeficient în sensul restrîns al eredității reprezintă cota parte a variabilității caracterului cantitativ **A** ce se transmite și se regăsește la descendenții generativi. El reprezintă raportul variantelor (dispersiilor sau variațiilor) $h^2 = S^2_g / S^2_p$ genotipică la fenotipică (sumară). Evident că în acest caz coeficientul h^2 este un mod de evaluare a variabilității genotipice prin unități relative de măsură $h^2 = \delta^2_g = S^2_g / S^2_p$. Deoarece componența principală din varianta genotipică este cea aditivă **h** ea se substituie și obținem $h^2 = S^2_g / S^2_p$. Acest coeficient are un impact direct pentru ameliorarea , deoarece se consideră că cu cît mai mare este valoarea h^2 pentru caracterul dat cu atît procesul de ameliorare va fi mai efectiv. Surpriza urmează în continuare, evaluare. Surprinzător este faptul că coeficientul h^2 se evaluează prin intermediul „coeficientului corelației” sau al regresiei .

Astfel relația unul din părinți descendeți se evaluează prin algoritmul $h^2 = S^2_g / S^2_p = 2r$ sau $h^2 = 2b_{y/x}$; unde **r** este coeficientul corelației și **b** al regresiei. Ambii coeficienți variază în limitele 0-1. Deci din start h^2 nu poate avea valorile $2r$ sau $2b_{y/x}$ deoarece $h^2 > 1$ nu poate fi mai mult ca acesta corelația operează numai cu variabile aleatorii x_i și y_i , care nu reflectă valorile reale ale caracterului **A** aflat în studiu.

Una din pricini e că **r** și **b** sunt considerați a fi de ordinul unu, iar h^2 e valoare patrată și deci la fel nu pot fi compatibile. Intenționat sau nu dar din punct de vedere statistic lipsește a treia componentă. Aceasta este variabilitatea paratipică a caracterului. Ea reprezintă o valoare aleatorie (întîmplătoare) și deaceia o semnăm prin $S^2_z = S^2_p - S^2_g$ sau $S^2_p = S^2_g + S^2_z$ sau în unități relative $1 = \delta^2_g \pm \delta^2_z$.

Semnificația variantei genotipice se apreciază prin criteriul teoretic **F (Fisher)**. Menționăm că aceasta este unul deplasat la dreapta, cu valorile teoretice > 1 . Aceasta înseamnă că la numărătorul fracției pentru **F** se află valoarea variabilității ce include în sine și componența aleatorie . Astfel $F = 1: \delta^2_z = S^2_p / (S^2_p - S^2_g)$. Un model de evaluare a variabilității caracterului **A** reprezintă experimentul monofactorial . Datele experimentale **Y_i** sunt repartizate pe variantele **A_i** (forme parentale și hibridă sau alte relații) cu mediile **A_i**. Variabilele $z_i = Y_i - A_i$ formează variansa $S^2_z = \sum z_i$.

Variația fenotipică sumară $S^2_p = \sum y_i^2$ unde $y_i = Y_i - M$; iar $M = \sum Y_i / n$. Astfel că $h^2 = S^2_p / S_z$ se compară cu **F_{teor}** pentru gradele de libertate $\gamma_1 = \gamma_2 = n$. Variansa genotipică S^2_g este semnificativă când **F** > **F_{teor}**.

Semnificația h^2 permite de a nemijlocit și relațiile forma parentală –hibrid prin raportul între aceste componente. Acest raport rămîne a fi semnificativ. Ele au atribuții directe la aprecierea heterozisului, capacității combinative și a altor particularități a formelor implicate în astfel de relații de ereditate.

STAREA ACTUALĂ ȘI UNELE OPORTUNITĂȚI DE REVITALIZARE ÎN PRODUCEREA SEMINTELOR DE LEGUME

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Agricultura Republicii Moldova, ca una din principalele ramuri ale economiei naționale, se află în fața unor provocări menite să demonstreze capabilitatea asigurării alimentare a țării și posibilităților de export în produse agricole. Rolul științei în dezvoltarea sectorului agrar este indiscutabil, în special al biologiei, geneticii și ameliorării, fiziologiei și protecției plantelor. Legumicultura a fost și continuă să rămână una din principalele ramuri ale agriculturii, menită să asigure populația cu legume în stare proaspătă și industria de conserve cu materie primă. Importanța social-economică a acestei ramuri rezultă din ponderea legumelor în alimentația populației, utilizarea intensivă a terenurilor și forței de muncă, obținere de producții sporite precum și a unor venituri esențiale. Condițiile pedoclimatice favorabile, tradițiile și experiența acumulată permit cultivarea a peste 60 specii de plante legumicole și obținerea unor recolte bogate precum și a unui beneficiu impunător la majoritatea culturilor.

Structura de cultivare a legumelor practică în ultimii ani este departe de cea optimă, care să corespundă mai pe deplin cerințelor pieții, securității alimentare, posibilităților de export și anume: tomatele - 40%, mazărea de grădină - 10%, ceapa, varza, ardeiul, și castravetele - 7-8%, vinetele, morcovul și sfecla de masă - câte 4-5%. Dezvoltarea durabilă a legumiculturii necesită un studiu permanent asupra nivelului de producere, creare a soiurilor și hibridilor noi, elaborarea și implementarea unor metode și tehnologii de producere a semințelor, răsadurilor și de cultivare a legumelor, în stare să asigure o producție cu o valoare biologică și competitivitate înaltă în condiții deficitare de apă și resurse energetice. În trecut 80 la sută din suprafețele cultivate cu legume erau însămânțate cu soiuri autohtone. În actualul Registru al soiurilor omologate în Republica Moldova sunt incluse 416 soiuri, dintre care create în instituțiile locale au rămas doar 20%. O situație mai dificilă se atestă în producerea semințelor de legume și materialului săditor de cartofi, din cauza insuficienței semințelor de categorii superioare (bază și prebază). La moment nu se efectuează lucrări de ameliorare conservativă și de menținere a soiurilor la culturile de varză, ceapă, rădăcinoase, cucurbitacee, leguminoase și verdețuri. În urma reformei agrare și privatizării gospodăriilor semincere s-a dispersat considerabil sistemul de producere a semințelor de legume. Mulți dintre producătorii actuali sunt dispuși să cuprindă un număr cât mai mare de specii și soiuri, ceea ce deseori complică amplasarea culturilor în spațiu și obținerea semințelor de calitate corespunzătoare. Întru evitarea amestecurilor în perioada recoltării, extragerii și condiționării semințelor se recomandă a limita la minimum soiurile din aceeași specie. Pentru îmbunătățirea calității semințelor trebuie ca producătorii să fie autorizați în producerea semințelor de soiuri concret nominalizate pe parcursul a mai multor ani. Aceasta v-a conduce la o eventuală specializare și însușire mai detaliată a particularităților soiurilor, reglare a volumelor de producție precum și evidențierea celor mai competitivi producători și realizatori de semințe. În scopul implementării mai eficiente a tehnologiilor avansate, exercitării unui control, care să contribuie la sporirea calității este necesară alocarea investițiilor și concentrarea producerii semințelor în baza unor asociații specializate cu asistență științifică corespunzătoare, care dispun de baza material-tehnică, specialiști și experiență în acest domeniu pentru:

- Menținerea fondului genetic, crearea soiurilor și hibridilor competitivi, producerea semințelor de categorii superioare de legume, cartofi și flori.
- Asigurarea necesităților pieței interne și exportului cu semințe de calitate corespunzătoare, construcția unei fabrici de condiționare și ambalare a semințelor de legume.
- Elaborarea elementelor tehnologice noi, perfecționarea tehnologiilor de cultivare și valorificare a legumelor, producerea și condiționarea semințelor de legume.
- Perfecționarea sistemelor de protecție a culturilor legumicole și cartofului de buruieni, boli și dăunători.
- Pregătirea cadrelor înalt calificate și dezvoltarea continuă a cercetărilor în legumicultură și la cultura cartofului. Organizarea sectoarelor demonstrative pentru promovarea rezultatelor științifice, instruirea specialiștilor și fermierilor.
- Promovarea progresului tehnic, implementarea de noi mașini și utilaje, rezultatelor științifice din alte domenii în scopul reducerii cheltuielilor de producere și resurselor energetic la cultivarea legumelor, sporirea competitivității acestora pe piața internă și externă.

ИЗУЧЕНИЕ МЕСТНЫХ ПОПУЛЯЦИЙ НЕСТРЕЛКУЮЩЕГОСЯ ЧЕСНОКА

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Рыночные и экономические преобразования отрицательно отразились на поддержании селекционно-семеноводческих работ, сохранении и расширении коллекций, особенно на культурах с ограниченными объемами производства. В последнее десятилетие не проводились какие либо исследования по улучшению сортового состава чеснока. По этой причине из-за отсутствия сортов местной или зарубежной селекции несколько лет назад чеснок, как овощная культура, не значился в списке районированных сортов для возделывания в Республики Молдова, хотя его всегда выращивали и традиционно использовали в местной кулинарии. В связи с этим в 2013 г нами зарегистрирован стрелкующийся сорт Изумруд, выведенный из местных форм, отличающийся высокой продуктивностью и особыми качествами.

Необходимо отметить, что в отличие от других овощных культур чеснок сильнее реагирует на изменение почвы и климата и значительно труднее приспосабливается к условиям произрастания. Причина тому вегетативное его размножение. Поэтому опытные производители обычно выращивают в основном местные формы чеснока, хорошо приспособленные к условиям данного региона. Сорта и формы, интродуцированные из других географических зон, используемые в качестве посадочного материала в почвенно-климатических условиях Молдовы плохо развиваются, в результате чего формируют низкий урожай, иногда погибают.

В Республике Молдова нестрелкующийся чеснок в основном выращивают на приусадебных участках, что связано с невысокой урожайностью и сложностью семеноводства, так как этот вид чеснока размножается только вегетативным способом - посадкой зубков. У нестрелкующего чеснока в процессе вегетации развиваются только листья, образующие в нижней части ложный стебель. Особенность таких растений-отсутствие семенного размножения, которое было утрачено в процессе эволюции вида. Вместе с тем следует отметить, что формы нестрелкующегося чеснока являются более устойчивы к болезням и обладают более длительным периодом хранения.

Селекционная работа по отбору форм нестрелкующегося чеснока ведется с 2010 года. Исходным материалом для селекции нестрелкующегося чеснока служили местные популяции. Собрана коллекция нестрелкующегося чеснока, которая поддерживается и постоянно расширяется из местных сортовых популяций, отобранных в северной и южной зонах. Для изучения характерных признаков собранный материал выращивался на высоком агрофоне. Коллекция включает разнообразные формы по количеству и плотности зубков, крупности луковицы, количеству чешуи и ее окраски и другим признакам. Отбор форм нестрелкующегося чеснока проводился по следующим селекционным свойствам: урожайность, качество луковиц, устойчивость к болезням, длительность хранения, крупность луковиц с хорошо сформированными зубками, однородность.

В ходе исследований выявили, что с расширением ареала выращивания у некоторых местных популяций нестрелкующегося чеснока проявляется неустойчивость биологических и морфологических признаков. Согласно полученных данных характерные признаки популяций чеснока северного региона являются более устойчивыми в сравнении с формами из южных зон. Вероятно, это свидетельствует о том, что при размножении чеснок вырождается не только под влиянием фитопатогенов, но и под действием экстремальных экологических факторов, что приводит к возникновению мутаций. Учитывая, что из вегетативно размножающихся растений сложно создать гибридные комбинации, полученные таким образом мутации являются селекционным материалом для создания местных сортов путем стабилизации и сохранения ценных признаков.

В результате изучения местных популяций и отбора по наиболее ценным признакам нами выделены стабильные однородные формы нестрелкующегося чеснока, отличающиеся высокой урожайностью и длительным периодом хранения, которые не формируют воздушную стрелку даже при осенней посадке. Дальнейшие отборы направлены на выделение сортов с устойчивыми хозяйственными признаками с последующим изучением реакции наиболее перспективных форм на различные агротехнические параметры – применение удобрений, орошение, густоты и сроков посадки в зависимости от величины посадочного материала, сроки уборки и хранения, экономическая эффективность, а также содержание биологически ценных веществ.

VARIABILITATEA PARAMETRILOR GENETICI PENTRU REZISTENȚA LA RAPĂN ÎN HIBRIDĂRILE DE DIALELĂ ÎN FUNCȚIE DE ANI

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Rapănul mărlui, *Venturia inaequalis* Cooke Wint, este una dintre cele mai periculoase maladii și aduce mari pagube recoltelor de mere în toate regiunile de cultivare a speciei. Impactul economic poate constitui o pierdere în recoltă de 50-80%. În anii epifitotici deseori toate fructele sunt atacate și nu pot fi folosite nici în stare proaspătă nici în scop de industrializare. Deaceia cercetarea eredității și variabilității parametrilor genetici în scopul alegerii corecte a soiurilor parentale și creării de noi soiuri imune este o necesitate irevocabilă.

Ca material pentru cercetare au servit 2400 hibrizi de măr obținuți de la încrucișarea soiurilor imune la rapănul mărlui în baza genei Vf: - Florina, Priam, Coremodet, Coredar, Remo, Coredana. Hibridările au fost efectuate conform metodei 3 B. Griffing în anul 2002. Semănatul semințelor hibride a fost efectuat în toamna anului 2002. În combinațiile hibride nu s-a efectuat selecția preventivă. Livada de selecție a fost plantată în primăvara anului 2005, în 4 repetări, cu plante hibride de doi ani. Distanța de plantare 4 x 0,75m. Colectarea datelor privind atacul de către rapăn a fost efectuată în anii 2007, 2010, 2013 prin notare de la 1 la 6 unde nota 1- atacul lipsește iar nota 6 atac foarte puternic. Analiza dispersiilor și de dialelă ne arată că indicii parametrilor genetici ai soiurilor parentale se comportă diferit în funcție de ani (tab. tab. 1, 2).

Tabelul 1. Variabilitatea indicilor heritabilității în funcție de ani

Anul	Heritabilitatea în sens larg h^2_{ws} , %	Heritabilitatea în sens strict h^2_{cs} , %	Cota de acțiune neaditivă h^2_{ns} , %	Efectul reciproc h^2_{rs} , %	Erori σ^2 , %
2007	93,0	83,0	8,9	0,0	8,0
2010	87,8	40,0	31,1	15,8	13,1
2013	91,6	40,7	32,1	16,9	10,3

Astfel heritabilitatea în sens larg h^2_{ws} , practic nu variază. Genotipurile soiurilor cercetate sunt foarte variate după caracterul dat și sunt influențate slab de condițiile mediului. Deci ereditatea rezistenței la rapăn depinde de genotipurile soiurilor parentale cercetate. Variabilă este acțiunea aditivă a genelor, determinată de heritabilitatea în sens strict h^2_{cs} , de la 83,0 până la 40,0%. Cota de acțiune neaditivă h^2_{ns} , la fel variază de la 8,9 până la 32,1%. În anii în care indicii acțiunii aditive și neaditive tind a avea o acțiune egală asupra formării caracterului cercetat la hibrizi s-a depistat și efectul reciproc.

Tabelul 2. Variabilitatea efectelor CCG (\hat{g}_i) și varianțelor CCG ($\sigma^2_{\hat{g}_i}$) și CCS ($\sigma^2_{\hat{s}_i}$) în funcție de ani

Soiul	Efectele CCG (\hat{g}_i)			Varianțele CCG ($\sigma^2_{\hat{g}_i}$)			Varianțele CCS ($\sigma^2_{\hat{s}_i}$)		
	2007	2010	2013	2007	2010	2013	2007	2010	2013
Florina	0,1554	0,1805	0,2707	0,0241	0,0323	0,0733	0,0014	0,0212	0,0311
Priam	0,0682	0,1402	0,2103	0,0046	0,0194	0,0422	0,0006	0,0131	0,0214
Coremodet	0,0066	-0,0376	-0,0564	-0,0001	0,0012	0,0032	0,0005	0,0185	0,0199
Coredar	-0,0286	0,0239	0,0358	0,0007	0,0003	0,0013	0,0001	0,0036	0,0096
Remo	-0,0920	-0,1695	-0,2542	0,0089	0,0285	0,0646	0,0014	0,0022	0,0034
Coredana	-0,1095	-0,1376	-0,2064	0,0119	0,0187	0,0426	0,0018	0,0151	0,0263
Greșala standard	$\pm 0,0106$	$\pm 0,0244$	$\pm 0,0365$	$\pm 0,0106$	$\pm 0,0244$	$\pm 0,0365$	$\pm 0,0097$	$\pm 0,0422$	$\pm 0,0514$

Efectele capacității combinative generale CCG (\hat{g}_i) la soiurile parentale variază în funcție de ani dar în general denotă tendința de a păstra însemnătatea acțiunii genelor la soiurile cercetate. La soiurile Florina și Priam efectele sunt stabil pozitive și semnificative. În combinațiile lor hibride pot fi anual multe plante atacate puternic de rapăn. La soiurile Remo și Coredana efectele CCG sunt semnificative și în funcție de ani stabil negative. În combinațiile lor hibride pot fi anual mai puține plante atacate puternic de rapăn. Varianțele CCG ($\sigma^2_{\hat{g}_i}$) ale soiurilor cercetate, cu toate că în multe cazuri sunt nesemnificative dar practic variază în conformitate cu variabilitatea efectelor CCG (\hat{g}_i). Varianțele capacității combinative specifice CCS ($\sigma^2_{\hat{s}_i}$) în majoritatea cazurilor sunt nesemnificative și au o variabilitate neînsemnată. Totodată din raportul dintre varianțele CCG ($\sigma^2_{\hat{g}_i}$) și varianțele CCS ($\sigma^2_{\hat{s}_i}$) rezultă că în fiecare an soiurile Florina și Priam posedă acțiune aditivă spre formarea mai multor hibrizi puternic atacați iar Remo și Coredana stabil posedă acțiune aditivă spre formarea de hibrizi mai slab atacați. La soiurile Coremodet și Coredar raportul dintre varianțele CCG ($\sigma^2_{\hat{g}_i}$) și varianțele CCS ($\sigma^2_{\hat{s}_i}$) indică variabilitate a acțiunii aditive sau neaditive ale genelor spre formarea la hibrizi a caracterului cercetat.

EREDITATEA CARACTERELOR FRUNZEI LA SOIURILE DE MĂR IMUNE LA RAPĂN ÎN HIBRIDĂRILE DE DIALELĂ

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Frunza este organul principal al plantei, care prin intermediul fotosintezei pune la dispoziția pomului și fructului substanțele nutritive. Mărimea frunzei are însemnătate pentru cantitatea și calitatea producției, posibilitățile de păstrare a fructelor, rezistența pomului la secetă etc. atât pentru soiurile tradiționale cât și pentru soiurile rezistente și imune la răpănul mărului care nu se stropesc contra acestei boli. Lungimea pețiolului are însemnătate pentru intensitatea de aerisire a coroanei și fototropismul frunzei în cursul zilei. Frunzele cu pețiolul lung ușor se dispun razelor solare fotosinteza fiind mai activă și efektivă. În același timp astfel de frunze, cu pețiolul lung, mai ușor se restrâng în caz de secetă decât cele cu pețiolul scurt. Astfel stabilirea eredității caracterelor frunzei are însemnătate și în procesul de alegere a soiurilor parentale pentru hibridări.

Ca material pentru cercetare au servit 2400 hibridi de măr obținuți de la încrucișarea soiurilor imune la răpănul mărului în baza genei Vf: - Florina, Priam, Coremodet, Coredar, Remo, Coredana. Hibridările au fost efectuate conform metodei 3 B. Griffing în anul 2002. Semănatul semințelor hibride a fost efectuat în toamna anului 2002. În combinațiile hibride nu s-a efectuat selecția preventivă. Livada de selecție a fost plantată în primăvara anului 2005, în 4 repetări, cu plante hibride de doi ani. Distanța de plantare 4 x 0,75m. Colectarea datelor privind caracterele frunzei a fost efectuată în anul 2012 prin notare.

S-a stabilit că nivelul de heritabilitate pentru suprafața frunzei h^2w este de 51,6%, pentru lungimea pețiolului alcătuiește 40,7%, pentru lățimea frunzei 14,8% iar pentru lungimea frunzei 27,39%. Raportul dintre lungimea și lățimea frunzei a indicat nivelul h^2w numai de 22,7% iar raportul dintre lățimea frunzei și lungimea ei- 61,0%. Indicii heritabilității în sens larg pentru caracterele enumerate sunt semnificativi cu nivelul de probabilitate mai înalt de 0,05. Heritabilitatea în sens larg h^2w determină ereditatea în totalitate, care cuprinde heritabilitatea în sens strict (h^2e acțiunea aditivă a genelor), acțiunea neaditivă (h^2s epistazia și dominanța) și efectul reciproc, h^2re .

Suprafața frunzei se transmite prin ereditate prin acțiunea aditivă a genelor parentale indicele heritabilității în sens strict h^2e este practic egal cu h^2w alcătuind 49,8%. Efectul acțiunii neaditive este numai de 11,5% iar efectul reciproc nu se depășește.

Ereditatea lungimii pețiolului este determinată de acțiunea aditivă a genelor formelor parentale deoarece indicele heritabilității în sens strict h^2e determinat de capacitatea combinativă generală (CCG) alcătuiește 38,2%, practic fiind aproape de indicele heritabilității în sens larg h^2w 40,7%. Efectul capacității combinative specifice h^2s (CCS) nu este semnificativ iar efectul reciproc al formelor parentale pentru acest caracter este egal cu 0.

Caracterul lățimea frunzei la soiurile parentale este mult influențat de condițiile mediului. Heritabilitatea în sens larg h^2w este inferioară 14,8%, iar în sens strict h^2e este egală cu 0, acțiunea neaditivă a genelor sau indicele CCS h^2s alcătuiește doar 12,01% iar efectul reciproc h^2re 2,79%.

Pentru lungimea frunzei, h^2w alcătuiește 40,7% iar h^2e care determină acțiunea aditivă a genelor spre formarea caracterului dat la hibridi este numai de 13,35%. Indicele CCS h^2s , de acțiune neaditivă a genelor, sa dovedit a fi semnificativ și superior lui h^2e alcătuind 20,95%, deci acțiunea neaditivă a genelor este superioară celei aditive. Efectul reciproc alcătuiește doar 4,2% fiind și nesemnificativ.

Raportul dintre lungimea frunzei și lățimea ei este influențat puternic de condițiile mediului extern. Indicele heritabilității în sens strict h^2e determinat de CCG alcătuiește doar 18,14%, practic fiind egal cu indicele heritabilității în sens larg h^2w 22,7%. Indicele CCS h^2s , de acțiune neaditivă a genelor, s-a dovedit a fi la fel neînsemnat 3,88% iar efectul reciproc al formelor parentale pentru acest caracter este egal cu 5,7%. Toți indicii obținuți sunt nesemnificativi.

Transmiterea prin ereditate a raportului dintre lățimea frunzei și lungimea ei are loc sub influența aditivă a genelor. Indicele heritabilității în sens strict h^2e determinat de CCG alcătuiește 54,2%, fiind aproape de indicele heritabilității în sens larg h^2w 61,0%. Indicele CCS h^2s , de acțiune neaditivă a genelor, sa dovedit a fi la fel neînsemnat 4,15% iar efectul reciproc al formelor parentale pentru acest caracter este egal cu 2,7%. Indicii h^2s și h^2re obținuți sunt nesemnificativi.

Efecte înalte ale CCG pentru caracterele suprafața frunzei, raportul dintre lățimea și lungimea frunzei și lungimea pețiolului au indicat soiurile de măr Florina, Remo și Coredana.

DETERMINATION OF SOYBEAN SEED OIL CONTENT IN VARIETIES ASSESSMENT

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Seed oil content is important in the study of the genetic diversity of soybean. The phenotypic value of oil content in seeds depends on the internal and external factors of plant development, ranging from 10 to 30%.

The use for biochemical purpose of the physical method, nuclear magnetic resonance (NMR) and its improvement allow us to do numerous rapid determinations of seed oil content during short period and significantly improve performance of analysis. The oil content as determined by NMR, corresponds to the amount of oil extracted from the seeds by organic solvents and the NMR method of determination of seed oil content is one of the international standards.

The object of research – the Moldavian varieties of soybean *Glycine max L.* The oil content in the seeds was determined by NMR relaxation method according to the international standard ISO/DIS 10565, Norm ISO 5725. Oil determination consist in comparing the NMR spin-echo amplitudes from the seeds and from the set of calibration standards. Standard selection defined by the natural biochemical composition of soybean oil, with the content of protons ~11,5%. A simple reference to the same content of protons is linoleic acid. In this work the reference samples were linoleic acid, soybean oil (refined or extracted from the seeds by toluene) and different batches of soybean oil in the defatted soy flour. Measurements were performed on a modernized RMN-relaxometer. Modernized NMR relaxometer, unlike the previous generation of NMR relaxometers, allows you to measure the maximum amplitude of the spin echo even in the case of spin echo NMR form distortion, with achieved S/N ratio of at least 32 dB and the operating temperature range to 10 ÷ 26 °C.

A significant factor for the seed oil content is the seed weight. Reduced oil content is typical for a start, and sometimes for the end of the vegetation period (the accumulation of oil in seeds is behind of the synthesis of other substances). For proper evaluation oil content should be assigned to a specific weight of the seeds. In addition, such parameter as the change in oil content per unit of seed mass change, contributes to a more differentiated characteristics of varieties.

On the basis of simultaneous measurements of the seed oil content and plant leaf surface in seeds is found that at optimum soil moisture soybean varieties with different potential productivity differ primarily in the amount of energy related to the oil (macroergic) component of grain yield per unit leaf area of plants. Although the bulk of the energy of seeds refers to proteins and this component of grain yield varies depending on growing conditions (especially in low-yielding phenotypes of plants grown under unfavourable conditions), but upon optimal growing conditions the soybean varieties different in their potential productivity did differ insignificantly in the amount of energy per unit of leaf area, refer to lowergic components of grain yield.

DESCRIEREA HIBRIZILOR-CLONE PERSPECTIVE DE LAVANDULA ANGUSTIFOLIA MILL.

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Scopul cercetărilor este evaluarea principalelor caractere biomorfologice cu impact direct asupra capacității productive a hibrizilor de levănțică, pentru evidențierea celor mai valoroase genotipuri. Acești hibridi pot fi utilizați direct la crearea de soiuri-clone noi prin multiplicarea vegetativă cu producție înaltă de materie primă, conținut sporit de ulei esențial și cu termeni diferiți de maturizare (timpurii, semitimpurii și tardivi). Acesta este și scopul cercetărilor preconizate. Din setul de hibridi evaluați pe parcursul anilor 2010-2012, unde ca martor au fost utilizate genotipurile de origine franceză (Fr.1) și (Fr.8), s-au evidențiat 7 hibridi perspectivi de levănțică, ce fac parte din diferite grupe de maturizare.

Din grupul hibrizilor timpurii fac parte: Fr.1-3-2V, Fr.1-3-23V, Fr.8-5-15V și Fr.8-5-23V, semitimpurii - Fr.1-3-9V și Fr.1-3-5V, Fr.8-5-34V tardivi. Acești hibridi sunt distinctivi după un sir de caractere cantitative valoroase. După cum știm, productivitatea la levănțică este influențată direct de caracterele cantitative precum, talia, diametrul plantei și numărul de tulpini florale per/plantă. Genotipurile hibride studiate sunt plante cu talie înaltă de la 52,8 cm (Fr.1-3-5V) până la 58,1 cm (Fr.1-3-9V), în comparație cu standardul, la care înălțimea plantelor este de 46,0 cm. Hibrizii ce provin de la forma maternă Fr. 8 sunt mai bine dezvoltati. Talia plantelor indică valori de până la 59,6-60,8 cm, cea ce este cu 9,4-10,6 cm mai înalți ca martorul.

Număr de tulpini florale la hibrizii studiați a constituit 606.3 unități la hibridul Fr.8-5-34V și 777.6 tulpini la Fr.8-5-23V. Forma standard (Fr.8) pe parcursul a trei ani de studiu a format în medie 465,6 lăstari florali cu 140,7 și 321,0 unități, corespunzător mai puține de cât la hibrizii menționați mai sus. La hibrizii policross F_1 de *Lavandula angustifolia* Fr.8-5-23V și Fr.8-5-15V cu un număr mare de tulpini florale, le corespunde și un diametru al plantei mai mare, 109,5 cm și 100,0 cm respectiv.

Important pentru soiurile de levănțică, pretabile pentru recoltarea mecanizată este lungimea tije florale. La acest caracter hibrizii Fr.8-5-23V, Fr.8-5-15V și Fr.8-5-34V indică valori de 24,2; 24,5 și 27,7 cm, s-au cu 7,7; 8,0 și 11,2 cm mai mari ca soiul martor. Tijele florale sunt groase și rezistă la cădere. Astfel de tije florale diminuează semnificativ acțiunea negativă a vânturilor puternice și micșorează distrugerea glandelor oleifere și respectiv, a pierderilor de ulei esențial. La trei genotipuri hibride cu forma maternă Fr. 1, tija florală are lungimea de 21,0-21,6 cm. Hibridul Fr.1-3-23V se evidențiază cu cea mai mare lungime a tije florale de 23,4 cm, sau cu 5,0 cm mai mare față de standardul Fr.1. Hibrizii studiați după caracterul "lungimea inflorescenței" au indici ce variază de la 27,5 până la 30,3 cm la cei descendenți de la forma maternă Fr. 1, iar la hibrizii descendenți de la forma maternă Fr. 8, lungimea inflorescenței este mai mare și constituie 34,7 cm. De menționat, că la plantele cu talie înaltă și inflorescențele sunt bine dezvoltate. De exemplu, la hibrizii Fr.8-5-15V, Fr.8-5-23V și Fr.8-5-34V cu talia plantelor de 59,6- 60,8 cm, lungimea inflorescențelor este de 31,7; 32,2; și 34,7 cm. Respectiv la hibridul Fr.1-3-5V cu cea mai joasă talie (52,8 cm) acest caracter este de numai 27,3 cm. Toti acești hibridi s-au evidențiat cu cele mai lungi spice florale de 7,2 cm la hibridul policross Fr.8-5-15V; 7,4 cm la Fr.8-5-34V și 8,2 cm la Fr.8-5-23V. La standard Fr.8 lungimea spicului floral este mai mică și constituie numai 6,1 cm.

Numărul de verticile în spic la hibrizii menționați au valori de 6,7; 6,8 și 7,0 unități. Din cele expuse rezultă, că hibrizii policross F_1 de *Lavandula angustifolia* Mill. evaluați ce provin de la forma franceză Fr. 8 (Fr.8-5-15V, Fr.8-5-34V și Fr.8-5-23V) după caracterele cantitative, sunt de perspectivă.

Conținutul de ulei esențial este o însușire la fel de valoroasă pentru soiurile-clone de perspectivă precum și numărul de inflorescențe. Sub acest aspect conținutul de ulei esențial pe parcursul anilor de cercetare la hibrizii evaluați este mai înalt de cât la forma maternă. Cercetările efectuate ne-au permis evidențierea celor mai performanți hibridi pe parcursul a trei ani de cercetări la conținutul de ulei esențial Fr.1-3-23V și Fr.1-3-9V, descendenți de la forma maternă Fr. 1, cu un conținut de ulei esențial 4,744-4,969 % (s.u.) și hibrizii Fr.8-5-34V și Fr.8-5-23V ce provin de la forma maternă Fr. 8, la care conținutul de ulei esențial este de până la 4.844%. Conținutul de ulei esențial la formele materne (Fr.1 și Fr.8) în medie este de 3,544 și 2,916% (s.u) respectiv.

Generalizând rezultatele obținute în acești ani la hibrizii menționați, putem confirma, că materialul evaluat ne permite crearea soiurilor-clone noi de levănțică cu productivitate sporită.

ACHIEVEMENTS AND REPRODUCTIVE CHARACTERISTICS OF SEED CROPS ELITE LEGUMES

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Progress in cultivation is directly related to the reproduction and use of seed varieties for sowing performance. The use of improved seeds in agriculture valuable biological characters increase grain yield per unit of area of 40-70%. The ongoing process of seed production provides the morphological and biological characteristics of the variety, quality and quantity production potential.

Creation, approval and production of basic seed stems for 8-10 years and is determined as the primary selection.

In the laboratory of Genetics and Plant Breeding Institute of Genetics Pulses Academy of Sciences (now the Institute of Genetics, Physiology and Plant Protection) were created in recent years and appreciated the State Commission for Variety Testing of RM 28 The leguminous crop varieties, of which 17 (soy - 6 chickpeas - 3 lentils - 2, beans - 2 peanuts - 2 chickling - 1 and grain - 1) were approved, patented and created basic seed .

The next stage is the scientific elite reproduction seeds of the variety newly created or the old one for implementation in production and should be carried out continuously by a team specialized in seed production, after multiplication scheme accepted for preliminary relief and arising for 7 - 8 years in institutions for creating seed variety or households.

Continuous production of elite seeds is necessary because the varieties within 5 - 6 years changes its genetic structure, lose their authenticity and production capacity after the intervention of natural selection, mutation, genetic drift, gene migration, and under the influence of mechanical impurities different technological processes in culture.

Reproduction and seed multiplication was initiated in 2011 to obtain new seed varieties elite chickpea and lentil Golden Ikel scheme supported by leguminous plants, including the following links: field sown with seed selection typical of the variety obtained the calibration process is selected seeds and plants (lines) most vigorous; selection field with grain sowing 8 cm distance between seeds in a row and between rows with 45 cm distance and harvesting individual progenies of elite lines I and -II eliminating less productive lines; Basic field of elite super valuable in the seed lines join the second year and third-year mechanical sown with row spacing of 45 cm, also the maintenance is performed to yield super elite seeds; propagating field is seeded first year 2015 with seed progenies most valuable elite of super elite field.

During reproduction seed all links elite seed breeding scheme were made phenological observations on plant growth and development, resistance to diseases and pests appreciated, selected elite progeny plant morphology after flowering and maturation during the nonspecific discarded pods and variety, the weak and sick.

In laboratory conditions were analyzed and found to yield structure elements consist of the number of pods with beans from a plant, number of grains of a pod and absolute weight of the grains. They also appreciated the biological characteristics of seeds (germination energy and germination).

PLUM HYBRIDS SUSCEPTIBILITY TO SHARKA IN THE CONDITIONS OF MOLDOVA .

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The main direction in breeding plum in Moldova is the selection of different varieties ripening, possessing a complex of economically valuable traits, increased resistance to adverse environmental factors especially dangerous disease Plum Pox Virus.

During five years we have created a gene pool of 4,500 seedlings of 54 crossing combinations. Thirty-five perspective hybrids were selected for further study and breeding Parental forms were: President, Stanley, Kirke, Ashatan, Renclod Hramovix, hybrid 3-4-35, hybrid 306420, Renclod Althan, Sopernita. Studies conducted in the plum garden, planting 2010-2011. Planting scheme 5 by 3 meters. Cultivars have been grafted on seedlings cherry plums. Natural infection background was about 22 percent. Evaluation susceptibility to Plum Pox Virus carried by the symptoms on the leaves and on the fruits during May and June, followed by testing Elisa or ISEM.

As a result of preliminary studies during the 2012-2014 period , we evaluated 9 hybrids susceptibility to the virus. The main strain that circulated in Moldova is a strain D(Dideron). Four hybrid available in the form of maternal Stanley showed high susceptibility to PPV of 65% of the hybrid 1-4-1 (Stanley x hybrid 306420) ,71% in the hybrid 2-4-91 (Renclod Hramovix x Stanley), 83% of the hybrid 1 -4-48 (hybrid 306420 x Stanley) to 100% in the hybrid 2-4-123(Renclod Hramovix x Stanley) In a study of 75trees of hybrid 328 (President x Sopernita), landing in 2011 only 6.7% showed symptoms on the leaves. Earlier experiments in 2004-2006 at the same hybrid in another garden, the percentage of trees with simtomy of the virus was 22%.

The studies selected two hybrids that showed resistance to plum pox virus, the parent pair was sort of Kirke. This III-33-75 (Renclod Althan x Kirke) dark purple, round fruit weighing up to 50 grams. Ripening time the second decade of August. The second hybrid 1-15-53 (Sopernita x Kirke), mature in early September, with the weight of the fruit to 40 grams. Investigations are continuing with the use of modern methods of diagnosis.

THE REACTION OF THE GENOTYPES OF WINTER BARLEY ON THE GROWING CONDITIONS

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Given the climatic changes at the present stage of plant breeding, it is important to create the varieties not only with the maximal yield potential, but also with high resistance to adverse environmental conditions. The indicators of the adaptive possibilities of the genotypes are characterized mathematically by the notions of plasticity and stability of varieties.

If the bi index value is less than 1, then the variety doesn't possess plasticity. If the bi index is equal to 1, then the variety is considered plastic, and if the bi index is higher than 1, then this variety has high plasticity. In case that the value of Si^2 is equal to 0, the variety is stable, and if Si^2 value is higher than 0, then the variety is considered unstable. The priority should be given to high plasticity and stability of varieties, that acquire the high yield in good growing conditions and insignificantly reduce the production, when they worsen.

At the moment, in the Republic of Moldova, there are 9 registered winter barley varieties, created in the Public Institution the Research Institute of Field Crops: Moldavskii 18, Tighina, Ciuluc, Speranța, Excelent, BȚ 14/02, Strălucitor, Scînteia, Tesaur. These varieties have the productivity potential between 7,0 and 8,0 t/ha. The average yield of the last 7 years (2007-2013) was between 3,94 t/ha and 4,71 t/ha. These varieties of barley can be divided into two groups: the typical winter varieties (the first 5 cultivars) and the universal or semiwinter varieties (the last 4 cultivars). The varieties differ significantly not only by the type of development, but also by the quantitative characters (height of plants, vegetation period, resistance to lodging, frost, diseases and drought, weight of 1000 seeds and others.). In this regard they respond differently to growing conditions, because that they have different levels of stability and plasticity (table 1).

Table1. The characteristics of the varieties of winter barley, registered in 2007-2013.

Varieties	The yield of grains, t/ha			The indicators	
	average	max	min	bi	Si ²
Moldavskii 18	4,20	5,09	2,25	0,90	0,13
Tighina	3,94	4,99	2,38	0,84	0,09
Ciuluc	4,09	4,92	2,57	0,81	0,09
Excelent	4,71	6,97	2,35	1,36	0,09
Speranța	4,32	5,23	2,55	0,86	0,05
BȚ 14/02	4,50	6,35	2,65	1,09	0,09
Strălucitor	4,34	5,99	2,57	1,03	0,07
Scînteia	4,59	6,50	2,96	1,06	0,06
Tesaur	4,34	5,48	2,35	1,06	0,09

The evaluation study of the winter barley varieties proved that the cultivars Moldavskii 18, Tighina, Ciuluc and Speranța are better adapted to the middle and the worst environmental conditions ($bi < 1$). The varieties BȚ 14/02, Scînteia, Tesaur and Strălucitor are the most responsive to good environmental conditions. The variety Excelent has the highest reaction on the favorable environment. Speranța has proved the highest stability among the winter barley varieties, and the variety Scînteia – among the semiwinter varieties.

The variety Scînteia, which was registered in the Republic of Moldova in 2012, has high plasticity and good yield stability. This variety is of middle height and proves good resistance to lodging, drought and diseases. It is recommended for cultivation on poor agricultural background. The average content of crude protein is 13,9 %. Due to these properties it acquires a large spread in the agricultural production of the Republic of Moldova.

LAVANDULA ANGUSTIFOLIA L. – ASPECTS THE *IN VITRO* REGENERATION

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L. angustifolia L. a medicinal and aromatic plant, known since antiquity with its origin in Mediterranean Basin. Of great interest is the volatile oil extracted from fresh vegetal material from inflorescences in the flowering phase, which is used in food industry, perfumery, cosmetics.

We have set ourselves to settle the micro-cloning and micro-propagation of this plant of great importance for national economy, which is cultured *in vitro*. In this work we will present a synthesis of results obtained during the study of micro-propagation for three lavender cultivars in Botanic Garden (I.) of ASM with a high productivity of vegetative mass, especially of inflorescences and as a result with the volatile oil of good quality and oil component of selected refinement. The entire aroma range of volatile oil is determined by organic and inorganic compounds. In order to maintain these qualities for regenerative plants is recommended the vegetative way of multiplication. By generative way there are obtained seed trees with a large spectrum of descendants, the deviation peculiarities came from initial plants, donor plants. Traditional way by tillering is limited by the number of twigs of mother plant, the rooting of cuttings in rhizogenes is of 50-70% at best, and the obtained cuttings are kept in nurseries with special care conditions for a long period (14-15 months) till the maturation, (M. Buiuclic, 1969, M. Gonciaruc, 2008).

The integral aerial part of the plant contains volatile oil; the maximum amount of oil is stored in inflorescences during the flowering period. There are made many investigations regarding the composition, quality and quantity of organic and inorganic substances of this plant and are written many literary works about this plant due to its cultivation in many countries. In the Republic of Moldova, due to favorable climate and soils was and is developed the branch of oleaginous plants, especially the cultivation of *Lavandula angustifolia* L. In Moldova these species were studied by M. Buiuclic (1996), M. Gonciaruc (2008, 2006, 2005, 2004, and 2000) and they have created volatile oil sorts with high productivity. In the Botanic Garden (I.) of ASM over the years the researchers were concerned about the mobilization and introduction of new species, varieties, cultivars and elaboration of regeneration and multiplication methods of these taxa. (M. Bodrug, A. Ciubotaru, M. Coltun, L. Chisniceanu, N. Ciorchina). During these studies we have tested the possibilities of obtaining the propagating material for species and sorts of lavender, which are regenerated by traditional methods and obtained from vitro-cultures. Vegetal biotechnologies represent one of the main achievements of science and technique of 20th century, with a great role in development of modern agriculture and horticulture. *In vitro* micro-propagation is the branch of vegetal biotechnology that represents an ensemble of multiplication methods of plants by using *in vitro* cell, tissue cultures and vegetal bodies. This technique allows the considerable increasing of yield by multiplication of different species, being at the same time a release method of pathogens of propagating material.

Applying this method to this culture we have settle the key moments, establishing the sterilizing reagents, exposure time for sterilization of vegetal material, size, type and phase of excising the explant. There were established main media MS-100% and 50%, there were selected the compounds of 7 solid and liquid types of media, with a supplement of hormonal balance of auxins and citochinine, used for processes of inoculation, development, growth and maintenance, callogenesis, rhizogenesis. Analyzing the results obtained during the periodical observations there were stated the following obvious aspects: inocula built from apexes are generating for all kinds of varieties seedlings in respective media. With other words there is being caused the callogenesis for a period of 15-30 days from merismatic apexes with one phytohormone ANA that causes a massive growth and strong multiplication at inoculated taxa. Other tested explants – inocula, like knots, inter-knots, inflorescences in different development phases in media with supplement of BAP, 2, 4-D are generating callus mass. Initially we have set ourselves to select the composition of a profitable and efficient media for a great micropropagation of those taxa in order to obtain a propagating material for industrial plantations. The first obtained results have shown that micro-propagation for lavender and especially for three taxa is possible by means of technique of *in vitro* cultures, where the explants are built from apexes, knots and floral buds. Callogenesis for all these three varieties is a frequent process; the regenerative capacity of callus mass is directly proportional to concentration of BAP from media. We have obtained a lot of plants received by *in vitro* culture, adapted and acclimatized to ex vitro conditions and planted in open-air run. The volatile oil will be exposed to a biochemical screening to compare the composition, quantity and quality of the oil these plants with the oil of propagated plants in traditional ways.

MOLECULAR AND GENETIC ASPECTS OF *CICER ARIETINUM* L. x *FUSARIUM OXYSPORUM* SCHLECHT. EMND SNYD. AND HANS. F. SP. *CICERIS* INTERACTION

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Poor health of the population and increasing incidence of cancer diseases dictates the need to implement the ecologic agriculture, based largely on the use of varieties with genetic resistance. Law on ecologic food production provides for a balanced and sustainable agricultural system – strategic area for food security and safety. In this context, leguminous crops have an important role, being considerate as fertilizers sideral for soil fertilization, good preceding in crop rotation and the remedy in cases of exposure to ionizing radiation.

The large number of phytopathogenic agents, their high toxicity, and absent of reliable genetic sources of resistance remain fairly current for agricultural crops. Using genetic resistance is the most economical and advantageous environmentally method to reduce losses caused by the attack of pathogens. Resistance gene activity and its heritability depend, in the most direct way, by host-parasite interactions, climate conditions and genotype ontogenetic stage.

Diminished *Cicer arietinum* L. genofod variability presents a real danger for the diseases expansion. A chickpea is attacked by about 50 pathogens more susceptible being to the fungus *Fusarium oxysporum* Schlecht emnd Snyder. and Hans. f. sp. *ciceris*. Under favorable conditions for the development of the pathogen (low temperatures and high humidity) epidemics caused by *F. oxysporum* f. sp. *ciceris* contribute to the total loss of the crop.

The purpose of the research and objectives: elucidating the molecular-genetic basis of *C. arietinum* relations with pathogens and resistance's control in chickpea protection; to determine the composition of the fungi species that attack chickpea and their pathogenicity; to characterize genetic-molecular features of chickpea genotypes based on enzymatic factors, SSR-markers and plant response to *F. oxysporum* f. sp. *ciceris*; germplasm evaluation and development of valuable chickpea genotypes resistant to pathogens and unfavorable abiotic factors possessing high indices of productivity; to determine the role of hereditary and acquired resistance in the integrated chickpea protection.

Polymorphism of SSR markers has been established. As a result of molecular analysis using SSR markers 96 amplicons have been obtained, which were analyzed based on molecular and genetics parameters of chickpea genotypes. H1G16 primer pair generated three amplicons: 265, 290 and 310 bp, only one (265 bp) was previously reported. An average number of 4,36 alleles per 1 SSR marker have been elucidated, showing 77,3% polymorphism. Polymorphic indices (PIC) average was 0,55, ranging from 0,14 (H6D02, H5D02) to 0,90 (H1K18). High polymorphism was manifested to genotypes: H1K18 (0,90); H6B11 (0,81); H2J04 (0,75); H1O10 (0,68); H1O01 (0,63); H4G08 (0,62); H5E05 (0,66); H1P17 (0,67). A high degree of similarity has been found for chickpea plant response to *F. oxysporum* f. sp. *ciceris* and polymorphism of SSR markers from different linkage groups, which indicates a polygenic control of the reaction and the opportunity to use molecular markers to identify resistant genotypes.

Strategies of genetic resistance control employment in crop protection from phytopathogens have been elaborated. An important role of genotype in manifestation of *C. arietinum* resistance to *F. oxysporum* f. sp. *ciceris* has been defined.

Polymorphism analysis of chickpeas peroxidases (POX) reveals evidence of Rf 0.3 and 0.9 polypeptides, their activity being higher in resistant varieties, especially under the action of the pathogen, which shows POX involvement in the formation mechanisms of chickpeas resistance to *F. oxysporum* f. sp. *ciceris*. Increase in the oxidation-reduction reactions involved in detoxification of mycotoxins and fungal cell walls degradation has been shown to be one of the mechanisms ensuring *C. arietinum* resistance to *F. oxysporum* f. sp. *ciceris*.

As the result valuable solutions have been proposed to reduce phytosanitary problems. *F. oxysporum* f. sp. *ciceris* strains with enhanced virulence useful for plant x pathogen interaction research and development of an artificial background for resistance tests have been identified. Three chickpea varieties have been developed; two varieties of chickpea have been approved for the Moldova conditions and included in the database of the FAO UNO and N.I. Vavilov Research Institute of Plant Industry, St. Petersburg, Russia.

SUPORTUL INFORMATIC ÎN MANAGEMENTUL GENOFONDULUI VIȚEI DE VIE AL ÎSPHTA

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Procesul complex de gestiune a Fondului genetic al viței de vie include un șir de activități, dintre care menționăm: colectarea, preservarea, descrierea, evaluarea, utilizarea și distribuirea genotipurilor. Fiecare dintre aceste activități, la rândul său, poate include propriul protocol de management. În acest context este necesară o evidență eficientă a tuturor aspectelor ce țin de fluxul de date și operații în banca de gene și care poate fi asigurată prin utilizarea sistemelor informatice.

Fondul genetic al viței de vie acumulat la Institutul Științifico-Practic de Horticultură și Tehnologii Alimentare include cca. 3000 genotipuri provenite din majoritatea centrelor viticole ale lumii, resurse genetice obținute în procesul de ameliorare a sortimentului. Monitorizarea și explorarea acestor resurse implică manipularea unui volum foarte vast și complex de informație, atât după natura sa (numerică, textuală, imagini foto), cât și după frecvența completării și actualizării.

În scopul asigurării unui instrument eficient în managementului acestor resurse s-a elaborat suportul informatic, unde în calitate de mediu de dezvoltare este utilizat Sistemul de Gestiune a Bazelor de Date Visual FoxPro 9.0. Structura Bazei de Date și aspectele funcționale au derivat din metodele de gestiune și documentare acceptate: Regulamentul Fondului genetic al viței de vie, Ghidul pentru fondarea și managementul Băncilor de gene, Lista de descriptori elaborată de Oficiul Internațional al Viei și Vinului (O.I.V.), Datele de pașaport conform listei de descriptori multi-cultură (MCPD - Multi Crop Passport Data).

Fiecare intrare în Genofond este identificată de un număr unic de acces, care conține, în calitate de parte componentă obligatorie, codul FAO atribuit institutului (MDA004). Astfel, accesile sunt identificate în mod univoc atât la nivel local, cât și la nivel internațional. În funcție de numărul de surse de material biologic inițial, pentru fiecare genotip în parte pot fi prezente mai multe accesii în cadrul Fondului genetic. Capacitățile funcționale elaborate asigură evidența „istoriei” fiecărui acces: sursa materialului inițial, data de intrare, regenerările în cadrul Genofondului, evoluția stării biologice a butucilor, difuzarea către alți beneficiari etc. Pentru fiecare genotip sunt acumulate descrierile ampelografice, datele observațiilor multianuale, provenite din diverse surse (desfășurarea fazelor fenologice, indicii de fertilitate și productivitate, alți parametri agrobiologici), asigurându-se completarea Fișelor de cercetare și a Cartelei ampelografice. Este asigurată compatibilitatea între metoda actuală de documentare a resurselor genetice, acceptată la nivel internațional (Descriptorii O.I.V.), cu metodele aplicate anterior (Constantinescu, Lazarevski, Ivanova). Totodată, numărul acestor metode poate fi lărgit, asigurându-se coordonarea între gradațiile fiecărui caracter și a celui de referință.

Informația acumulată poate fi prezentată și livrată, în format necesar, către diverse destinații, inclusiv este pregătit setul de bază pentru transmitere în Baza de date *Vitis* europeană.

Pentru a eficientiza procesul de acumulare a informației, în special reducerea manipuleărilor și operațiilor manuale voluminoase ce țin de colectarea, prepararea și păstrarea ierbarilor, efectuarea ulterioară a măsurărilor ampelometrice cu rigla și raportorul, s-a realizat posibilitatea măsurării parametrilor morfologici ai frunzei mature în baza imaginilor digitale, păstrate în baza de date. Același procedeu este utilizat și la efectuarea măsurărilor, în baza imaginii digitale, a parametrilor ce caracterizează sămânța viței de vie.

Interfața realizată în formă de meniu principal și submeniuri de diferit nivel, formularele pentru introducerea și afișarea informației, rapoartele predefinite, permit diverselor categorii de utilizatorilor, familiari deja cu alte aplicații informatice, să exploateze eficient sistemul informatic.

VARIABILITATEA UNOR CARACTERE CANTITATIVE LA HIBRIZI DE SALVIA SCLAREA L., ÎN PRIMUL AN DE VEGETAȚIE

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Până nu demult, soiurile cultivate de *Salvia sclarea* L., înfloreau numai în anul al doilea de vegetație. Grație cercetărilor efectuate în laboratorul Plante Aromatice și Medicinale al Institutului de Genetică, Fiziologie și Protecție a Plantelor, au fost creați hibrizi și soiuri de șerlai, care înflorească, formează producție de materie primă în anul întâi, al doilea și al treilea de vegetație. Pentru crearea hibrizilor de *Salvia sclarea* cu productivitate înaltă, cu perioade de vegetație diferite, rezistenți la ger, iernare și secetă, a fost utilizat material inițial de ameliorare cu o variabilitate pronunțată la caracterele cantitative ce influențează direct productivitatea. Hibrizii creați sunt rezistenți la ger, iernare și secetă, formează inflorescențe lungi, compacte cu număr mare de ramificații, sintetizează și acumulează ulei esențial în concentrații ridicate. Obiect de studiu au servit 16 hibrizi de diferită complexitate, de *Salvia sclarea* L., în primul an de vegetație, la care a fost evaluată variabilitatea, exprimată prin coeficientul de variabilitate (V).

La caracterul "talie plantelor" acest indice a variat de la 97,2 la hibridul în trepte [(K-36 x 0-41)F₂ x 0-19)F₁ x 0-22)B₄ x L-15)F₈ x (M-44S₄ x L-15)F₁ x L-15)B₆]F₃ până la 106,9 cm, la hibridul complex [(S.s.Turcmenia/N)S₇ alb x (Rubin x S1122 9S₃)F₁ x (0-56 x V-24)F₇)F₇]F₄. La 13 din hibrizii studiați, inclusiv un hibrid triliniar, un hibrid dublu și 11 hibrizi în trepte și complecși, variabilitatea la acest caracter este joasă, fiind mai mică de 10% (V% < 10). Cel mai mic coeficient de variație (V% - 3.1) s-a înregistrat la hibridul complex [(S.s.Turcmenia/N)S₇ alb x (Rubin x S1122 9S₃) F₁ x (0-56 x V-24) F₁) F₇]F₄ și tot acest hibrid a avut cele mai înalte plante - 106,9 cm. Pentru hibrizii testați observăm, că acest caracter este constant.

Lungimea inflorescenței la hibrizii evaluați în primul an de vegetație a fost destul de mare și a constituit de la 50,0 până 70,6 cm. Cel mai viguros panicul (70,6 cm lungime) au format plantele hibridului complex [(S-1122 528 S₃ x (Rubin x S-786)F₁ x (0-33 S₃ x L-15)F₇ x M-69 655 S₉)F₃. Cota parte a inflorescenței din talia plantei la hibrizii evaluați a fost mai mare de 50%, la șase din ei acest indice a fost mai mare de 60%, iar la doi din hibrizii studiați acest raport a constituit 67,8%. Lungimea inflorescenței, caracter de care depinde randamentul hibridului, variază neînsemnat la 13 din ei (V=4,2 - 9,1%). Un hibrid în trepte și doi hibrizi complecși au avut valori medii (V%=14,3 și 14,7%).

Inflorescențele lungi, compacte, puternic ramificate sunt caractere importante pentru șerlai. Numărul de ramificații de gradul întâi al inflorescenței, la hibrizii studiați a fost destul de mare și a variat de la 10,8 la hibridul în trepte [(M-69 655 S₉ x (S-1122 528 S₃ x (Rubin x S-786) F₁ x (0-33 S₃ x L-15)F₇) F₃ până la 17,2 unități la hibridul complex [(K-36 x 0-41) F₂ x 0-19) F₁ x 0-22) B₄ x L-15) F₈ x (M-44S₄ x L-15) F₁ x L-15)B₆] F₃. Variabilitatea ramificațiilor de gradul întâi la zece din hibrizii evaluați este joasă (V% < 10), iar la un hibrid în trepte variabilitatea este relativ înaltă (20,9%).

Este necesar de remarcat faptul, că caracterele cantitative la *Salvia sclarea*, cât și la alte plante aromatice și medicinale depind în mare măsură de condițiile climatice ale anului și se caracterizează printr-o anumită variabilitate genotipică și fenotipică pentru fiecare caracter în parte. Cele mai evidente diferențe ale coeficientului de variație s-au înregistrat la caracterul "numărul de ramificații" de gradul al doilea al inflorescenței. Coeficientul de variație la acest caracter este mult mai mare față de celelalte caractere studiate. Astfel, la hibridul triplu [(V-24-86 809 S₃ x 0-33 S₉)F₇ x Cr.p. 160 S₁₁]F₂ - V%=34%, la hibridul dublu [(V-24-86 809 S₃ x 0-33 S₉)F₇ x (S-1122 9 S₃ x K-17)F₉)F₂ - V%=22%, iar la hibrizii în trepte și complecși a variat în limitele 7-26%. Coeficientul de variație a numărului de verticile pe spicul central al paniculului demonstrează aceeași regularitate. Astfel, la hibridul triplu V%=5,4%, dublu - V%=6,1% iar la hibrizii în trepte și complecși este în medie - 9%.

Valoarea indicilor conținutului de ulei esențial în inflorescențe, recalculat la substanță uscată, la hibrizii evaluați, a demonstrat, că majoritatea au acumulat ulei esențial în concentrații ridicate. Cel mai înalt conținut de ulei esențial a înregistrat hibridul în trepte [(M-44S₄ x L-15)F₁ x L-15) B₅ x (M-44S₄ x L-15)F₁ x L-15) B₆]F₃ - 1,845 % (s.u.).

În rezultatul cercetărilor efectuate, putem concluda, că prin hibridări se poate crea un material inițial de ameliorare valoros și important pentru crearea soiurilor perspective. Majoritatea caracterelor la hibrizii studiați sunt destul de constante, deoarece coeficientul de variație este unul relativ mic și variază în limitele 3,1-19,0%.

VARIABILITATEA PARAMETRILOR GENETICI PENTRU REZISTENȚA LA FĂINARE ÎN HIBRIDĂRILE DE DIALELEA ÎN FUNCȚIE DE ANI

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Făinarea, *Podosphaera leucotriha* Salm, este o maladie periculoasă pentru soiurile de măr aducând mari pagube recoltelor de fructe mai ales în regiunile sudice inclusiv în Republica Moldova. Impactul economic poate constitui o pierdere în recoltă de 40-50% și mai mult. În anii epifitotici aparatul foliar fiind puternic atacat fructele pierd mult în calitate. Deacea cunoașterea eredității și variabilității parametrilor genetici în funcție de condițiile anilor poate favoriza alegerea corectă a soiurilor parentale. Ca material pentru cercetare au servit 2400 hibridi de măr obținuți de la încrucișarea soiurilor imune la rapănul mărlului în baza genei Vf: - Florina, Priam, Coremodet, Coredar, Remo, Coredana. Hibridările au fost efectuate conform metodei 3 B. Griffing în anul 2002. Semănatul semințelor hibride a fost efectuat în toamna anului 2002. În combinațiile hibride nu s-a efectuat selecție preventivă. Livada de selecție a fost plantată în primăvara anului 2005, în 4 repetări, cu plante hibride de doi ani. Distanța de plantare 4 x 0,75m. Observațiile privind atacul de către făinare au fost efectuate în anii 2007, 2010, 2013 prin notare de la 1 la 6 unde nota 1- atacul lipsește iar nota 6 atac foarte puternic. Datele obținute ne arată că indicii parametrilor genetici ai soiurilor parentale se comportă diferit în funcție de ani (tab. tab. 1, 2).

Tabelul 1. Variabilitatea indicilor heritabilității în funcție de ani

Anul	Heritabilitatea în sens larg h^2_{vs} , %	Heritabilitatea în sens strict h^2_{es} , %	Cota de acțiune neaditivă h^2_{ns} , %	Efectul reciproc h^2_{rc} , %	Erori σ^2_e , %
2007	80,2	39,8	35,6	4,6	20,0
2010	91,3	59,4	27,6	5,4	7,7
2013	95,7	62,4	28,6	6,1	2,9

Tabelul 2. Variabilitatea efectelor CCG (\hat{g}_i) și varianțelor CCG ($\sigma^2_{\hat{g}_i}$) și CCS ($\sigma^2_{\hat{s}_i}$) în funcție de ani

Soiul	Efectele CCG (\hat{g}_i)			Varianțele CCG ($\sigma^2_{\hat{g}_i}$)			Varianțele CCS ($\sigma^2_{\hat{s}_i}$)		
	2007	2010	2013	2007	2010	2013	2007	2010	2013
Florina	0,3218	-0,0545	-0,0967	0,1020	0,0022	0,0094	0,0939	0,0651	0,0724
Priam	0,0264	0,1943	0,2118	0,0008	0,0369	0,0449	0,0167	0,0645	0,0695
Coremodet	-0,1485	-0,3232	-0,2496	0,0205	0,1036	0,0623	0,0344	0,0849	0,0212
Coredar	-0,1888	-0,1013	-0,1975	0,0341	0,0095	0,0390	0,0598	0,0715	0,0639
Remo	-0,3717	-0,3801	-0,2965	0,1366	0,1437	0,0879	0,0592	0,1015	0,1097
Coredana	0,3608	0,6649	0,6286	0,1287	0,4413	0,3950	0,0615	0,0392	0,0834
Greșala standard	$\pm 0,0605$	$\pm 0,0439$	$\pm 0,0523$	$\pm 0,0605$	$\pm 0,0439$	$\pm 0,0523$	$\pm 0,0600$	$\pm 0,0761$	$\pm 0,0694$

Astfel heritabilitatea în sens larg h^2_{vs} , practic nu variază. Genotipurile soiurilor cercetate sunt foarte variate după caracterul dat și sunt influențate slab de condițiile mediului. Deci ereditatea rezistenței la făinare depinde de genotipurile soiurilor parentale cercetate. Variabilă este acțiunea aditivă a genelor, determinată de heritabilitatea în sens strict h^2_{es} , de la 39,8 până la 62,4%. Cota de acțiune neaditivă h^2_{ns} , practic nu variază. Cu vîrsta pomilor acțiunea aditivă a genelor la soiurile parentale crește.

Efectele capacității combinative generale CCG (\hat{g}_i) la soiurile parentale variază în funcție de ani dar în general denotă tendința de a păstra însemnătatea acțiunii genelor la soiurile cercetate. La soiurile Priam și Coredana efectele sunt stabil pozitive și practic semnificative. În combinațiile lor hibride pot fi anual multe plante atacate puternic de făinare. La soiurile Coremodet, Coredar și Remo efectele CCG sunt semnificative și în funcție de ani stabil negative. În combinațiile lor hibride pot fi anual mai puține plante atacate de făinare. Varianțele CCG ($\sigma^2_{\hat{g}_i}$) ale soiurilor cercetate, practic variază în conformitate cu variabilitatea efectelor CCG (\hat{g}_i). Varianțele capacității combinative specifice CCS ($\sigma^2_{\hat{s}_i}$) variază atât în funcție de soi cît și în funcție de ani. Din raportul dintre varianțele CCG ($\sigma^2_{\hat{g}_i}$) și varianțele CCS ($\sigma^2_{\hat{s}_i}$) rezultă că soiul Priam posedă acțiune neaditivă spre formarea mai multor hibridi puternic atacați iar Coredana stabil posedă acțiune aditivă spre formarea de hibridi puternic atacați. La soiurile Florina, Coremodet și Coredar acțiunea aditivă și neaditivă variază diferit în funcție de ani. Raportul dintre varianțele CCG ($\sigma^2_{\hat{g}_i}$) și varianțele CCS ($\sigma^2_{\hat{s}_i}$) indică variabilitate a acțiunii aditive sau neaditive ale genelor spre formarea la hibridi a caracterului cercetat acțiuni, care probabil depind de interacțiunea genelor soiurilor parentale cu condițiile mediului ce se instalează în fiecare an pentru dezvoltarea ciupercilor raselor de făinare.

MOLECULAR SCREENING OF LOCAL SUNFLOWER GERmplASM FOR DOWNY MILDEW AND RUST RESISTANCE

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Study of sunflower plant resistance to various diseases have a great importance for improvement of commercial hybrids. The majority of diseases of this oilseed crop are caused by fungi. The most frequent pathogens of sunflower are: *Plasmopara halstedii* (downy mildew), *Puccinia helianthi* (rust), *Verticillium dahliae* (verticillium wilt), *Phoma macdonaldii* (black stem), *Sclerotinia sclerotiorum* (stem rot), *Diaporthe helianthi* (stem canker), *Alternaria helianthi* (blight), *Erysiphe cichoracearum* (powdery mildew) etc.

In our country fungal diseases are frequently encountered on sunflower cultivated surfaces in wet years. The parasitic fungi have a negative impact on the quantity and quality of yield. Selection and breeding of crops aided molecular markers associated with resistance genes enables to select valuable genotypes to produce hybrids in short time and with high precision. For *Pl* genes that ensure resistance to downy mildew and *R* genes for rust resistance in sunflower were proposed various molecular markers (SSR, RAPD, CAPS, SCAR etc.). Aim of the study was to evaluate the potential for resistance to fungal pathogens on local genotypes and hybrids to effectively control the epidemic spread of these phytopathogens in sunflower fields.

Genetic material used for study was offered from the breeding collection of company "AMG-Agroselect", Soroca. The set contained 42 sunflower genotypes: 12 lines with cytoplasmic male sterility (CMS), 22 *Rf* (restore of fertility) lines and 8 commercial *F*₁ hybrids. Sunflower plants were grown in the laboratory until the two cotyledon leaf stage. DNA extraction was performed using fresh vegetative material with GeneJET Plant Genomic DNA Purification Mini Kit (*Thermo Scientific*). Molecular screening of *R₁* and *Pl₁* gene was realized with molecular SCAR and CAPS markers respectively previously reported in literature. Assessment of *Pl₆* and *Pl₅/Pl₈* locus was performed by PCR with specific primers (STS markers). All the amplicons and the digestion fragments were visualized in agarose and polyacriamide gels (CAPS) of various concentrations.

The molecular screening of *Pl₁* gene within showed the presence of 363 bp amplicon after PCR, which generated after digestion with *Tas509I* four DNA fragments, thus indicating resistance of all 42 studied genotypes to race 100 of downy mildew. Identification of *Pl₆* gene using HAP3 primer showed the presence of fragments associated with resistance in only 22 analyzed genotypes. Identification of genotypes with resistance to downy mildew caused by the presence of *Pl₅/Pl₈* locus was revealed seven *Rf* lines and two CMS lines containing markers associated with these resistance gene. Amplicons, indicating presence of *R₁* rust resistance gene were revealed in only 10 paternal lines and three hybrids.

Apparition of infection with downy midley and rust it is typical for Northern part of the Republic of Moldova (Soroca). Evaluation of the downy mildew and rust resistance potential on sunflower germplasm using molecular markers demonstrated the presence of all four investigated genes in only one *Rf* line – MS-2540C, which can be useful for various breeding programs in quality of the original resistance source. Obtained data could be applied at the initial stage of parental lines selection for production of elite sunflower germplasm, allowing the application of gene pyramiding to obtain hybrids resistant to fungal diseases.

**DIFFERENTIAL EXPRESSION OF ROS-SCAVENGING GENES IN SUNFLOWER
INFECTED WITH DIFFERENT BROOMRAPE POPULATIONS**

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Rapid generation of reactive oxygen species (ROS), which is a specific early feature of the hypersensitive response (HR), play a central role in plant pathogen defense. ROS produce not only direct antimicrobial effects, but also participate in cell signalling associated with expression of defense genes, HR, bonding between proteins within the cell wall, phytoalexin synthesis, callose deposition and systemic acquired resistance (SAR).

ROS production, including superoxide radical ($O_2^{\cdot-}$), hydrogen peroxide (H_2O_2), hydroxyl radical (OH^{\cdot}) and other, is associated with the normal metabolism of the plant cell. Under the stress, plant cells could produce an "explosion" of ROS referred as "oxidative burst", which is primarily composed of H_2O_2 . Production ROS is among the first crucial events that occur in plants during pathogen recognition. ROS are molecules that contain oxygen and this is extremely chemically reactive, interacting with proteins, DNA and membrane lipids, thus reducing photosynthesis, increasing leakage of electrolytes, accelerating senescence and cell death. Elevated level of ROS induces not only the biosynthesis of antioxidant molecules such as ascorbate, glutathione and polyamines but leads to increased activity of antioxidant enzymes such as superoxide dismutase (SOD), catalase (CAT), ascorbate peroxidase (APX), glutathione-S-transferase (GST) and other.

The purpose of this study was to determine expression levels of ROS-scavenging genes (*MnSODI*, *APX3* and *AOX1A*) in leaves (90 days after sowing) of seven sunflower CMS lines (MS-2161A, MS-2098A, MS-2091A, MS-2077A, MS-2067A, MS-2039A and MS-1589A) infected with three *Orobanche cumana* Wallr. populations (from Romania, Tulcea and Republic of Moldova, Soroca and Anenii Noi).

Screening of broomrape infection and evaluation of three phytopathological parameters (frequency, intensity and attacking rate) were performed for estimation of resistance of studied genotypes. The total RNA from three plants was isolated using TRI Reagent (*Ambion, Applied Biosystems*) and mixed into a single bulked sample. First strand cDNA for was synthesized using RevertAid RT Kit, Oligo(dT)18 and Random Hexamer primers (*Thermo Scientific*) following the manufacturer's instructions. Primers for quantitative real-time RT-PCR were designed using web based primer designing tool Primer3Web v.3.0.0. The mRNA expression levels of selected samples were estimated by quantitative real-time PCR (qPCR) using Maxima SYBR Green/ROX PCR Master Mix (*Thermo Scientific*) according to the manufacturer's protocols on DT-96 (*DNA Technology, Russia*). Each sample was analysed in three replicates performed in three different runs and the mean value of each triplicate was used for further analysis. Fold changes greater than 1,5 or lower than -1,5 were considered biologically significant.

Five lines from seven investigated were highly susceptible to all three populations. MS-2161A was resistant to broomrape population from Anenii Noi and tolerant to those from Soroca, and MS-2039A resistant only to Anenii Noi broomrape. The expression of studied genes was much more altered in highly susceptible genotypes than in those resistant, which showed higher metabolic stability. Significant differences in number of cases of ROS-scavenging genes with modified transcriptional activity in infected and non-symptomatic plants were not ascertained. The transcriptional activity of *MnSODI*, *APX3* and *AOX1A* genes was weakly influenced by infection with broomrape (67 % cases) or was down-regulated (24 % cases). Some up-regulation cases (9 %) for *MnSODI* (MS-2039) and *AOX1A* gene (MS-2067) were revealed. *AOX1A* was the most responsive gene, especially when infection was produced by population from Anenii Noi. In summary, the results obtained in this work showed that in sunflower infected with broomrape ROS-scavenging genes (*MnSODI*, *APX3* and *AOX1A*) did not significantly change their transcriptional activity, pointing out that in the late stages of infection plants were adapted. Further investigations regarding ROS accumulation, enzymatic activity and isozyme patterns are required for a better understanding of molecular events in sunflower-broomrape pathosystem in earlier stages of infection.

EXPRESSION GENES INVOLVED IN METABOLIC PATHWAY FOR SYNTHESIS OF SCLAREOL *Salvia sclarea*

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The sclareol (*labd-13-en-8-ol diphosphate*) is a natural product from labdane diterpenes class of particular value in the perfume and cosmetics industry [1], identified in *Salvia sclarea* L. (*Lamiaceae*), *Cistus reticus* L. (*Cistaceae*), *Nicotiana glutinosa* L. (*Solanaceae*) and *Cleome spinosa* L. (*Brassicaceae*) [2].

In the production and isolation of this compound was used predominantly the species *Salvia sclarea* L., because it is characterized by the largest amount of sclareol - about 0,4-2,6% [3].

Starting point in the synthesis of sclareol is **geranylgeranyl pirofosfate** (GGPP) from metabolic path of terpenoids formation (KEGG-ath009000), subsequently transformed into **labda-13-en-8-ol** and respectively - in **sclareol** [4]. These processes develop with the involvement of enzymes *labda-13-en-8-ol diphosphate synthase* (*LPPS*) and *sclareol synthase* (*SS*), for which in bioinformatics resources have been identified ARNm sequences: AET21246.1 - *Salvia sclarea* (*clary*) *sclareol synthase* [5] and JQ478434.1 - *Salvia sclarea labd-13-en-8-ol diphosphate synthase* (*LPPS*) [2].

Sequences of interest were the basis for generating *in silico* (*PRIMER3web*) pairs of specific primers to generate an amplification product of 75 – 150 pb, were used to estimate the level of expression of transcripts of genes involved in the the metabolic pathway of synthesis of the *Salvia sclarea* L.

Real-time amplification revealed relevant results only for gene JQ478434.1, responsible for the synthesis of *labda-13-en-8-ol diphosphate synthase*. In reaction qRT-PCR was obtained one specific amplicon with the size of about 133 pb in all genotypes (28) included in the study, tested primers generating a common profile.

In most cases the expression of genes correlated with the heterosis effect, increased amounts of the transcript were identified in hybrids, compared with the parental forms, in particular at hybrid [*M-69 655 S₉ x (M-69 429-82 S₃ x 0-40 S₅)F₇*]*F₁* followed by hybrid [*S. s.Turkmen/N S₇ x (K-36 x 0-41) F₂ x 0-19)F₁ x 0-22) B₁ x L-15)F₈*]*F₁*, and lowest values is observed for the hybrid [*S. s.Turkmen/N S₇ x (Rubin x S1122 9 S₃)F₁ x (0-56 x V-24) F₁)*]*F₇*. If parental forms, the highest level of transcripts is highlighted in the form *S. s.Turkmen/N S₇*, followed by parental form *Rubin x S1122 9 S₃)F₁ x (0-56 x V-24) F₁*, while the lowest value was obtained in the form (*K-50)F₅ x S 1122(102+113)F₂ x K-43)F₄*.

In conclusion, it was determined the gene expression *LPPS*, being demonstrated the involvement of it in the biosynthesis of sclareol at *Salvia sclarea* L. *SCLAREA* L.

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MOLECULAR DIVERSITY OF *O. CUMANA* POPULATIONS ASSESSED
USING ISSR MARKERS

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Broomrape (*Orobancha cumana* Wallr.) is an obligate chlorophyll-lacking holoparasite that lives attached to the roots of sunflower (*Helianthus annuus* L.), depleting the plant of nutrients and water.

This parasitic plant has spread especially in the central and south-eastern regions of Moldova. Actually, a considerable expansion has been noticed also in the north region with increasingly frequent cases of occurrence during the last 5 years. The damage caused by the pathogen is often devastating with reported yield losses up to 50-90% (Linke et al., 1989; Bulbul et al., 1991; Dominiques et al., 1996; Blamey et al., 1997; Hibberd et al., 1999), leading to a significant reduction in the amount and quality of the oil (Alonso et al. 1996; Duca et al., 2012).

Over the past few years, the broad distribution and the continuous expansion of this parasitic plant have been pointed out. Knowledge of the variability in the population genetics of weedy broomrapes is important in any attempt to develop resistance-breeding strategies for the relevant host crops against these parasites.

Morphological traits are, in the case of obligate plant parasites, of limited use to diversity studies. Because of their variation with environmental changes and estimation errors, there are a reduced number of characteristic features available, as holoparasites have no leaves and present only false roots. In contrast, molecular marker-based genetic diversity analysis has potential for assessing changes in genetic diversity over time and space. Methods that rely on DNA polymorphism have been increasingly applied to determine fixed interspecific differences and to identify species difficult to distinguish based on their anatomical and morphological characteristics. Numerous taxonomic studies using various molecular marker classes have shown that true biological species differ in both qualitative and quantitative terms at the molecular level.

In recent years, as an effective and a relatively new molecular marker technique called inter simple sequence repeat (ISSR) (Zietkiewicz et al., 1994) has been widely used for plants diversity analysis, DNA fingerprinting drawing or variety identification.

The ISSRs are semiarbitrary markers amplified by PCR in the presence of one primer complementary to a target microsatellite. The primers are 16-18 bp long composed of a repeated sequence and could be flanked at the 3' or 5' end by 2-4 arbitrary nucleotides –anchored primers (Zietkiewicz et al., 1994). Such amplification does not require genome sequence information and leads to multilocus and highly polymorphous patterns (Zietkiewicz et al., 1994; Nagaoka & Ogihara, 1997).

Recent ISSR studies of natural populations have demonstrated the hypervariable nature of these markers and their potential use for population- level studies (Culley & Wolfe, 2001; King et al., 2002). The genetic diversity of several invasive plant species has been successfully assayed using ISSR markers.

A confirmation that the ISSR can be a precise method for identification (Roman et al. 2002) is the molecular diversity in and between the populations of *O. crenata*. Benharrat et al. (2002) used ISSR markers to study genetic diversity among four species of *Orobancha*. All primers detected polymorphisms in five populations of *O. cernua* and *O. cumana* collected in different countries, allowing the differentiation of the two species. Buschmann et al. (2005) reported that using of ISSR markers allowed characterization of two *P. ramosa* populations with different levels of virulence parasitizing nine tobacco cultivars grown in Europe. Hristova et al. (2011) and Stoyanov et al. (2012) identified a useful set of ISSR markers for characterize the biodiversity and the phylogenetic relationships different *Orobancha* species in Bulgaria. Altogether, genetic studies can provide a general framework for strategies to control the parasite.

The research regarding genetic variation of *Orobancha cumana* populations at country level are very scarce. Our study advances the understanding of the genetic variation within and between broomrape populations sampled throughout Republic of Moldova by applying the ISSR technology. Also, the use of these molecular markers may be a suitable method for the identification of pathogenic groups in parasite populations and for comprehension of the sunflower broomrape phylogeography on the entire territory of the country.

**VAVILOV'S COLLECTION OF CULTIVATED PLANTS AS THE BASIS FOR
SUCCESSFUL BREEDING AND FOOD SECURITY**

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The genetic resources of cultivated plants and their wild relatives are basic components that determine the food and environmental security of each sovereign state, including Russia. At present, they have become especially topical and strategically important in connection with growing genetic erosion and the disappearance of agricultural plant varieties, species, and genera from the face of the earth. According to the data of the UN Food and Agriculture Organization (FAO) for 1998, 75% of the global genetic diversity of crops was lost in the 20th century. In addition, the number and assortment of arable crops and species have decreased substantially. Only 30 crops ensure 95% of humankind's needs for caloric vegetable food, and four of them (rice, wheat, corn, and the potato) cover more than 60% of the needs for energy and proteins. Under global and local climate changes, the loss of diversity and the shortage of crops used in production are a serious food threat to the entire world community. Hence, at the present stage, the preservation and rational use of plant genetic resources (PGRs) are nationally significant and strategically important for every country. In Russia, the problems of PGR mobilization, preservation, research, and use have been studied for 120 years by the Vavilov Research Institute of the Plant Industry. Scientists of several generations have been collecting the VIR collections, and today the monitoring, collection, preservation, documentation, detailed investigation, and efficient use of the world's PGRs continue and develop based on modern achievements of science and technology. These collections include more than 324000 plant samples (representing 64 botanical families, 376 genera, and 2169 species), occupy the fourth place in the world by the quantity of samples preserved, and are the richest in terms of their botanical, genetic, geographical, and environmental diversity. It is not by chance that the FAO steering council on plant genetic resources included VIR and its collections in the list of Future Harvest Centers, which unites the 11 largest PGA preserving gene banks.

The significance of the collections for domestic selective breeding, food production, and the development of environmentally safe agriculture can hardly be overestimated. Suffice it to say that, in the second half of the 20th century, as a result of studying and using samples from the VIR collections by plant breeders from Russia and the former Soviet republics, unique varieties were created, which made it possible to increase grain crop yields by two to five times, boosting considerably gross grain yields. By introducing wild potato species, collected in South and Central Americas by VIR expeditions, into selection alone, this culture was disseminated across Russia and acquired the traits of early maturation, resistance to diseases and pests, and a better storage capacity, and its crop yield increased by three to four times by the end of the 20th century. Thanks to the use of the sources of the early maturation and cold endurance of wheat, oats, sunflower, soybean, cotton, rice, corn, and other cultures in selection, their arable areas have moved far northward; in particular, the area of corn for grain has spread almost 2000 km northward and 10000 km eastward. The country's fields cultivate 80% of the varieties and hybrids of agricultural crops obtained owing to the collections' samples. For example, on the basis of the collections, more than 300 varieties of rye, oats, and barley were bred, more than 120 of which were regionalized. The dominant shortstem gene, found in the rye collection, served as the parent material for a general global trend in winter rye selection—the creation of erect varieties. Shortstem varieties, bred using this gene, now occupy about 80% of the rye planted area in Russia and the CIS countries, annually yielding an income of billions of rubles. Comprehensive studies over the past ten years have allowed us to extract and create by traditional methods more than 20000 genetic sources and 500 donors of selection crucial gene alleles and polygenes, and to form numerous trait and gene collections of major agricultural crops for their target application in selection programs. New effective and rare gene alleles, previously unused in selection, were identified and mapped to determine their photoperiodic sensitivity and reaction to vernalization, resistance to pathogens and other traits. In addition, quantitative trait loci (QTLs) were marked and molecular genetically mapped in order to identify and transfer under control chromosome loci that determine the manifestation of economically valuable traits of major agricultural crops. The genetic diversity of the collection material, studied using traditional technologies and molecular genetic approaches, is a powerful basis for valuable source material and an effective tool for the efficient development of breeding.

INFLUENCE OF SOME GLYCOSIDES OF PLANT ORIGIN ON THE SEED YIELD OF FENUGREEK (*TRIGONELLA FOENUM-GRÆCUM* L.)

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Trigonella foenum-graecum L. (fenugreek) is an annual crop and dicotyledonous plant belonging to the subfamily *Papilionaceae*, family *Leguminaceae* (the *Fabaceae*) with trifoliate leaves, branched stem, white flowers, roots bearing nodules and golden yellow seeds. Fenugreek seeds are an important source of essential phytochemicals such as steroids, galactomannans and 4-hydroxyisoleucine, content of which varied from 0,6 to 2,2% depending on the seed origin, growing area and the meteorological factors of the growing season. In addition, the composition of secondary metabolites includes the polyphenolic substances that possess antioxidant activity. Numerous researches were carried with the goal to achieve an increase in yield and improvement of the quality of fenugreek seeds as well as plant protection. To this aim the biologically active substances of plant origin, namely; steroid, iridoid and flavonoid glycosides could be used as stimulants of growth, development and productivity of this non-traditional for Moldova plant species at their cultivation in new agro-climatic conditions.

The purpose of this investigation was to study the influence of two biologically active compounds obtained from the aerial part of *Linaria vulgaris* L. (linarioside) and *Scrophularia nodosa* L. (scrophularioside) on seed productivity of fenugreek. Fenugreek seeds used in this investigation were produced in the plant collection of the Institute of Genetics, Physiology and Protection of Plants. The method of pre-sowing treatment of seeds by soaking as well as foliar treatment with aqueous solutions of 0.01% glycosides was applied. The effect of these treatments on plants growth, yield and quality of fenugreek seeds (total phenolic content and antioxidant activity) were investigated during the seasons of 2013-2014. Total phenolic content was determined by Folin-Ciocalciu method, and antioxidant activity of extracts was appreciated *in vitro* using potentiometric procedure of peroxy radical scavenging. Method is based on peroxy radicals generation by the 2,2'-azobis(2-amidinopropane)dihydrochloride. Both the total phenolic content and antioxidant activity were expressed in gallic acid equivalent (GAE) per g of seed.

Obtained data have shown that fenugreek crop responded positively to the application of treatment with glycosides solutions by increasing the quantities of plants with high vitality (approximately 1,42-1,86 times) and formation of more pods per plant. Thus, the plants treated by scrophularioside and linarioside formed $49,70 \pm 1,65$ and $49,18 \pm 1,61$ pods/plant, respectively. These values were higher by 17,70 and 16,60% than that of non-treated plants. Fenugreek also enhanced its productivity by numbers of the seed in the pod (11,06-11,42 seeds per pod). Furthermore, there were observed the statistically significant weight increasing of seeds collected from one plant. The highest weight ($9,22 \pm 0,37$ g) had the seeds obtained from one plant of fenugreek treated with linarioside. Seeds weight of non-treated fenugreek and treated with scrophularioside were equal $7,22 \pm 0,32$ g and $8,77 \pm 0,36$ g per plant, correspondingly. Moreover there was determined that application of linarioside also leads to weight increasing of 1000 seeds, which reached to $16,95 \pm 0,18$ g. This index of seeds from non-treated plant was $15,75 \pm 0,17$ g. To conclude, obtained results confirmed that pre-sowing and foliar treatment of fenugreek with glycosides of plant origin contributed to higher seed productivity, which increased in our experiences by 18,66-23,83%.

Total phenolic content of seeds obtained from non-treated plant was $3,95 \pm 0,29$ mg/g of seed. The same indexes of seeds from fenugreek treated with linarioside and scrophularioside were $4,01 \pm 0,40$ mg/g and $4,23 \pm 0,37$ mg/g respectively. There was observed a tendency for greater accumulation of secondary metabolites such as phenolic substances, but these data had no significant statistical difference. Phenolic substances constituted approximately 2,1-2,8% from all compounds extracted by 70% ethylic alcohol. The antioxidant activity of seeds extracts of non-treated plants as well as treated with scrophularioside did not differ significantly and varied from $3,52 \pm 0,71$ to $2,98 \pm 0,12$ mM GAE/g of seed, respectively. The highest antioxidant activity ($3,92 \pm 0,08$ mM GAE/g of seed) belonged to the fenugreek treated with linarioside. Due to the fact that fenugreek crop needs warm and dry conditions for proper growth the accumulation of phenolic substances and its antioxidant activity were higher by more than 1,5 times in drought season of 2012 in comparison with the same indexes determined in 2014. In conclusion, the utilization the biologically active substances such as iridoid and flavonoid glycosides contributed to the improvement of fenugreek crop through increasing plants vitality, seed yield and accumulation of secondary metabolites with antioxidant activity.

THE USE OF BIO-REGULATORS OF PLANT ORIGIN FOR GROWING *LINUM USITATISSIMUM* L.

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Linum usitatissimum L. belongs to the family of *Linaceae* S.F.Grai., which consists of 18 genera and 330 species widespread on all the continents. The most populous genera is *Linum* L., which has about 200 species. The greatest practical interest has *L. usitatissimum* L., which is known for various species, forms, cultivars and distinctive characteristics.

A considerable interest for the introduction and breeding of this species could be noticed in the last decade among the specialists. This is due to the emergence of new properties of flax fiber and oil, beneficial to the human health.

It should be noted that the most widespread in crops, in a wide variety of species, is the flax curly. As object of the conducted research served the seeds of a local population, periodically sown to preserve their field germination.

In the spring of 2014 a pre-sowing seed treatment and foliar fertilizer with aqueous solutions of various concentrations were carried out on two glycosides - linarioside and scrofularioside, which were obtained from the aerial parts of two representatives of the *Scrophulariaceae* family (*Linaria vulgaris* L. and *Scrophularia nodozsa* L.), common in the local flora.

According to the obtained results, it was concluded that the pre-sowing seed treatment and the foliar solution dosing of the above mentioned biologically active substances did not have a pronounced effect on the growth process of the plant. The only observation was the significant difference in the plants height from that of the monitored indicator, which can be confirmed by the values of the coefficients of variation. Slight differences were noticed between the treatment methods when distributing the plants into groups according to the vitality level.

However, the solutions of glycosides stimulated the development of the plants' generative organs. In result of using a 0,01% linarioside solution, the number of fruits on a single plant was of $121,71 \pm 5,56$ pieces, when using scrofularioside, $121,71 \pm 5,14$ pieces were obtained, while in the monitored method, $99,35 \pm 4,70$ pieces were achieved; which is by 18,4% and 18,1% more.

The quantities of seeds in the experiments were almost the same: $6,44 \pm 0,30$ g per plant for linarioside and $6,58 \pm 0,29$ g per plant for scrofularioside, while in monitoring, the quantity was of $5,26 \pm 0,28$ per plant. The pattern of distribution of plants in groups according to their level of vitality also differed slightly, but the greatest number of individuals (53% and 52%) with a low vitality were observed in the control embodiment.

Therefore, in order to stimulate the biological productivity of *Linum usitatissimum* L., it is recommended to conduct seed pre-sowing soaking and foliar dosing with a 0,01% solution of biologically active substances derived from the Figwort family.

CURRENT STATUS AND PERSPECTIVES OF CONSERVATION OF HAZELNUT AND CORNELIAN CHERRY IN THE REPUBLIC OF MOLDOVA

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Under the conditions of global climate change, dramatic degradation of natural resources, growth of human population on the Earth, food shortage and maldistribution of food, the role of crop wild relatives (CWR) as one of stabilizing factors of civilization development significantly increases. CWR of natural flora have the wide spectrum of valuable traits and properties – resistance to draught and limiting temperatures, diseases and pests, increased content of bioactive substances and main nutrients. Gene pool of these species created during evolution is a treasurable source material for plant breeding. Along with cultivated species CWR form a part of plant genetic resources, the diversity of which determines national food security.

Laboratory of Plant Genetic Resources of the Institute of Genetics, Physiology and Plant Protection of Academy of Sciences of Moldova conducts complex studies on inventory, collection, investigation and conservation of genetic resources of crop plants and their wild relatives. Special attention is paid to investigation of natural populations of CWR, search for the germplasm possessing useful properties. Among CWR of fruit crops growing in forest ecosystems of Moldova, hazelnut (*Corylus avellana* L.) and cornelian cherry (*Cornus mas* L.) play a very important role. *In situ* conservation of hazelnut biodiversity represent a challenging issue of high priority in the context of current trend of reduction of areas of pure hazelwoods and decreased presence of this culture in many forest types. On the other hand, share of wild fruit tree-shrub cultures including cornelian cherry grows among the main species used for restoration of forests. For this purpose, for example, more than 2 million seedlings and young plants of *Cornus mas* from the material grown in forest tree nursery were planted in 2002-2008.

For the purpose of identification of current status of growth of abovementioned species populations, routing investigations were conducted aimed at their inventorying and GPS-positioning in forests of different soil-climatic zones of Moldova in more than 30 forest stations and protected natural areas. Cornelian cherry and hazelnut are found in the undergrowth in forest associations with common oak, durmast oak, hornbeam, *Tilia*, ash tree, smoke-tree, forming sometimes brushwood of various density. It was noted that land topography and vegetation conditions influence on the growth, functional state and fructification of populations of *Corylus avellana* and *Cornus mas*, their resistance to drought and other unfavorable environmental conditions. Plants growing in forest stations located in Edineț, Soroca, Nisporeni, Strășeni, Călărași and some other regions were in the best state among natural populations of hazelnut. Biometric measurements of plants were made and showed that cornelian cherry brush height varied within 2-7 m, and tree stem diameter was 3-20 cm. Diversity was noted with regard to seed shape and size and taste qualities. Hazelnut bushes were as high as 4-7 m, sometimes up to 10 m, and had 2-5 main stems 2-16 cm in diameter.

The issue of intraspecific diversity of said species in their natural habitats is a noteworthy point important for determination of degree of genetic erosion under the conditions of dramatic fragmentation and significant anthropogenic pressing on forestlands, characteristic for the Republic of Moldova. On the other hand, examination of mono-dominant and mixed artificial plantations of hazelnut and cornelian cherry in different forest stations (Vadul lui Vodă, Susleni, Anenii Noi, Singerei, Chișcăreni, Bobeica, Căzănești and etc.) revealed their ineffective use (solely for fruit harvesting) in the absence of any management. However it should be noted that these sites are ideal “natural laboratories” where valuable genotypes can be searched for and selected, reproduced and introduced in the forestry. Selected forms could become valuable sources for traits important for breeding of new productive cultivars and hybrids resistant to environmental stresses.

Conducted studies allowed to identify current distribution and reliability of conservation of 2 species of fruit crop wild relatives in forest ecosystems in different soil-climatic zones of Moldova, and to determine the perspectives for implementation of methods of their further conservation and sustainable use.

GENETIC RESOURCES OF SUNFLOWER AT VIR COLLECTION

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The sunflower collection at VIR totals 2780 accessions that include 2230 accessions of cultivated (*Helianthus annuus* L.) and 550 of wild sunflower belonging to 24 species (of which 5 are annual and 19 are perennial species). The collection of cultivated sunflower is represented by local varieties and landraces, by cultivars of national and foreign breeding, as well as by populations collected by collecting missions. The collection includes 150 sunflower ancient varieties of domestic breeding such as Chernyanka, Smena, Voshod, Peredovik, Armavirsky, etc., are the progenitors of the modern sunflower inbred lines for heterotic breeding. More than 300 lines were created by VIR researchers, and by Russian and foreign breeders. The collection also includes the first CMS-based hybrids which are conserved as hybrid populations, and lines. For instance, VIR 114 was produced by crossing VIR 104 (CMS PET) with cv. Sputnik (bred at the Armavir Station). Parental forms of VIR 104 are the CMS source from Leclercq and cv. Armavirsky 1813. VIR 116 was created on the basis of an American line CMS HA 234, which, in its turn, had been obtained from a hybrid between a CMS source from wild sunflower from Texas and cv. Smena (bred at VNIIMK). The Vympel cultivar was used as the paternal form for VIR 116. Pedigrees of both lines feature a CMS source obtained from an interspecific hybrid between the wild annual and cultivated sunflowers and also a Russian variety. The collection of lines has been composed either as a result of studying intraspecific and varietal diversity and repeated self-pollination, or on the basis of interspecific hybridization. The genetic collection includes 189 lines with different manifestation of morphological characters; 120 pollen fertility restorer lines; 20 CMS lines and their fertile analogs; 46 lines with genes for resistance to strains (330, 710 and 730) of downy mildew (*Plasmopara halstedii* (Farl.) Berl. and de Toni); 90 lines among 362 lines of genetic collection are marked by seed storage protein polymorphic variants. The collection includes field entries from different parts of the world. Of particular interest among the latter are the materials collected by M. Duca during the collection missions to the Primorskiy krai. These are tall (with a plant height between 4.0 and 3.5 m), large fruited and resistant to *Phomopsis helianthi* Munt.-Cvet. accessions. The other 86 large fruited accessions (with 1000 seed weight of 90-130 g) entered into the collection from Armenia (Gâr-Gâr), Argentina, Bulgaria, China, and the modern varieties were obtained from Russian institutions.

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BIOCHEMICAL DIVERSITY OF THE *ORIGANUM VULGARE* SSP. *VULGARE* L. AND *ORIGANUM VULGARE* SSP. *HIRTUM* (LINK) IETSWAART GENOTYPES FROM MOLDOVA

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Origanum vulgare L. is a herbaceous perennial species in the family of *Lamiaceae* known and used for ages. Multiple *Origanum* biotypes, genotypes, forms, and taxons have a strictly local distribution and are distinguished through accentuated morphological and biochemical diversity, which is confirmed by the studies on the species and subspecies in the definite areal of prevalence. The relevance of the studies on *Origanum* is also determined by the importance of the species as a medicinal, aromatic, culinary, spicy, ornamental, and meliferous plant, supported by the chemical composition that includes flavonoides, vitamins, bitter substances, and essential oil, synthesized and accumulated in the aerial part of the plant. This is essential oil and, especially, carvacrol, that is responsible for the antimicrobial, antifungal, and antioxidant action, as well as the capacity to inhibit bacterium growth. *Origanum* essential oil is also successfully employed for its antispastic and antiseptic action. *Origanum* is known to possess sedative, carminative, emenagogous, diuretical and other actions. It is also utilized as flavouring and preservative agents in food products. It is known that the *O. vulgare* decoction possesses antioxidant activity, while its hydroalcoholic extract demonstrates antimicrobial effect.

This investigation was carried out during the 2014 growing period and included ten genotypes of *Origanum vulgare* ssp. *vulgare* L. and *O. vulgare* ssp. *hirtum* (Link) Ietswaart. In order to evaluate the essential oil content, the samples of fresh herbs and the aerial part of the plant were harvested in the morning hours at the flowering stage. The essential oil was isolated by hydrodistillation. Qualitative and quantitative analyses of the essential oil were conducted using GC GC-MS. Previous researches have demonstrated that *O. vulgare* ssp. *vulgare* and *O. vulgare* ssp. *hirtum* plants are characterized by the formation of shrubs 65-80 cm tall and a large number of floral stems: 100-115 in ssp. *vulgare* and 120-240 in *O. vulgare* ssp. *hirtum*. The inflorescences of the genotypes belonging to the species *O. vulgare* ssp. *hirtum* develop 20-30 ramifications, while those of *O. vulgare* ssp. *vulgare* - 15-18. These two species also differ in the inflorescence ramification length - *O. vulgare* ssp. *hirtum* has shorter ramifications, thus, the inflorescence being more compact. Flowers have a white corolla in *O. vulgare* ssp. *hirtum* as opposed to a pink flower corolla in *O. vulgare* ssp. *vulgare*.

The genotypes differ in the content of the essential oil extracted using hydrodistillation. Its content in the *O. vulgare* ssp. *vulgare* genotypes varies from 0,077% to 0,360% (dry matter). The *O. vulgare* ssp. *hirtum* genotypes are richer in essential oil the content of which ranges between 2,315% and 4,923% % (dry matter). The quantitative and qualitative analyses carried out using GC GC-MS techniques have shown that the number of the components identified in the *O. vulgare* ssp. *vulgare* genotypes varies from 17 to 28 depending on the genotype. The number of the components identified in the essential oil of the *O. vulgare* ssp. *hirtum* genotypes is different and ranges from 18 to 25, the identification rate making 98,09% to 98,80 %. The concentration of the components identified in the essential oil is also different in all the genotypes of the both subspecies. Thus, carvacrol is the major component in the essential oil of five genotypes *O. vulgare* ssp. *hirtum*, its concentration being 74,63% to 88,13% depending on the genotype. In these genotypes, we can mention the presence γ -Terpinene in the essential oil at relatively high concentrations ranging from 3,59% to 10,69%, and that of p-Cymene at concentrations of 2,23% to 5,06%. The *O. vulgare* ssp. *hirtum* genotypes divided into three chemotypes: 1- carvacrol/p-Cymene; 2- carvacrol/p-Cymene / γ -Terpinene; 3- carvacrol/ γ -Terpinene /p-Cymene.

The essential oil isolated from the *O. vulgare* ssp. *vulgare* genotypes differs in that it contains four major components, D-Germacrene occurring at higher concentrations (26,01-33,98%). The second major component is β -Caryophyllene at concentrations of 12,16% to 33,16%. The third component, γ -elemene, was certified at concentrations of 3,82% to 16,79% in the essential oil of the *O. vulgare* ssp. *vulgare* genotypes. Another component in the essential oil this subspecies - (+) β -bisabolene was certified in concentrations from 0 to 16,4%. The *O. vulgare* ssp. *vulgare* genotypes divided into three chemotypes: D-Germacrene/ β -Caryophyllene/ γ -elemene; D-Germacrene/ γ -elemene/ β -Caryophyllene; β -Caryophyllene/D-Germacrene/ β -bisabolene.

The research conducted has shown that the polyphenols content varies in all the samples evaluated. Thus, the polyphenol concentrations are within the range of 12,7 - 17,08 mg/g (dry mat.) in the genotypes of ssp. *hirtum*, while their range is greater in the ssp. *vulgare* genotypes (11,1-25,9 mg/g).

**THE MARKING OF RESISTANCE GENE *Or₅* TO BROOMRAPE RACE E OF
OROBANCHE CUMANA WALLR. IN SUNFLOWER LINES OF VNIIMK'S BREEDING**

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The sunflower is one of the most important sources of oils in the world and the main source in the Russian Federation. Currently a vigorous limited factor in oil production is broomrape (*Orobancha cumana* Wallr.). It is a phytoparasite with a high potential of reproduction.

In present time, a real problem in the domestic breeding is a search of donors of resistance to the highly virulent races of broomrape: F, H, and G. In collections of inbreds the amount of samples resistant to race E is restricted. It is possible to isolate such resistant samples by means of molecular-genetic methods in laboratory. It assigned the aims of the present work to identify the gene *Or₅*, controlling the resistance to broomrape race E in genetic collection of VNIIMK. The identification was conducted with PCR methods with known (Tang et al., 2003; Lu et al., 2000) SSR- and SCAR-primers.

There was investigated a population F_2 , developed as a result of cross of lines VA330V (resistant to race E, genotype *Or₅Or₅*) and I6SL-Lakomka 1 (susceptible to race E, genotype *or₅or₅*) of VNIIMK breeding.

Seven primers revealed the polymorphic products of DNA amplification in studied combination. But after hybridological analysis only DNA loci showed the actual segregations corresponding to theoretically expected ones. We carried out linkage analysis of gene *Or₅* with these three loci.

A test on an independent inheritance proved the linkage of loci *Or₅* – ORS1036 with frequency of recombination $0,12 \pm 0,04$ (12 cM) and *Or₅* – RTS 05 with frequency of recombination $0,10 \pm 0,04$ (10 cM). Locus RTS40 was inherited independently of the resistance gene to the race E.

These data on recombination frequencies between the studied genes are differed from the results of Tang et al. (2003) and Lu et al. (2000). Due to data obtained Tang et al. (2003) SSR-locus ORS1036 was located at 7,5 cM far from locus *Or₅*. In researches of Lu et al. (2000) SCAR-marker RTS 05 was mapped at 5,6 cM to distal end of *Or₅*, and RTS40 was linked with the gene of resistance to the race E and was located at 14,1 cM away.

Such discrepancy of the results obtained by the different researches can be caused by some reasons. In present time, it is almost impossible to differentiate the broomrape races per se. The broomrape populations where the race E is dominant always contain impurities of more virulent biotypes. In a population we used the race E was prevailed but there were also impurities of a more virulent race F. In view of this fact we considered as resistant the sunflower plants having no more than two-three broomrape tubercles on roots. However, a presence of a more virulent race in a population leads to a distortion of segregation in any case. It should be also considered that the populations with the identical race composition but developing in the different geographically distant places are really differed with their virulence; and this fact effects a reproduction of the genetic researches conducted in the different countries. In work of Tang et al. (2003) there was also noted that distances between *Or₅* and DNA-markers can be influenced with phenotypic mistakes. It occurs as an assessment of a susceptible phenotype as a resistant one at classification leads to increasing of a class of recombinant genotypes and distorts the distances on a genetic map. In some cases, unidentified genes, which expression depends on a temperature and ecological factors, can effect on expression of the dominantly inherited genes of resistance (Honiges et al., 2008). Thus, two loci of DNA – RTS05 and ORS1036 – in genetic collection of VNIIMK are linked with the gene of resistance to the broomrape race E with a frequency of recombination $0,10 \pm 0,04$ and $0,12 \pm 0,04$, respectively.

**OBTAINING LINES OF TRITICUM AESTIVUM WITH
PERSPECTIVE FOR IMPLEMENTATION IN THE SELECTION**

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Classical experiences, carried out at the Institute of Physiology and Plants Biochemistry of ASM, have shown that under the influence of the solar radiation regime of alternative high altitude with the darkness in the period of morning and evening, different varieties of common wheat have induced mutations. Using this method to improve wheat, C. Moraru obtained more valuable forms through artificial mutagenesis of plants of Caucaz variety. One of them, called *mA 8/1*, form medium size spikes and thin stem. This form has proved to be resistant to drought. In extremely dry years this form plants survived and cropped on the experimental field of the Institute of Genetics, Physiology and Plant Protection (IGFPP). Their high resistance to drought is manifested by those that plants of mentioned variety have green stems, leaves and spikes during drought period when the sowing of the initial variety (and other varieties of wheat) were yellow and grains on spike were shriveled.

Another factor that caused genetic and phenological changes was the introduction in Moldova of varieties from different regions of the globe. Centurk variety descendant, from the USA, in the collection of the Institute of Field Crops from Balti gave new forms of spontaneous mutations. Form *Lp 2/3* highlighted by reducing plant height by 20 cm compared to the native (variety of Centurk). The plants of *Lp 2/3* form have a greater spike mass and is manifested by increasing 1000 seeds mass by 30% - 40% and the amount of wet gluten in grains by 4% over the original variety seeds. In so gluten quality of seeds of *Lp 2/3* line corresponded to the highest quality standard.

Mutant forms of Autumn common wheat created by changing the length of duration of solar radiation, *mA 8/1*, and the introduction from a distant geographical area, *Lp 2/3* have been proposed for use in improving wheat as sources for obtaining hybrid grains that brings grain high yield and quality with increased resistance of plants to winter frosts and droughts possible during spring and summer. In addition it was very important that plants showed a high resistance to diseases. To achieve the purpose the plants of *Lp 2/3 x mA8/1*; *Lp 8/1 x mA 2/3* were crossed. Obtained forms were crossed too with the most resistant to frost variety - Albidum 114 (Albidum 114 x *Lp 2/3*; Albidum 114 x *mA8/1*) as well as with the Sapphire variety (Sapphire x *Lp2/3*; Safir x *mA 8/1*), which is characterized by large grains. As the result of these crosses were obtained characteristic grains for 10 hybrids. They were planted and tested on the experimental field of IGFPP during 7 consecutive generations in the years 2007-2014. During this period 2007 year was characterized by severe drought and 2012, 2013 and 2014 years were heavy rains during flowering and grain ripening. In these adverse conditions the resistance of plant obtained from hybrid seeds was much better than of parental forms plants and of the Odesscaia 267 variety. In 2007 all hybrids were highlighted by increased resistance to drought, wet gluten grains containing 28-30% when the initial forms and beans contain 24-26% of gluten. In the year with excessive humidity (2012, 2013 and 2014) hybrids and mutant *mA 8/1* were affected by brown rust and blight. Therefore we developed a rigorous selection of genotypes resistant to diseases. Based on these selections were obtained few lines with valuable properties, lines that have been recommended for use in breeding programs in 2015. Among them we mention the following:

№14 (mA 8/1 x Safir variety), medium-sized plants, resistant to drought and winter, medium resistance to major foliar diseases (*Puccinia recondite f. sp. tritici*, *Puccinia striiformis*, *Tiletia spp.*, etc.), coefficient twinning is 2,4-2,8 and red seeds with high protein content and with good bakery features.

№ 16 (mA 8/1 x LP 2/3), plants with thick stems medium sized, resistant to drought, winter and foliar diseases, coefficient twinning is 2,6-3,0; 10-15% increase in yield compared to the parental forms.

№ 18 (Albidum 114 x LP variety 2/3) - resistant to drought and winter, differs from the other lines with high resistance to rust (*Puccinia recondite sp. P. striiformis*); beans have very good baking qualities.

In general, most indices characterizing the quality of grain and grain productivity at obtained lines were superior to Odesscaia 267 variety. So, obtained lines can serve as sources for the selection of new varieties with increased resistance to winter, drought and fungal diseases, at the same time showing high productivity and quality of grain.

**IMPROVING ASSORTMENT OF PLUM (PRUNUS DOMESTICA L.) IN THE
REPUBLIC OF MOLDOVA DUE TO LOCAL BREEDING VARIETIES**

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Plum is the main stone fruit in Moldova. Plantations occupy an area of 32,000 hectares. As a result of continuous selection held about 200 combinations of crosses on a set of positive attributes and grown more than 17 thousand hybrid seedlings. 22 hybrid transferred to the State Cultivar Tests in 1987 and 2002 from 140 selected perspective forms. Plum varieties bred at the Research Institute of Horticulture and Scientific Practical Institute of Horticulture and Food Technology of the Republic of Moldova included in the prospectus of Plant Varieties of the Republic of Moldova in 1994-2014 years. The parent form for new sorts were the 10 varieties of world and Moldovan selection: Udlinionnaia (Tuleu Gras x Queen Victoria),

- Ranniaia Hramova (Tuleu Gras x Italian Prune)
- Pozdniaia Hramova (bud mutation Grand Duke)
- Pamiati Vavilova (Green Gage x Italian Prune)
- Crasa Oseni (Green Gage x Giant)
- Ajur-1 (Renclod Hramovix x Stanley)
- Super President (President x Sopernitsa).

New released varieties Moldovan selection of different ripening have many economically valuable traits, and high annual yield (15-25 t / ha), early entry into fruition (4 year), high quality and attractive appearance of fruits (fruit weight 50- 70 grams), of the complex resistance to diseases, undemanding to growing conditions.

Fruit varieties Ranniaia Hramova, Pamiati Vavilova, Crasa Oseni have a very elegant appearance and excellent dessert taste. Fruit varieties Udlinionnaia, Ajur-1, Super President universal use and highly transportable. Fruit varieties – Udlinionnaia, Crasa Oseni and Super President can be stored in a refrigerator at a $t=1-2^{\circ}\text{C}$ within 3-4 months, while maintaining normal appearance and high taste of dignity.

**HERBICIDE RESISTANCE BREEDING IN SUNFLOWER, CURRENT
SITUATION AND FUTURE DIRECTIONS**

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Sunflower is one of the most important oil crops in the world due to higher adaptation capability, mechanization use and preference by customers as vegetable oil. Sunflower areas are not larger in the world because the income is lower due to influenced more from environmental conditions as a summer crop. To increase production, it is important for decreasing of effects of factors reduced seed yield utilizing from higher production techniques in addition to develop higher yielding cultivars. Weeds and broomrape (*Orobancha cumana* Wallr.) exist among most limiting factors for sunflower production in especially in Eastern Europe and Black Sea Region which have more than 60% of world sunflower planted areas. After discovering the resistance genes against Imidazolinone (IMI) herbicide then transferring into cultural ones in US, Clearfield Technology which using post emergence IMI herbicides with IMI resistant cultivars has been used widely for 10 years. IMI herbicides (Imazamox (40 g/l)) control efficiently both broomrape and major broadleaf weeds such as *Xanthium strumarium* Wallr. *Chenopodium album* L., *Echinochloa crus-galli*, *Sinapsis arvensis* L., *Amaranthus* spp., *Solanum nigrum* L., *Datura stramonium* L. *Avena* spp. etc. resulting important yield losses in sunflower. On the hand, Sulfonyl Urea (SU) herbicide resistant sources also were developed with backcrossing method from same wild population in Kansas, US and then transferred into cultural types after discovering IMI genes. SU herbicides control more weeds and also cheaper than IMI are used widely in sunflower production in the world. However, SU resistant hybrids have the less control on both broomrape and some common weeds such as *Xanthium*, *Cirsium*, etc. so they should be combined with broomrape resistance together. On the other hand, new IMI and SU resistant genes also developed by chemical mutation then they are also available to use by sunflower breeders but it needs provisional contracts to use widely. Broomrape parasite develops new aggressive races historically against sunflower resistant genotypes. New races of broomrape such as F, G and H other than 5 known races (A, B, C, D and E) were observed in mostly in Balkan and Black Sea region and also in Spain. Therefore, new hybrids should have resistant genes against these new races or Clearfield technology. Farmers like this technology due to offering well control on both broomrape and also common weeds but they should wait until 6-8 leaves stage to apply IMI herbicide for efficient broomrape control. These delaying applications result sometimes not well control of already grown weeds. Therefore, combining broomrape resistant genes with IMI resistance in the same hybrid give farmers more options both for application time and amount depending on weed infestation in their fields. Additionally, seed companies also develop new tolerant hybrids every years mostly combining or adding new traits to broomrape tolerance such as downy mildew resistance as well as IMI herbicide resistance together because Clearfield system is one of the best and efficient option to control both broomrape and major broadleaf weeds. Now, sunflower hybrids combined these traits (IMI + Orb, Orb + SU) have started to sell recently and are preferred widely by sunflower growers. Therefore; in the future, broomrape and herbicide resistant hybrids combined all three traits (Orb + IMI + SU) will be developed with using IMI and SU resistant genetic material soon. These new hybrids combined these traits present more economical results to sunflower producers as reducing cost and increasing income per area with giving herbicide selection based on broomrape and weeds in their fields.

**SCREENING OF ACTIVATION-TAG ARABIDOPSIS MUTANTS WITH ALTERED
SUSCEPTIBILITY TO PHELIPANCHE RAMOSA AND P. MUTELII**

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The majority of species in family *Orobanchaceae* are facultative or obligate root parasites that subsist on broad-leaf plants, depleting them of nutrients, minerals and water. Several broomrape species parasitize important crops. They are widely spread in Bulgaria, Southern Europe, Russia, Middle East and Northern Africa and cause losses in crop productivity estimated at hundreds of millions of dollars annually.

The tiny seeds of broomrapes can support newly emerging seedling for very limited time. Therefore several mechanisms exist that insure tighter coordination between developmental stages of parasites' life cycle and the one of the host plants. The first critical step is germination. Seeds are capable of persisting for over a decade in the soil. For the germination to proceed, seeds require not only favorable abiotic conditions but an exposure of the right concentration of exogenous chemical xenogonins (usually named germination stimulants [GS]), that are emitted from the roots of host plants. Changes in the levels of synthesis and emission of GS can allow the development of practical measures for control of the crops-harming parasitic species. However, the genes encoding enzymes responsible for GS biosynthesis are still unknown. We performed a large-scale screening of 62,000 Arabidopsis activation-tag mutants for alteration in susceptibility to *Phelipanche ramosa* and *P. mutelii*. After five successive screenings we identified 36 lines with altered susceptibility to *Phelipanche ramosa* and *P. mutelii*. Seven of them displayed altered levels of GS production. By using a combination of Southern blot and thermal asymmetric interlaced polymerase chain reaction (TAIL-PCR), we pinpointed the location of activation-tag constructs in these lines. A combination of differential display and quantitative real-time PCR (qRT-PCR) allowed us to identify several affected genes. Two of them are directly involved in isoprenoid biosynthetic pathway in chloroplasts, and we believe that their activation led to increased levels of GS production.

EFFECTIVENESS OF GAMETOPHYTE SCREENING IN THE BREEDING OF LETTUCE SAMPLES WITH LOW CADMIUM ACCUMULATION

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Gamete selection is a prospective direction in the breeding on the resistance to heavy metals. Gamete selection has been used in the breeding of tomato forms resistant to heavy metals (Kilchevsky et al., 1993). Reaction of lettuce gametophyte in the F₁ progeny under cadmium provocation background and spectrum of progenies variability by resistance to cadmium accumulation have been studied. Materials of study were F_{1,3}-progenies of Novogodnii x Rubin hybrid combination of lettuce.

The treatment of lettuce flowers with water solution of cadmium was decreased significantly number of seeds in the ovary of F₁ plants. Number of seeds was decreased on 8-77% after treatment with water solution of low-cadmium concentration and on 45-123% after treatment with water solution of high-cadmium concentration. Trend of increasing of factorial effect from the beginning of blossom to the end of blossom has been observed. Analysis of cadmium accumulation in F₂ progenies show, that using of provocation with cadmium background is effective in the full blossom-period. The number of plants with high concentration of cadmium is decreased significantly (on 30%). The number of plants with low concentration of cadmium is increased on 25%. It is testified, that elimination of genotypes accumulated cadmium in leaves resulted in the treatment of flowers at the stage of blossom. Lowering of cadmium accumulation in F₂ progenies was about 28%. Effectiveness of gametophyte selection after flowers treatment was higher, than effectiveness of gametophyte selection after foliar treatment before 3 days to blossom (fig.1). Effect of screening is more obvious after treatment with water solution with high cadmium concentration. We haven't plants with high concentration of cadmium in leaves in F₂ progenies (fig.2). Comparison the number of seeds in the ovaries in the F₁ progeny under cadmium background with the level of cadmium accumulation in the F₂ progeny show, that lowering the number of seeds more, than 50%, was decreased the cadmium concentration on 19% as to standard. Backgrounds with cadmium concentration lower that 12,5 mg/l were not effective. Using of cadmium background in the period of blossom for the gametophyte screening in the F₁ progeny was decreased the number of plants have been studied in the F₂ progeny. The number of plants with low concentration of cadmium were decreased in F₃ progeny resulted in the treatment of flowers of F₁ progenies. Identification of lettuce forms, which will be stable by this trait (low accumulation of cadmium) will the object of our future investigation.

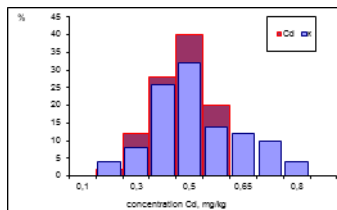


Fig.2. Distribution of lettuce plants by the level of cadmium accumulation in the F₂ progeny after the treatment of flowers with water solution of cadmium (Cd – 25mg/l). K-standard is water.

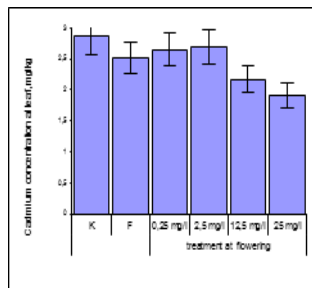


Fig.1. Concentration of cadmium in leaves of lettuce in F_{1,3} progenies on different cadmium backgrounds after the treatment with water solution of cadmium: F – leaves and flowers. K is standard.

EVALUATION OF CHICKPEA BREEDING MATERIAL (F₆-GENERATION) BY AGRONOMIC PARAMETERS AND SOME OF ITS BIOLOGICAL FEATURES

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Intraspecific crossing of initial (parental) forms: ♀MDI 02432 (seeds *desi*, black) and ♂ MDI 02419 (*kabuli*, beige) resulted in hybrid material, the offsprings of which have a rather high level of genetic variation.

Breeding material was evaluated in field experiment in 2014 using “pedigree” method. Individual selections were divided into 2 groups: with beige seeds (standard – *Ichel* cultivar) and pigmented ones (standard – *Botna* cultivar). Standards were sown after each 8 selections; comparative analysis of parameters was performed by their average values compared with 2 closest standards. Offsprings were evaluated by 5 agronomic traits: 1st and 2nd – productivity expressed as g/plant and g/m², 3rd – germinability (%), 4th and 5th – number of productive plants and their ratio to the number of seedlings (%). From total offsprings Δ (%) 31 families were distinguished by 5 traits as compared to standards and their values were at the level of standards or higher: C76, C90, C101, C152/c, C48/1, C48/2, C53, C187, C152, C156 and C131. The best values were noted in the family C48/28 and in 7 families from original family C53, exceeding standard by 44.6% and 21.2- 39.5%, respectively (brown *desi*-seeds). Average values of productivity 1 in experiment and the range for rows of *Ichel* cultivar were: 19,7 and 18,6-21,3 g/plant; for *Botna* cultivar – 18,8 and 16,8-20,5 g/plant. In selections with beige seeds the range for this parameter was: 17,4 (C76,6) - 25,0 (C90/6), for other selections: 15,7 (C187/2) - 23,3 (C53/6). Values of this parameter that exceeded standards by more than 10% were noted in 17 families (54,8%). Average values of productivity 2 in experiment and the range for rows of *Ichel* cultivar were: 171,2 and 165,6-178,3 g/m²; for *Botna* cultivar: 211,6 and 192,2-227,8 g/m². In selections with beige seeds the range for this parameter was: 160,9 (C76,6) - 193,3 (C152/c1), for others: 195,6 (C48/14) - 258,9 g/m² (C53/6). By germinability of seeds the following families were distinguished as exceeded the standards: 4 families from C53 - by 14,0-18,2%, C152/c1 (16,8%), and 2 families (C48/14 and C187/4) by 12,0%. Average value of germinability for *Ichel* cultivar was 87,1 \pm 1,2%; for *Botna* cultivar – 95,5 \pm 0,9%. 23 offsprings (74,2% of their total number) had germinability at the level of standards or higher. Values of parameter “number of productive plants” in 25 families (80,6%) were at the level of standards or higher. Values of parameter “number of productive plants and their ratio of the number of seedlings” in *Botna* cultivar and in selections were within 70,1% to 84,0% and 69,3% (C48/18) to 86,1% (C48/28), respectively. Values of this parameter in selections with beige seeds and *Ichel* cultivar were a shade less (66,3-73,5%).

It should be emphasized that manifestations of agronomic traits of genotypes are significantly influenced by their resistance to the main pathogens - *Ascochyta rabiei* and *Fusarium spp.* (*F.solani* prevailed). It was noted that selections of *desi* and *gulabi* groups (pigmented seeds) had higher resistance to both pathogens as compared to *gulabi* (beige seeds) and *kabuli*.

It is important to note that special features of breeding material tested include its difference from parental forms, formation and preservation of new traits: (1) pigmentation (brown, cherry, orange, beige - for *desi*), (2) seed type (pea-shape, 85,0% and 51,3% among beige seeds and other pigmented seeds, respectively), (3) wide spectrum of seed sizes (weight of 100 seeds): 23,4 g (C101/5) – 35,7 (C152/8), (parental forms: MDI 02432 had 29,1 g, MDI 02419 had 31,3 g; (4) maturation period: from 16th of July (C187 offspring) to 1st of August (C152/8, C152/9).

Evaluation of homo/heterogeneity of selected offsprings by tested traits revealed that higher homogeneity occurred in 2 parameters: maturation period (3 families were heterogeneous, all were C152 offsprings); seed type (4 were heterogeneous (*desi/gulabi*), with 2 from each C48/1 and C48/2). 9 families (29,0%) had segregation by pigment, and the highest heterogeneity was found in seed size: evenness at the level of standards and/or paternal forms was found in 11 selections (35,5%).

CHANGES IN THE NITRATE REDUCTASE AND PEROXIDASE ACTIVITY IN THE LEAVES AND LEAF APOPLAST OF SUGAR BEET PLANTS UNDER APPLICATIONS OF MICROCOM-T FERTILIZER, SUSPENSIONS AND METABOLITES OF MICROORGANISMS

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The purpose of the research - to reveal changes in nitrate reductase as well as peroxidase activity in leaves and leaf apoplast depending on seeds treated before sowing with fertilizer Microcom-T and suspensions of microorganisms (*Azotobacter chroococcum*, *Bacillus subtilis*) and under application of foliar fertilizer Microcom-T, metabolites of microorganisms and by the action of temporary water stress.

Soil moisture controlled conditions were created (in house experience vegetation) using Mitcerliih vessels with a capacity of 22 kg of soil. Soil - calcareous chemozem. Each vessel was administrated based fertilizers (NPK - 500 mg of active substance / kg of soil). Treatment of sugar beet seeds (hybrid Vilia) was performed by soaking seeds in 0.5% aqueous preparation Microcom-T for a period of 5 hours according to the proportion of seeds: solution as 2:1. Simultaneously, seeds treatment with suspension of microorganisms - *Azotobacter chroococcum*, *Bacillus subtilis* was carried out. The foliar treatment of plants with the metabolites of microorganisms was carried out in the intensive growth phase of the plants, and a temporary water stress (35% water capacity of the soil, WCS) - 7 - 10 days after the treatment with the complex of trace elements and metabolites of microorganisms. Field experiments - on experimental basis of the institute land. The area of experimental plot was 20 m², repetitions - 3. Activity of *in vivo* nitrate reductase in leaves was determined by Mulder method, peroxidase in leaves - by Boiarchin, leaf apoplast peroxidase enzyme extraction - by Brovenco and Riabischina, monosaccharide's and sucrose content - by Bertran.

Foliar treatment of plants with Microcom-T led to significant increase in leaf nitrate reductase activity both under optimal soil moisture and temporary water stress. Under seeds treatment with microelements the primary reducing process of nitrates in leaves increased compared to control but was less significant comparing to foliar treatment. Treatment effect was increased when applying fertilizer in complex with microorganisms, both under optimal soil moisture and soil moisture insufficiency (35% WCS). It should be mentioned that under water stress the nitrate reductase activity in leaves decreases significantly in virtually all variants, which confirms breakdown in nitrogen metabolism in such conditions. Therefore the reduction of nitrate reductase activity under unfavourable conditions of soil moisture is directed towards the improvement of biochemical pathways that are not crucial for survival in extreme conditions. Regulatory role of nitrate reductase in optimal growth conditions due to the application of trace elements and suspensions of microorganisms consists in fostering the reduction of nitrate in leaves. Simultaneously the leaf peroxidase activity decreased significantly both under optimal soil moisture and temporary water stress. Under seeds treatment with mentioned substances the decrease of peroxidase activity under optimal soil moisture was less significant compared to the variations subject to water stress. The content of monosaccharide's both under optimal soil moisture and temporary water stress under the application of Microcom-T, suspensions and metabolites of microorganisms decreased slightly. Simultaneously sucrose content tended to increase foliar application metabolites of microorganisms. These changes in carbohydrate content and enzyme activity were followed by increasing root mass and sugar content in them.

In field conditions the leaf nitrate reductase activity remained high under seeds treatment with complex of microelements and suspensions of microorganisms and the peroxidase activity usually tended to decrease compared to the control. Peroxidase activity in leaf apoplast of control variant was higher compared to variants with application of fertilizer and suspension of microorganisms and constituted 10.7% of the total activity of the enzyme. So the results show that changes in the activity of peroxidase in the apoplast are determined by physiological status of plants. On the background without application of basic fertilizers (NPK) seeds treated with Microcom-T separately increased productivity of crops stem with 14.6% and the application of the suspension of microorganisms - 10.9% (yield of crop roots in the control variant - 38,2t / ha, the sugar content - 22.6%). Increased harvest of crops roots under application of suspension of microorganisms is probably due to the growth of accessibility of plant nutrients and the use of Microcom-T - directly efficient supply of plants with microelements.

FACTORIAL ANALYSIS OF THE SPECIFICITY FUNGUS *ALTERNARIA ALTERNATA* (FR.) KEISSLER, ISOLATED FROM TOMATOES

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Lack of varieties with durable resistance is largely related to strong interactions of the plant x pathogen x environment. The three elements are in tight interdependence, forming a pathological system which determines the orientation and the character of the disease. One of the most widespread and damaging diseases encountered in recent times at the crop plants is alternariosis, caused by fungi of genus *Alternaria* Nees.

Alternaria presents a genus of fungi with numerous species, the strains of whom manifest saprophytism as well as parasitism. According to contemporary bibliographical sources, between the species *Alternaria*, *A. alternata* is manifest in recent times with frequency and high aggression, occupying the ecological niches or decrease area of extending of species *A. solani* (Ell. et Mart.) Sorauer which in many regions presents the basic species of tomato. The species *A. alternata* has a wide specialization, causing various diseases at a wide circle of plant species agricultural and technical: tomatoes, wheat, sorghum, barley, sunflower, rapeseed, cotton, etc., causing enormous economic losses.

A. alternata presents an important source of mycotoxins, such as: alternariol, alternariol monomethyl ether, tenuazonic acid, altertoxins, which have a pronounced specificity for the host plant, but also are dependent on environmental conditions.

In this context the purpose of the research was to elucidate the role of tomatoes, wheat and soybean factor genotype of at interaction with culture filtrates of the fungus *A. alternata*, isolated from tomato leaves.

The research was carried according to the model of two-factor analysis, in which the 3 cultures taxonomically distant – tomatoes, wheat and soybeans, each represented by 4 varieties were examined based on the reaction of plants to culture filtrates of *A. alternata*, isolated from tomato leaves with signs of ulceration or browning, and subjected to the ANOVA test (software package STATISTICA 7).

Seeds/beans were treated for 18 hours with culture filtrates (CF) of the 4 strains of *A. alternata*, and as a witness quality served the distilled water variant. The experience was performed at optimum temperature: 24-25°C (6 days) and low: 14-15°C (21 days). As test indices of the plants reaction, have served the important characters of growth and development at the early stage of ontogenesis – root and stem length.

It was found that the plants response to the 4 CF was differentiated, depending on the culture, genotype, character and isolated fungus, this falling in to the categories: lack of reaction, inhibition, stimulation. The weighting genotypic factor in the source variation of root growth constituted 88,9; 70,0; 34,0% and 89,9; 52,6; 53,9%, respectively, tomato crops, wheat and soybeans, on the background one of the optimal temperature and unfavorable. Regarding the role of genotypic factor in the variation source of stem growth, values to the constituted 80,4; 76,4; 17,8% and 86,0; 67,9; 66,4%, respectively, tomato crops, wheat and soybeans, on the background one of the optimal temperature and unfavorable.

The fact that the genotypic factor in reaction of seedlings the strains *A. alternata* (isolated from tomato) submitted higher values for tomato seedlings, reveals that the specificity of their reaction is more pronounced, compared to wheat and soybeans, and so – a possible specialization of *A. alternata* pathogen for tomatoes.

GENES ASSOCIATED WITH MICROSPORE EMBRYOGENESIS IN BARLEY (*HORDEUM VULGARE* L.)

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In the 21st century, cereals continue to constitute the most important crops with an annual output of 139.74 million tons (according to World Barley Production 2014/2015). In today's worldwide production, barley ranks fourth among cereals. Barley (*Hordeum vulgare* L.), first domesticated in the Near East, is a well-studied crop in terms of genetics, genomics, and breeding and qualifies as a model plant for *Triticeae* research. Advances made in barley genomics mainly include the following: a) rapid accumulation of EST sequence data, b) growing number of studies on transcriptome, proteome, and metabolome, c) new modeling techniques, d) availability of genome-wide knockout collections as well as efficient transformation techniques, and (e) genome sequencing effort (Nese Sreenivasulu et al., 2008).

In this paper I proposed to study the literature on genes associated with microspore embryogenesis.

According to the literature, the first genes identified in barley as being differentially expressed in the early stages of microspore embryogenesis encoding a lipid transfer protein (LTP), a glutathione S-transferase (GSTs) and a previously unknown protein (ECA1, possibly an arabino-galactan like protein). The GST family during the initial steps of microspore embryogenesis has been described by Vrinten et al. (1999) and Maraschin et al. (2006).

The availability of large sets of expressed sequence tags (ESTs) have triggered the development of macro- and microarray technology approaches that provide a rich source of information of the molecular processes that take place during microspore embryogenesis, specially at the microspore reprogramming phase (Maraschin et al. 2006; Muñoz-Amatriain et al. 2006).

The up-regulated transcription factors were divided into two groups according to the processes they regulated: abiotic and biotic stress responses (ICE1, ATAF2-like, WRKY46, AtbZIP60 and HSF7) and changes in developmental programmes (YABBY5, ZML2 and CURLY LEAF). The induction of CURLY LEAF, a repressor of floral homeotic genes (Goodrich et al. 1997; Katz et al. 2004) together with the downregulation of the ABC model genes *APETALA1*, *APETALA3*, *AGAMOUS* and *AGAMOUSLIKE 9* (*AGL9*).

The Affymetrix Barley1 GeneChip is a platform technology that contains at least 21,439 genes (Close et al. 2004) that facilitates extensive and globally comparable transcript profiling experiments in this crop. Two important classes of genes are those designated as hypothetical (i.e., those that have no reasonable homology-based functional annotation) and those that encode regulatory factors. The Barley1 GeneChip contains at least 6,789 and 1,059 probe sets representing hypothetical genes and regulatory factors, respectively. Using this resource, 1787 genes present on the Barley 1 GeneChip could be assigned to of the seven different chromosomes of barley of which (365 genes to 2H, 271 to 3H, 265 to 4H, 323 to 5H, 194 to 6H, and 369 to 7H) (Cho et al., 2006).

Platform Barley1 GeneChip offers a high degree of confidence, it has been tested on a technical level, extensive and was subsequently demonstrated to provide information rich data when used to investigate specific biological systems (Caldo et al. 2004).

An atlas of gene expression patterns that occur throughout the development of the barley plant was constructed with the aim of characterizing global expression patterns of each gene as well as determining the transcriptome characteristics that differ among different tissues during development (Druka et al. 2006).

In conclusion, YABBY5, ZML2, CURLY LEAF and ICE1 play a direct role in microspore reprogramming. Previously in barley, Maraschin et al. (2006) revealed that genes encoding an alcohol dehydrogenase 3 (*ADH3*), and proteolytic genes as a metalloprotease FtsH, a cysteine protease precursor, an aspartic protease, and a 26S proteasome regulatory subunit, could represent bio-markers for the embryogenic potential of microspores. These genes are involved in metabolic changes and proteolysis have a critical role in the dedifferentiation phase of microspore embryogenesis.

СОЗДАНИЕ СТЕРИЛЬНЫХ ФОРМ ТОМАТА ДЛЯ ГЕТЕРОЗИСНОЙ СЕЛЕКЦИИ

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Первостепенной задачей любой селекционной программы при получении гибридов F_1 , как для открытого грунта, так и защищенного является использование в качестве материнских форм стерильных линий. Создание таких линий возможно двумя способами: 1) скрещивание линий с разными типами стерильности и 2) скрещивание стерильных линий с фертильными формами. В связи с этим в исследования были включены образцы томата (6) с разными типами стерильности: лонгостилия, функциональная мужская стерильность типов Джон Бер и Врыбчанский низкий (ps), пыльцевая (ms-35, c) и тычиночная (pis). Скрещивание этих форм с линиями собственной селекции (J111, J112, J113 и J115) обладающих высокой комбинационной способностью явилось базой для получения новых стерильных линий. Морфологический анализ гибридных популяций ($F_2 - F_3$), разных гибридных комбинаций, полученных на основе вышеприведенных форм по целому ряду признаков репродуктивной системы: тип соцветия, тип цветения венчика, тип стерильности цветка – функциональная, пыльцевая выявил высокий спектр изменчивости по заявленным признакам. Степень стерильности, определяли путем подсчета процента завязавшихся плодов от свободного опыления, которое зависело не только от генотипа, но и в значительной степени от условий внешней среды (температура, движение потока воздуха и др.) в течение вегетации растений. Это позволило выделить полустерильные и высокостерильные формы томата. Выделена форма полудетерминантного типа роста, раннеспелая (100 дней), крупноплодная (150-240 гр). Анализ растений, выращенных из 20 семян, полученных в F_2 , показал, что в F_3 только на трёх растениях пыльники не растрескивались, и не происходило самопроизвольного завязывания плодов, хотя пыльца была высокожизнеспособна (51,3%). Из другой гибридной комбинации (детерминантной стерильной х детерминантной фертильной) выделены 4 растения детерминантного типа роста. При свободном опылении на двух растениях образовалось по два плода, третье растение имело 1 плод (5семян) на четвертом растении завязалось 3 плода, в каждом из которых было минимальное количество семян – 3, 2 и 5.

Необходимым условием использования этих форм в гетерозисной селекции является изучение взаимодополняющих при гибридизации морфобиологических признаков, благоприятных для выражения эффекта гетерозиса. Определяющими многие хозяйственные свойства, в частности продолжительность вегетационного периода, жизнестойкость и, как результат величину урожая являются архитектоника и габитус растения. Поэтому основной акцент при анализе гибридных комбинаций, полученных от скрещивания форм с разными типами стерильности, направлен на изучение образцов с полудетерминантным, детерминантным и супердетерминантными типами роста в сочетании со степенью выраженности определенного типа стерильности.

Анализ группы генотипов детерминантного и супердетерминантного типов (11), позволил выделить раннеспелые формы (3) с высотой главного стебля 50-65 см, средние (7)- и позднеспелых (1) 70-98 см. Выделенные образцы различаются по стеблеобразованию, что особенно ценно для гетерозисной селекции. Среднее число ветвей у раннеспелых генотипов варьировала от 2 до 4,7 шт., а габитус растений от 53,5 до 94 см. У средне- и позднеспелой группы, генотипы имели в среднем 3,5...7,5 штук стеблей на растение, габитус при этом составил 101 – 142 см. длина междоузлий у данной группы генотипов составила 3,5 – 5,0 см, что определяет сближенное расположение соцветий их число, а также общее количество сформировавшихся плодов в зависимости от яруса расположения кисти.

Изученные образцы (4) с полудетерминантным типом роста имеют длину главного стебля 110 – 138 см. Число ветвей насчитывается у них 5,5 – 6. Габитус растений характеризуется приподнятым-полустоячим стеблем и сильной облиственностью. Длина междоузлий имеет 5,5 – 6,0см. Окраска листьев темно-зеленая, с разной формой листа – яйцевидные, удлинено-яйцевидные, широколанцетные и с картофельным листом. Различия выявлены и по продолжительности вегетационного периода 100-109 дней.

Полученный исходный материал, различающийся по морфобиологическим характеристикам (тип стерильности, тип куста и срок созревания) отвечает поставленной задаче исследований и будет использован при создании гетерозисных гибридов F_1 .

USE OF *LS* GENE IN TOMATO BREEDING TO OBTAIN A NEW SOURCE MATERIAL

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Excessive formation of laterals shoots is a big problem when growing tomatoes in the greenhouse, which adversely affects the fruiting and increases labor costs for their removal, and consequently the cost of the products. In this regard, particular relevance is the task of creating new varieties and hybrids of tomato with weak laterals shoots development ability.

To obtain the desired results in this area of research, forms with the *ls* gene (lateral suppresser) characterized by the absence of side shoots, complete or partial absence of the corolla of a flower with partial male sterility, have been included in this study.

Three samples with *ls* gene were used in the cross. One line (L1751 American origin), represented by plants of cultural indeterminate type with short compact buds, fruits rounded shape with no green spot at the stem, red when ripe, weight 30-80gr. The flowers are typical of genotypes with the *ls* gene – without the corolla, with a long curved pestle. The second line (L 1169), obtained from professor S.I.Ignatova (VNIIO) is represented by plants of half cultivated type with a whole set of marker signs. Flower typical forms of a *ls* gene, without long corolla significantly protruding from the anther column pestle (ps). Laterals shoots on plants are absent or rarely appear one or two. The third mutant line (443 Mo), with *ls* gene manifestation, have been obtained from the collection of mutant tomato of the Institute of Genetics, Physiology and Plant Protection. It is characterized by determinant type of plants growth, no side shoots or individual self-limiting, reduced number of flowers in inflorescence, depressed corolla, often without lobes, partial male sterility.

A series of crosses of submitted forms with lines of varying laterals shoots development ability: L 828 and L 187 – weak, L 556 –medium and L 965 – very strong have been performed.

In the F₁ hybrids populations have been observed 100% domination of laterals shoots development capacity. But sprouts were in different sizes depending on the specific combination. In combination of *ls* gene forms and lines 828 and 187 with a weak laterals shoots development capacity during the growing season produced 2...5 sprouts at 18-25 knots. Fruiting in F₁ and F₂ hybrids combinations in the data combinations of L 1751 x L 828 and L 1169 x L 187 was more abundant, although the weight of the fruit was not entirely fit for purpose, and consisted 35 - 60gr only. In combination with the mutant line 443 and lines 828 and 187 laterals shoots have been absent and medium size fruit consist of 55 - 65gr.

In hybrid combination with the line 965, which is characterized by strong laterals shoots ability, there was a predominance of cultivated type with strong laterals shoots in each leaf axils. In these combinations in F₂ generation a strong splitting of this parameter and many other features have been observed. There were forms with signs of the reproductive system not peculiar to both the parent component.

In the hybrid combination L 1169 x L 965 dominated half cultivated type of plants, also with small fruits (30-50gr) and, in all nodes of plants evolved shoots, but they were very underdeveloped self-limiting growth, which is particularly valuable for the further work with these forms. In F₂ generation, plants without laterals shoots have been identified, but their fruiting were at very low level and a different number of sterile flowers was identified.

Strong variation observed in almost all cross combinations with the *ls* gene shape and morphology of the flower: without corolla, full corolla, but fused into a tube petals and unbreakable, with normal corolla.

From different hybrid combinations of studied hybrids, single cultural type plants with normal flowers and no lateral shoots or with single underdeveloped have been isolated.

The obtained results seems to be very interesting and will be studied in the next F₃ hybrid generation and further in order to select new forms with big fruits without or limited amount of laterals shoots and also sterile tomato forms with varying degrees of branching.

EFFECT OF INDUCED MUTAGENESIS WITH GAMMA RAYS ON GROUNDNUT

(Arachis hypogaea L.)

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Peanut, also known as ground nuts are consumed by many people around the world, being characterized by the impressed nutritive value. The high content of minerals like zinc, magnesium, copper, potassium or phosphorus, also fibers and antioxidants make peanuts a source of nutrients that can help the human body to remain healthy and fit. Today, the importance of peanuts increased a lot; they are one of the most important crops in the world, being used in food, in cosmetics and industry. Peanuts breeding are conducted according to the seed destination. Peanuts that are intended for oil extraction must have a debarking rate more than 72% and oil content more than 50%. For the fresh or roasted peanuts for human nutrition the content of oil should be less than 45% and weight more than 150gr /100 pods (Pop L.,1986). According to the agronomic particularities peanuts have reduced requirements to soil (good results on sandy soil), require smaller amounts of fertilizers, enriches the soil with nitrogen (above 100kg/ha) due to bacterial nodules on the roots. The countries with the highest number of population – China and India – together produce over 50% of the global peanuts harvest. The peanuts cultivation is carried out in warm regions, sunshine, with moderate rainfall, but also in temperate regions, which includes Moldova too.

In our country the peanuts are cultivated due to the high adaptability of this plant and the presence of early varieties homologated in Moldova (Fazenda I, Kovarschii 17, etc). Peanut can be considered as one of the most advantageous and cost-effective crop due to the production that can be obtained and multiples fields of use of obtained harvest. For this reason peanuts present a great interest for scientific researches performed at IGPPP, of the ASM. But climatic changes that are produced in last years at global and regional levels intensified the frequency of various climatic risk factors as: sudden changes of temperatures, a very long drought period, disastrous floods, short and warm winters, etc., which often diminish the value of the production. The main goal of research is to improve the quality of existing varieties, create early varieties, with high food productivity, high resistance to disease, pests, drought, and different climatic risk factors – which present a major problem of Moldovan agriculture. The experimental mutagenesis methods are successfully used in plant breeding for obtaining the valuable initial material. One of these methods is the gamma-ray-induced mutagenesis. The gamma radiation is useful in the induction of genetic variability, that presents a large spectrum of mutations and a high frequency of their manifestation. This method was used in our researches in order to obtain the valuable initial material for peanut breeding. The used biological material was peanut Fazenda I, approved in Moldova, that was treated with gamma rays doses of 100, 200 and 300 Gy. The result of gamma-ray-induced mutagenesis was the obtaining of peanut mutant forms with significant increase of oil and protein content. The biochemical analysis of allowed selecting the forms that are evidenced by a high oil content: from 56.57% to 54.15% (M101, M107), and protein: from 27.86% to 28.52% (M124, M91). The obtained results confirm the already known fact that the oil content in seeds is in negative correlation with protein content. It should be noted that in case of peanuts, the oil and protein content depends on the climatic conditions and is characterized by specific genotype variability for each variety. The analyses of the mass of 100 seeds of studied forms attested that characterized by high productivity. The mass varied from 51,2gr to 54,1gr (M91, M96), while in the control the mass was 50,8 gr. Thus, based on the above, it is clear that the weight varies between the forms and controls and largely depends on the climatic conditions of the year. The results of research in the field of gamma ray induced of mutations have allowed to determine the important features of mutational variability and by careful selection of peanut, there were obtained the mutant forms with high oil content, protein and productivity. The obtained forms represent a great interest for peanuts breeding and reproduction in Moldova.

**THE OPPORTUNITY OF USING VIRAL INFECTION IN INCREASING OF
VARIABILITY IN TOMATO VALUABLE TRAITS**

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The use of interspecific cross to obtain new forms of variability in tomato is a classic technique, and the application of additional factors to expand the variability is a useful method in breeding programs of cultivated plants. The purpose of this study was to analyze the possibility of using viral infection as recombinogenic factor in inducing variability of valuable traits in tomato interspecific hybrids compared with the traditional method. Five hybrid combinations obtained from crosses of cultivars Nistru and Novicioc with wild species were used: Nistru x *Solanum pimpinellifolium* (syn. *L. racemigerum*) - Ns28, Nistru x *S. pimpinellifolium* - Ns27, Nistru x *S. cheesmaniae* - Ns25, Novicioc x *S. pimpinellifolium* - Nw12, Novicioc x *S. pimpinellifolium* (syn. *L. racemigerum*) - Nw14. F₁ hybrid plants were inoculated with mixt infection - Tobacco Mosaic Virus and Potato Virus X (TMV + PVX). The seeds from F₁ uninfected plants served as control variant of F₂ generation, and the seeds formed on infected plants were used to obtain experience variants. F₂ plants were free of viruses. The measurements were carried out for each evaluated trait in about 150 plants per variant. Data were processed using the statistical software package Stagraphics Plus 2.1.

Assessment of distribution spectrum of values for analyzed traits in F₂ indicates a specific event based on genotype. At the same time differences for spectrum and frequency of traits values in descendants of infected and healthy plants were attested. In combination Ns27 the rate of plants accomplishing interested traits was always higher (up 1,33 to 2,47 times) in experimental population compared with the control one. Thus the number of fruits per plant, within 30-55 limits was attested for 18,22 % of plants in control population compared with 45,0 % in experience, and similarly for Soluble Solid Contents (SSC) in fruit, 6,4-7,8 g % was attested for 24,2% of plants in control variant and 32,11 % for the same range of trait SSC in experience. For the rest of combinations - Nw12, Nw14, Ns25 and Ns28, depending on traits, were attested cases of decrease as well as increase the frequency of phenotypes in experience variants compared to control ones. So, for the best fruit weight 50-110 g for Nw12 cross was attested for 13,3% and 44,81% of plants in control and experience, respectively. The earliest ripening forms were in Ns27 hybrid, but distributions of plants as well as the rate was different: within interval 103-110 days for ripening was for 66,06% of plants in control variant and earlier around 86-110 days at a rate of 83,47% plants in experience. Values of major content for SSC were certified in Ns28 cross - 6,8-8,3 g% in 26,71% of plants in the control and 6,8-7,8g% in 23,07% of plants in the experience. The hybrid combination Nw14 was found more attractive in result of analysis of fruit number and their rate. In the limit of 30-60 fruits per plants were attested 18,2 % of plants in control and in limit 30-70 fruits in 48,0% of plants in experience. Special interest presents a less elucidated in studies traits, namely shelf life. For one of the five combinations (Ns25) has registered a good storage of fruit under uncontrolled conditions of temperature and humidity up to 150 days without significant changes in the appearance of the fruit.

Differences in the average of fruit weight (increased or decreased) were confirmed in 4 of 5 hybrid combinations in the case of hybrid F₁ progeny populations derived from infected plant compared with the control.

The key moment for the selection of valuable plant forms, however, is the possibility of combining 2 or more traits of interest (large or small fruit weight, early or late ripening, increased number of fruits per plant, major content of SSC) within the same genotype. The incidence of such forms was higher in the offspring of the F₁ infected plants than control for 4 of 5 analyzed populations, although the spectra of traits combinations varied depending on genotype. Thus, the rate of selected F₂ plants that combines 2 or more valuable traits, was maxim in combination Nw14 and represent 8,85% in experience variant compared to 0% - missing of such plants, in the control one, followed by Ns27 cross - 5,36% in experience variant compared to 0% too in control, in Nw12 cross 4,92 % of valuable forms were in experience and 3,08% in control, in Ns28 cross 3,93% of valuable plants in experience compared with 0,8% and finely in Ns25 cross 4,4% of plants within experience compared to 0% - missing of plants in control variant.

The results show that descendants of virus infected F₁ interspecific hybrids differed after distribution and frequency spectrum of phenotypes compared to uninfected offspring, contributing to the emergence of phenotypic classes that were missing in the control population.

WINTER HARDINESS OF POLY-CROSS HYBRIDS F₁ LAVENDER (*LAVANDULA ANGUSTIFOLIA* MILL.)

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Lavender (*Lavandula angustifolia* Mill.) is the most "sensitive" culture to the changes of climatic parameters. The negative influence on this culture is low and extremely low winter temperatures. Exactly this fact explains the significant fluctuations in crop inflorescence and yield of lavender essential oil.

The purpose of this research phase was to determine winter hardiness and to select the polycross hybrids F₁, which are stable to low temperatures. During the researches in phase of the budding had been defined winter hardiness of the polycross hybrids F₁. To define winter hardiness of these hybrids were calculated died out skeletal branches and shoots on the bush on five-point scale.

Studying of the polycross hybrids showed that 36 perspective hybrids planted in 2007, the winter hardiness varies from 3,3 to 5,0 points. Five from these hybrids possess the highest winter resistance (5 points); these hybrids are Cr.26S-31; Cr.26S-56; Cr.26S-57; Cr.26S-58 and Cr.26S-130. The 12 hybrids F₁ from this group winter hardiness is above average 4,0 – 4,8 points. Representatives of this category are hybrids F₁ Cr.26S-18; Cr.26S-21; Cr.26S-44; Cr.26S-85. The middle winter resistance from 3,0 until 3,8 points is defined at 11 polycross hybrids F₁. For example, the hybrid Cr.26S-126 during 4 years the winter hardiness in middle makes 3,8 points. The same points of resistance have the following hybrids: Cr.26S-79, Cr.26S-73 and Cr.26S-66.

The hybrids Cr.13S-6-41, Fr.5S-8-16 and Fr.5S-8-24 planted in 2008 also have the highest points of winter hardiness. Other group of 12 hybrids planted in this year has 4,0 – 4,6 points and were noted with the resistance above average. Representatives of this group are hybrids Cr.26S-9-4 with 4,6 points and Cr.26S-9-6, Cr.26S-9-7, Cr.13S-6-12, Fr.5S-8-3 with 4,3 points of winter hardiness. The middle winter resistance was determinate at nine polycross hybrids F₁ planted in 2008, and these hybrids are Cr.26S-9-11 and Cr.13S-6-31 with the middle hardiness of 3,7 points.

After determination of the winter, hardiness at the polycross hybrids F₁ was established that level of the resistance and quantity of this kind of hybrids depends from mothers form. The acquired information demonstrates that the most resistant hybrids are hybrids that belong to Cr.26 mothers form. In this way, to this form concern five hybrids with high winter hardiness, 24 hybrids with above average hardiness and 12 with middle winter resistance. The mothers form Cr.13 has only three hybrids with high level of winter resistance, these hybrids are Cr.13S-6-41, Fr.S-8-16 and Fr.S-8-24. The most hybrids of mothers form Cr.13 and Fr.5 have the winter hardiness above average and middle.

After analysis of received information about the winter, hardiness of polycross hybrids F₁ was not established the hybrids with below an average or low level of winter resistance. The highest winter hardiness are determinate at 8 polycross hybrids Cr.26S-31; Cr.26S-56; Cr.26S-57; Cr.26S-58; Cr.26S-130; Cr.26-6-41; Fr.5S-8-16; Fr.5S-8-24 with the 5,0 points, 13 hybrids have the resistance varied between 4,0 – 4,8 points that is above average, and 21 hybrid has a middle winter resistance from 3,0 till 3,8 points.

For centuries, *Lavandula angustifolia* is used in folk medicine to treat a wide range of ailments. The essential oil from inflorescences of lavender is known for its antidepressant, analgesic, antispasmodic, antiseptic and healing action.

The ninety genotypes of *L. angustifolia* with remarkable quantitative traits were evaluated for essential oil content in the fresh inflorescences. The essential oil was separated by hydrodistillation using the Ginsberg apparatus. Seven hybrids showed the highest content of essential oil: 4,320 % - 5,790 % (dry matter).

Qualitative and quantitative composition of essential oil was determined by GC-MS. Have been identified from 15 to 25 components, depends on the polycross hybrids. Major components are linalool and linalyl acetate. The perspective polycross hybrid is Fr.5S-8-24 with 5,790 % (dry matter) essential oil content and the concentration of major components: linalyl acetate – 44,713% and linalool – 37,250%, followed by nerol, 3,899%; caryophyllene oxide, 3,595%; 4-terpineol, 1,718%; germacrene D, 1,260%. Other minor components in number of 16 are in a concentration from 0,737% to 0,157%.

METHODS AND GENETIC MECHANISMS THAT PROVIDE HIGH QUALITY HYBRID SEEDS OF MAIZE (ZEA MAYS L.)

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Maize is one of the most important agricultural crops at both national and global levels and holds the first place in crop production in the last ten years (according to FAOSTAT). This realization is based on implementing genetic accomplishments in maize culture – in the last 70 years it has switched from cultivating landraces and local populations of maize to applying the heterosis by growing corn hybrids.

One of the key moments in producing high quality hybrid corn seeds is maintaining genetic purity and assuring thorough hybrid pollination in seed producing sectors. In order to do so it is necessary to isolate the involved parental forms from other sources of pollination in space and/or time, to assure the conjunction in flowering of parental forms and assure the sterility of maternal form.

The sterility of maternal form can be provided by several methods, which can be grouped in 3 categories:

I) Physical methods:

- Manual castration - emasculation of tassels of maternal forms. Performed incorrectly, this method can affect the productivity of maternal form and requires significant expenditures of labor force.
- Mechanical castration – mechanical emasculation of tassels of maternal forms. Though it is widely used it has its flaws and can seriously damage the plant. The efficiency of this method depends of climatic conditions and the quality of seeding process.

II) Chemical method:

- Inducing male sterility by using chemical agents – it's a successful method in breeding autogamous plants, but can affect other vital plant systems.

III) Genetic methods:

- Cytoplasmic male sterility – the cms-Rf mechanism proved a viable method of producing high quality corn seed at a low price. Currently two types of cytoplasmic male sterility are used – cms M(S) and cms-C, that replaced Texas type (cms-T), vulnerable to disease (Helmintosporium maydis, T race), and that was used predominantly in hybrid corn seed production through the '70s of XXth century.
- Nuclear male sterility (*ms*) – the use of nuclear male sterility is complicated by the complexity of sterility inducing mechanism and its possible affinity with some mutations that affect valuable characters for crop production.
- Subsidiary genetic mechanisms, that include tassel-seed (*ts₂-ts_{2m}*), adherent (*ad*) and tassel-less mutations, that can affect directly (*ts₂-ts_{2m}* – by forming female pestles in tassel; *tls* – by gradual reduction of tassel and the amount of pollen) and indirectly (*ad* – by coalescing of superior leaves around the tassel) the male fertility of plant and that can serve as sources in creating a new mechanism of assuring the male sterility of maternal forms, used in the production of hybrid corn seed.

In conclusion: ensuring full male sterility of maternal forms on maize seed production sectors can be obtained by multiple methods, among them the most efficient would be based on applying genetic mechanisms.

AMELIORAREA PLANTELOR NECESITĂ AMELIORARE

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Ameliorarea culturilor agricole a contribuit cu peste 40-50% la sporirea productivității acestora, devenind un factor decisiv în eficientizarea agriculturii. Republica Moldova în anii 1980-2005 a avut realizări performante în ameliorare, producerea semințelor și materialului săditor la principalele culturi agricole. Actualmente în acest domeniu sunt mai multe probleme și aspecte, dar și soluții posibile. Am putea indica doar unele dintre acestea:

I. Nu este evaluat și exploatat pe deplin genofondul existent, fenomenul heterozisului. Culturile la care a fost studiat în profunzime și se exploatează fenomenul heterozisului, inclusiv androsterilitatea, se cultivă cu hibrizi. Timpul soiurilor tradiționale a trecut (inclusiv și la culturile autogame). Timpul hibrizilor actuali, tradiționali, trece... Vine timpul soiurilor hibride, cu multiplicare vegetativă, cu fixarea efectului maximal al heterozisului.

II. Majoritatea lucrărilor de ameliorare sunt orientate spre îmbunătățirea culturilor agricole existente. E necesar să fie create culturi noi, mai eficiente ca acumulate de energie, (în special culturi energetice), acumulate de hidrocarburi. Și această problemă are soluții în genetică și fiziologie.

III. Stimularea creației intelectuale în domeniul ameliorării este o problemă de cea mai mare actualitate. Cea mai bună soluție ar fi privatizarea creațiilor intelectuale, ele trebuie să aparțină autorilor, care trebuie să dețină supremația asupra creației intelectuale. Altfel ei nu o vor crea.

IV. OMG – reprezintă cea mai importantă creație în domeniu (genetica și ameliorarea plantelor). Nu-i bine să o ignorăm. UE deja a ridicat bariera, a deschis calea utilizării acestor realizări în Europa.

V. În ameliorare, dar și în genetică, e absolut necesar viziunea în profunzime, în dinamică și evoluția evenimentelor.

VI. Calea de la idee, de la creația cu valoare ameliorativă, la aplicarea acesteia – este prea lungă. Reducerea duratei acestor etape este de cea mai mare actualitate.

VII. Dotarea tehnică a procesului de ameliorare a plantelor, de producere a semințelor și materialului săditor este foarte slabă.

VIII. Finanțarea – Menținerea și studierea resurselor genetice, lucrările metodice și teoretice, studierea în profunzime a heterozisului și sterilității, pregătirea cadrelor – e necesar să fie finanțate din buget. Crearea noilor soiuri și hibrizi – parțial (doar primele etape) din buget, producerea de semințe și material săditor – din contul vânzărilor acestor produse.

IX. Posibilități și soluții de a depăși problemele din ameliorarea plantelor există în toate aspectele – organizatoric, metodic, genetic, tehnologic, informațional, socio-uman.

INHERITANCE OF SOME QUANTITATIVE TRAITS IN HYBRID POPULATIONS OF *CUPHEA*

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Creation of cultivars and hybrids combining high productivity and increased ecological sustainability, is one of the main approaches in agricultural crop breeding. As early as in 1960-s, the scientists faced the problem to find renewable sources of plant oils that can replace or supplement industrial oils. It was revealed that some plants of *Cuphea* genus (family: *Lythraceae*) can synthesize and accumulate oils in seeds (16 – 42%) containing, inter alia, medium-chain triglycerides. They serve as important raw material in production of the wide range of commercial products including soap and cleansing agents, personal hygiene products as well as nutraceuticals. At present researches are carried out aimed at acclimatization of *Cuphea* species and development of more productive and resistant forms. The most important stage of work on breeding of high productivity and resistance to unfavorable environmental factors includes identification of inheritance pattern of the main qualitative and quantitative traits. Purpose of these studies was to identify productivity inheritance pattern in two populations (F_1 and F_2) of hybrid combinations ♀MDI 02172 x ♀MDI 02166 (1), ♀MDI 02172 X ♀MDI 04085 (2) of *Cuphea viscosissima* Jacq. and *Cuphea lanceolata* Ait. species. Description of plants by morphological parameters revealed their intermediate inheritance in F_1 hybrids. Degree of dominance of quantitative traits varied widely. As a result of analysis of data on inheritance of the trait “plant height” we revealed negative dominance in the first combination and positive over-dominance in the second combination. The latter type of dominance was also noted for the trait “number of side shoots” and varied from 3.08 (1) to 4,2 (2). Negative dominance was revealed for such parameters of productivity as the number of flowers, fruits and seeds per plant. Positive over-dominance was noted for the 1000 seed weight that was equal to 37,0 (1) and 27,2 (2). Heterosis is commonly considered the superiority of hybrids over the average value of certain trait of parental forms. Positive heterosis manifested itself in the following parameters: “number of side shoots”, “number of leaves” and “1000 seed weight”. All hybrids had negative heterosis in productivity.

Quantitative traits in F_2 – population were evaluated and analyzed by the most important parameters – plant height, number of side shoots, number of flowers, fruits and seeds per plant. It was revealed that the average values of all tested traits in the first hybrid combination were distinguished by higher level as compared to parental forms. Thus, for example, the average number of seeds in hybrid plants was 247,7 flowers and the best parent had 165,0. The second hybrid combination was distinguished by higher values of traits: *number of side shoots, flowers and fruits per plant*. Average plant height was at the level of 83,01 cm and didn't differ significantly from the average value of the best parent (85,0 cm). Number of seeds per plant in F_2 – population was 378,33 seeds and the best parent had 586,2. In general, inheritance coefficient varied from low to medium. The first combination showed the best inheritance of plant height (60,1) and number of flowers per plant (71,6). The second hybrid combination showed intermediate inheritance of all tested traits.

Thus, the inheritance pattern of some quantitative traits was studied in *Cuphea* plants in hybrid populations F_1 and F_2 . In terms of quantitative traits the degree of dominance in F_1 hybrids varied from negative to positive over-dominance. Somatic heterosis was found in all hybrid combinations, and reproductive heterosis manifested itself in the parameter *1000 seed weight*. Analysis of segregation of the main quantitative traits revealed different pattern of their inheritance that depended on original forms included in hybridization.

ГЕНЕТИЧЕСКИЙ КОНТРОЛЬ ГЕТЕРОЗИСА У КУКУРУЗЫ: ОЦЕНКА СРЕДНЕЙ СТЕПЕНИ ДОМИНИРОВАНИЯ

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We propose an unbiased estimator for the average degree of dominance eliminating in average the linkage effect. The use of this estimator to the analysis of experimental data showed that the most likely cause of heterosis in maize is dominance, not overdominance.

До сих пор не сделан окончательный выбор между основными гипотезами, объясняющими гетерозис – гипотезой доминирования и гипотезой сверхдоминирования, и вопрос о генетических причинах гетерозиса по-прежнему остается открытым. Причина этого затруднения – искажение, вносимое сцеплением в генетические оценки. Чтобы выбрать между этими двумя гипотезами, надо оценить степень доминирования в генах, контролирующих гетерозис – превышает ли она единицу (сверхдоминантные взаимодействия) или нет (доминантные взаимодействия). Однако принятый в классическом анализе способ оценки средней степени доминирования по соотношению дисперсий (оценка Комстока-Робинсона) дает верный результат лишь для несцепленных генов. Применение этого способа к анализу экспериментальных данных обычно сопровождается оговорками, что полученное значение может быть завышенным из-за сцепления. Проблема искажающего влияния сцепления сохраняется и в анализе количественных признаков методом молекулярных маркеров.

Нами предложена несмещенная оценка для средней степени доминирования, которая в среднем элиминирует влияние сцепления:

$$\bar{D}_{\text{вер}} = \sqrt{\frac{\sigma^2(DIF) - \bar{\rho}(F_1 - MP)^2}{\sigma^2(SUM) - \bar{\rho}(\Delta P)^2}},$$

где F_1 – среднее значение признака у гибрида, MP – среднеродительское значение, ΔP – полуразность между родителями, $\bar{\rho}$ – неравновесие по сцеплению, равное в нашем случае 0,0543, SUM и DIF – функции $LxP_1 + LxP_2$ и $LxP_1 + LxP_2$, где L – компоненты выведенной из F_1 гетерогенной тестерной популяции.

От формулы Комстока-Робинсона наша формула отличается наличием членов, зависящих от F_1 , MP и ΔP . Она справедлива при случайном расположении генов, контролирующих значение количественного признака. При неслучайном расположении она может давать оценки завышенные или заниженные, но при массовом ее применении следует ожидать, что частота случаев завышения и занижения будет примерно одинаковой.

Мы применили эту оценку для анализа продуктивности трех гибридов кукурузы. Тестерными популяциями у нас были наборы удвоенных гаплоидных (DH) линий. Результаты в таблице. Для всех трех гибридов после поправки на сцепление средняя степень доминирования оказалась меньше единицы. Это означает, что со статистической точки зрения более вероятной причиной гетерозиса у кукурузы является доминирование, а не сверхдоминирование.

Таблица 1. Оценка средней степени доминирования в различных гибридах кукурузы

Гибрид	Год	Число DH-линий	Примечание	Продуктивность F1, г/раст.	Оценка средней степени доминирования	
					по Комстоку-Робинсону	с учетом сцепления
Rf7 x Ky123	2009	9	линии повторены	154	0,89	0,31
	2011	25		193	1,61	0,99
	2014	18		151	0,95	0,50
MK01 x A619	2010	27	18 общих линий	168	1,56	0,78
	2013	31		241	1,22	0,76
Rf7 x MK109	2014	9		140	1,35	0,73

STRATEGIES FOR CONTROL OF *OROBANCHE CUMANA* IN SUNFLOWER

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Orobanche cumana Wallr. is one of the most important constraints on sunflower production in all European sunflower-growing countries, as well as in the Middle East. It produces a large number of small seeds that are easily disseminated, leading to the buildup of *O. cumana* populations and the constant appearance of new, more virulent races. As the economic losses caused by this parasite are high, different measures, such as crop rotation and cultivation of herbicide resistant hybrids, have been used with more or less success, with the aim to limit its impact on sunflower crop. However, breeding for resistance proved to be the most economic and environmental friendly method for broomrape control. Hence achieving efficient and sustainable resistance to broomrape is currently one of the main goals of sunflower breeding programs. Nevertheless, sustainable resistance to broomrape cannot be achieved only by introduction of resistance genes into sunflower, but also by integration of other control measures and development of long-term strategies for broomrape management. This integrated approach to control of broomrape in sunflower is discussed in the paper.

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COMPLETAREA COLECIEI NAȚIONALE DE REFERINȚĂ LA PORUMB, IMPORTANTĂ ÎN TESTUL LA DISTINCTIVITATE

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Dreptul de autor asupra soiului de plantă în țara noastră este protejat prin Legea 39-xvi/2008, care prevede protecția juridică a tuturor varietăților „clar distincte”, „suficient de uniforme” și stabile la reproducere. În acest scop, fiecare varietate solicitată pentru protecție juridică necesită examinarea tehnică în câmp două cicluri de vegetație.

Procedura de examinare tehnică a varietăților este dirijată de Convenția UPOV, prin elaborarea și aprobarea Ghidurilor de testare la DISTINCTIVITATE, UNIFORMITATE și STABILITATE pentru mai mult de 7000 de taxoni botanici. Pentru cultura porumbului, este aprobat Ghidul TG/2/7 din 2009-04-01, care descrie particularitățile fiecărui test în parte.

Din testele recomandate la protecția juridică a soiului, testul la DISTINCTIVITATE este cel mai dificil, dar fiind faptul, că unele caractere necesită a fi evaluate prin măsurare, numărare, cântărire etc, altele – prin comparare cu soiuri de referință. Unele soiuri de referință, identificate ca marker/martor pentru o anumită stare a caracterului, sunt indicate în Ghid. O parte din ele sunt absente în colecțiile autohtone de porumb însă, acestea pot fi înlocuite cu mostre, selectate din colecțiile existente ale amelioratorilor. Reieșind din aceste considerente, IF „Porumbeni” menține o colecție de linii consangvinizate publice, cu termenul de protecție expirat, și soiuri populații, care pot fi utilizate în fondarea unei colecții naționale de referință pentru a ușura procesul de examinare tehnică a varietăților de porumb. În această ordine de idei, în ultimii ani, s-a inițiat un program de evaluare a expresiei caracteristicilor recomandate UPOV la un șir de linii consangvinizate și de acumulare a datelor, în vederea identificării markerilor, cel puțin pentru extremele de expresivitate a caracterelor.

Astfel, la etapa inițială au fost semănate linii consangvinizate cunoscute, menținute în colecția Institutului. Toate liniile au fost evaluate conform recomandărilor UPOV. Caracterele cantitative au fost evaluate prin măsurare/ numărare, iar media calculată a fost raportată la scara stărilor de expresivitate a caracterului, recomandat de UPOV. Starea de expresivitate a caracterelor calitative a fost înregistrată prin comparare cu starea caracterului la un soi de referință recomandat sau soiurile, cu cea mai joasă sau cea mai puternică expresivitate a caracterului evaluat.

În urma investigațiilor efectuate, datele obținute au fost sistematizate pentru caracterele: 8 "epoca de înflorire a paniculei" și 15 - "epoca mătășirii", dat fiind faptul că lista soiurilor recomandate de UPOV conține doar 2 mostre (A632 și B73) cunoscute și menținute în colecția institutului. Rezultatele obținute în ultimii 3 ani de testare, permit completarea colecției de referință cu mostrele: Z10, Co125, F2, W117 și A654, pentru înregistrarea stării de expresivitate a caracterelor 8 și 15, respectiv cu notele 1, 2, 3, 4.

Mostrele recomandate pentru diferențierea soiurilor la epoca înfloririi organelor reproductive au fost analizate și din punct de vedere al expresiei caracterelor ce țin de colorația antocian a diferitor părți/organe ale plantei (caracterele 1, 9, 10, 11, 16, 17, 19, 20, 41). Reieșind din datele acumulate pentru acest șir de mostre, colorația antocian a tecii primei frunzulițe (caracterul 1) a fost înregistrată cu nota 3 la liniile Co125 și A632 și nota 5 - Z10, F2, W117, A654, B73. Intensitatea colorației antocian a bazei glumelor pe panicul (caracterului 9) a fost înregistrată cu nota 1 (Co125, F2, A654 și B73), nota 3 (A632) și nota 9 (Z10), iar a glumelor paniculei exclusiv baza (caracterul 10) - cu nota 1 (A654), nota 3 (Co125, F2, W117, B73), nota 5 (A632) și nota 9 (Z10). Absența culorii antocian în antere (caracterul 11) a fost înregistrată la liniile Co125 și B73 cu nota 1, intensitate slabă, de nota 3 a fost observată la liniile Z10 și A632, iar intensitate medie, de nota 5, la linia W117. Referitor la intensitatea antocian în rădăcinile aeriene (caracterul 17), colecția recomandată UPOV poate fi completată cu linia A632, la care expresia caracterului observată este mijlocie (nota 5) și linia Z10, înregistrată cu cea mai înaltă stare de expresie, nota 9. În mostrele selectate pentru analiză, culoarea antocian în teaca frunzei mature (caracterul 19) sunt absente sau foarte slab evidente (nota 1) la 5 din 7 linii evaluate sau au o expresie slabă, de nota 3, la liniile W117 și A632. Referitor la intensitatea antocian în internodurile tulpinii (caracterul 20), în șirul analizat de linii n-au fost observate mostre lipsite de antocian (nota 1), iar la liniile A632 și B73 a fost înregistrată o expresie slabă a caracterului (de nota 3); la liniile Co125, W117 și A654 – medie (de nota 5); puternică, de nota 7, la linia F2 și foarte puternică, de nota 9, la linia Z10. Spre deosebire de alte caractere, în colecția sistematizată în rezultatul cercetărilor, expresivitatea antocian în glumele rahisului (caracterul 41) a fost la nivelul mostrelor, recomandate de UPOV. Cercetările inițiate vor continua pentru completarea colecției la caracterele menționate la toate gradele de expresie.

REALIZĂRI ÎN AMELIORAREA CULTURILOR DE SORG

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Conform Hotărârii Guvernului Republicii Moldova nr. 1262 din 01.11.2006 materialul biologic și finanțarea cercetărilor științifice în domeniul ameliorării culturilor de sorg au fost transferate din Institutul de Fitotehnie „Porumbeni” în gestiunea Institutului de Protecție a Plantelor și Agricultură Ecologică al AȘM. Din anul 2013 investigațiile nominalizate au fost continuate în Institutul de Genetică, Fiziologie și Protecție a Plantelor (IGFPP) al AȘM.

Obiectivul studiului în cadrul AȘM a constat în efectuarea cercetărilor experimentale și teoretice cu folosirea soiurilor (hibrizilor) omologate de sorg, surselor din colecții de linii consanguinizate și androsterile, descendențelor autopolenizate de sorg pentru boabe, sorg zaharat, sorg pentru măhuri și iarbă de Sudan. Investigațiile a fost direcționate: pentru menținerea genofondului, crearea materialului inițial de ameliorare, sintetizarea, încercarea și transmiterea în Comisia de Stat Pentru Testarea Soiurilor de Plante (CSPTSP) a hibrizilor competitivi de sorg boabe, sorg zaharat și sorg x iarbă de Sudan; înmulțirea seminței de categorii superioare a formelor parentale și de generația întâi; perfecționarea tehnologiei de cultivare și procesare a culturilor de sorg. Principalele obiective care au configurat tematica de ameliorare: precocizarea, sporirea capacității de producție, rezistența la polignire, secetă și arșiță, creșterea toleranței la atacul afidelor, ameliorarea calității boabelor și a masei vegetale.

În rezultatul investigațiilor efectuate pe parcursul anilor 2007-2014 au fost: evidențiate 65 linii de sorg boabe, 86 polenizatori de sorg zaharat, 30 forme de sorg măhuri și 6 linii androsterile; finisată crearea a 10 linii consanguinizate și 4 linii androsterile; evidențiați hibridi: 37 de sorg boabe și 48 de sorg zaharat; depozitate la Banca de Gene al IGFPP 1119 mostre de sorg; stabilită schema optimă de alternare a formelor parentale pe loturile semincere: 10 rânduri de formă mamă și 2 rânduri - formă tată; brevetat procedeul și instalația de uscarea a masei vegetale de plante pe rădăcini în câmp; adoptate 3 tipuri de uscătoare pentru uscarea seminței culturilor de sorg; elaborate 2 standarde de firmă și 1 instrucțiune tehnologică pentru utilizarea bobului de sorg în produse alimentare; transferați în CSPTSP și omologați în Republica Moldova hibridi de sorg: 2 de sorg zaharat, 1 de sorg boabe și 1 de sorg x iarbă de Sudan.

Dintre aceștia, hibridul **SAȘM 3** asigură obținerea recoltei de boabe la nivelul porumbului, dar având bob cu endospermul sticlos (asemănător cu orezul și respectiv numit **soriz** prin înbinarea cuvintelor Sorghum orizoydum) cu succes pot fi obținute produse similare cu acelea procesate din bob de orez: crupe întregi și brizate, griș, făină, amidon, bere, nutrețuri combinate etc. Rândamentul crupe, grișului și făinii din **soriz** atinge **75 – 82%**, pe când din orez este de **53 – 63%** și respectiv **1.5 – 1.7%** - a grișului din grâu. **Conținutul de proteine în aceste produse constituie 9,4 – 10,6 % (la orez 6,5 – 8,5 %), amidon 82 – 88%, grăsimi 0,2 – 0,5 %**. În colaborare cu savanții Universității Tehnice din Republica Moldova sunt elaborate tehnologii de fabricare din **soriz** a produselor **aglutenice** pentru bolnavii de **celiacă**: pâine, pandișpan, biscuiți, clătite, iaurt și băuturi combinate. Rândamentul de masă verde în condițiile Republicii Moldova a hibridului de sorg x iarbă de Sudan **SAȘM 4**, omologat în anul 2014, constituie 60-80 t/ha de la 2 coase. Al doilea cosit coincide cu deficitul de masă verde pe parcursul lunilor august-octombrie. Rezultate cu perspectivă sunt obținute în urma utilizării **heterozigotei Aa₁** în hibridarea sorgului zaharat prin metode tradiționale de ameliorare. Acest procedeu a permis crearea hibrizilor omologați **SAȘM 1** și **SAȘM 2** cu productivitatea biomasei de până la **80-90 t/ha** în condiții fără irigare și peste **130 – 140 t/ha** - la irigare. Hibridul de acest tip Porumbeni 4 în testarea internațională a sorgului zaharat a Uniunii Europene în anul 2008 s-a evidențiat ca lider după productivitate, acumulând recolta de biomasă de **184 t/ha**. Datorită **similarității** proprietăților morfologice (înălțimea plantei, grosimea tulpinii, particularitățile frunzei etc.) și agronomice, inclusiv și a utilizării biomasei plantelor hibrizilor respectivi, cu cea a **plantelor trestiei de zahar**, hibridii de acest tip au fost denumiți ca **sorg – trestie (sorgreed** în limba engleză), respectiv **sorggrid** în limba română. Procedeul de obținere a acestui tip de hibridi de sorg zaharat – **sorggrid** a fost brevetat în anul 2012.

Hibridii de **sorggrid** suportă suficient salinizarea solului, asimilează din atmosferă până la **50-55 t/ha CO₂** (pădurile de foioase pe latitudini temperate până la **16-18 t/ha/an CO₂**), sunt rezistenți la polignire și adaptivi la cultivarea și recoltarea mecanizată. Costul de producție a unei tone de biomasă de **sorggrid** cultivată, recoltată și transportată la 10 km, la stația de procesare, constituie **180/270 lei (11,92/17,87\$)** în prețurile anului 2014, în funcție de productivitatea plantației și nivelul de dezvoltare al gospodăriei.

EXPERIMENTAL ANALYSIS OF BIOENERGETICAL POTENTIALS OF MISCANTHUS IN MOLDOVA

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Limited capacity of the Earth to absorb ever increasing carbon dioxide emission directly linked with growing human activities in non-sustainable energy production via fossil carbon oxidation processes may (at some point in the future) trigger sudden climate change catastrophe which can be dangerous for survival of mankind. From other point of view the problem of self-sufficiency in energy production is an important economic issue for many countries in the world. Even those two considerations would be enough for stimulating scientific interests in plants useful for developing new sustainable ways of large-scale energy production. The logic of modern development required prompt and perfect answers on the wide range of questions essential for building and optimizing bioenergy industries which must be both economically sustainable and technologically suitable for preserving our ecological habitats.

After many years of systematic international research *Miscanthus x giganteus* was recognised as one of the most efficient bioenergy crops. It has many advantages: C4 plant with amazing ability of greater photosynthetic efficiency even under lower temperature and water conditions, lower nutritional requirements allowing using lower fertility soils, naturally originated (it is commercially open plant form) sterile hybrid propagating vegetatively via rhizomes etc. *Miscanthus x giganteus* rhizomes can be cost-efficiently planted during middle spring and the resultant established plantations can last for as long as 15-20 years. During late winter these plantations can contain up to 25 t/ha of well dried biomass suitable for cost-efficient harvesting and manufacturing high-density pellets which can be conveniently transported and stored. Subsequently these pellets can be used for heat and electricity generation in various power automatic devices depending on local energy requirements. At the same time *Miscanthus x giganteus* plantations can help us restoring soil fertility via carbon sequestration processes. Moreover the protecting effect of *Miscanthus x giganteus* plantations on natural biodiversity has also been shown.

The total scale of *Miscanthus x giganteus* plantations in EC is continuously growing together with expanding bioenergy industry. Apparently the fraction of *Miscanthus x giganteus* in total renewables is also growing and sustainably contributing to the total energy production.. Currently EC aiming achieving 20% of total energy production via usage of renewable resources.

Certainly, the progress in *Miscanthus* research field continues helping generating novel improved forms of *Miscanthus*. Several years ago EC provided financial support for the Optimisc Project aiming expanding testing of novel forms of *Miscanthus* including commercially used *Miscanthus x giganteus* in different geographical locations across several European countries. One expected outcome of Optimisc project would be the model predicting the distribution of productivities of *Miscanthus x giganteus* and novel forms of *Miscanthus* in changing climate conditions in any geographical location in Europe supporting expanding bioenergy industries.

The wealth of previous experiences in *Miscanthus* field accumulated in UK, the advantages of adding more geographical locations with different climate conditions for developing more accurate model predicting *Miscanthus* productivities as well as new tantalising potentials for developing bioenergy industry in Moldova were taking into serious considerations before initiating long-term scientific collaboration between British and Moldovan scientists starting at the beginning of 2014. at the present we need more time before starting discussing our first experimental results, therefore we are looking forward to systematic analysing of bioenergetical potentials of *Miscanthus x giganteus* and several novel forms of *Miscanthus* grown under climatic conditions of Kishinev in Moldova. The obtained data will be included into the general model allowing predicting the productivities of *Miscanthus* in Europe. This project has good potentials for developing new international collaborations and future business opportunities in the field of bioenergetics in Moldova.

INBRED LINES OF EARLY MAIZE: GERMPLASM, EVALUATION AND CLASSIFICATION

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Basically, the major focus of maize breeding programs is the development of new inbred lines having valuable agronomic properties and characters as parents of hybrids. The spreading of earlier flowering single-cross hybrids in shorter-season growing areas has been made by improvement of lines. Modern inbred's development is characterized by some new requirements for germplasm of basic material, evaluation of lines per se and selection of progenies with highly yielding ability. This report intend to made a review of the present state of research in a long term breeding programme of maize for export in northern areas of cultivation.

The basic material in 1970's years were represented by early hybrids created in different companies of France, Germany and USA. From 461 breeding sources only 12 (2,6%) were useful in inbred development, including two USA dent hybrids from which originated the first inbreds used in some registered three way and double cross hybrids. At the next stage the main basic material were planned crosses involved germplasm of 79 public inbreds from 21 heterotic groups and subgroups. In the last 15 years single-cross hybrids, related crosses and one backcross sources are the most frequently used basic material. More strictly, attention is given to maintaining known heterotic patterns in planned crosses and to general combining ability of involved inbreds. The testing program of hybrids generated the information that the best heterotic patterns for early maize are crosses of Reid Iodent germplasm as seed parent with Euroflint, BSSS-B37 and Lancaster groups of germplasm as male component. From this reason in present in inbred development are included the best sister/related lines from each germplasm group. For increasing of genetic diversity in heterotic groups, BSSS-B37 and Lancaster in some basic material are introduced favorable alleles from indentified new germplasm sources.

Phenotypic selection during the inbreeding were effective for good germination, vigorous seedling and juvenile plant grow, simultaneous pollen shedding and sulking, heat and drought tolerance, leaf and ear diseases tolerance, plant and ear height, stalk lodging, stay-green, kernel moisture and texture, grain yield and other agronomic characters. For a better discrimination of progenies has used high plant density (more than twice normal), extra early and late planting dates, artificial inoculation with *Sporisorium reilianum* and *Ustilago maydis*. Late generations of inbreeding (S_4 - S_5) are evaluated for combining ability with testers from alternative heterotic groups. Results from our experiments suggested that inbred grain yield correlated with test cross yield ($r=0,464$ in S_3 , $r=0,599$ in S_4 and $r=0,719$ in S_5) and selection of high yielding progenies per se is a efficient procedure. The best progenies are transferred in operational collection for multiplication and a more accurate evaluation in testing trails for cold tolerance, grain yield and moisture, reaction to cytoplasmatic male sterility. This information permits to appreciate the preferable position (female or male parent) in hybrid combinations. During 1982-2014 years the operational collection included more than 200 original inbred lines from which 30 have been used as parents in advanced testing, transfered for official state testing and registered hybrids. These lines are from heterotic groups: Euroflint – 8, Reid Iodent – 8, Dent Canadian – 7, BSSS-B37 – 5 and two with Lancaster germplasm. The new inbreds realized in the last ten years are significant more performing then first commercial lines, in special from Reid Iodent heterotic group. A visible improvement of 3-4 breeding cycle lines was obtained for stalk lodging associated with stay-green plants, lower grain moisture at harvest and general combining ability. In our experiments general combining ability (resulted from cumulative, additive gene effects) was more important than specific combining ability.

The classification of related inbreds into heterotic groups and the development of hybrids into heterotic patterns increase efficiency of breeding program. Heterotic (germplasm) groups are identified using pedigree dates, UPOV phenotypic descriptors and grain yield in test crosses transferred into index of heterosis (H) and index of genetic diversity (DG). The last procedure was more accurate and permits to establish the genetical distance between related inbreds which is very important for substitution of weakest inbred in popular single-cross hybrids or improvement of seed parent using related crosses. Original lines have been used as parents of 17 registered and 5 new hybrids.

VIABILITY IN TIME OF SEEDS OF SOME AROMATIC AND MEDICINAL PLANTS

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Moldova was and remains an essential area to growing medicinal and aromatic plants. Their cultivation is closely linked to the production of seeds that are central in production technologies. At some species not all years are favorable for obtaining seeds, at others seeds have seminal prolonged rest and are viable only 1-2 years after harvest. Therefore creating seed reserves of some species is strictly necessary. In this connection was studied dynamics of seed germination while the new species found during the implementation in industrial crops: Savory, Passiflora and Dragonhead.

As objects of study were seeds of savory Alfa-14 variety, of the Dragonhead Aroma-1 variety and of Passiflora variety. The seeds were produced in seed lots in the period 2010-2014 and were kept in dark storage in packages (bags) of paper craft.

Germination was determined on the plates Petri, on filter paper in the dark, in thermostat TS-80 at a temperature of 24-26°C in accordance with the recommendations (L.S. Muntean, 1996). The energy of germination was determined in 5-7 days and total germination - over 15 to 21 days. The mass of 1000 grains (seeds) was determined in three repetitions for each segment of seeds dried at natural moisture of conservation. Germination was determined repeatedly in time, in the analysis using average values.

It was determined that Savory (*Satureja montana* L.) in plantations (III-VIII year of vegetation) at a density of 10,000 shrubs/ha achieved production of viable seeds of 40-50 kg/ha.

At the Alfa-14 variety seeds are formed beginning from second half of September. Seeds are small - mass is 0,3-0,4 g per 1000 units. In harvest year seed have low germination (up to 20%), they being in rest period. Over 1 year after harvest the seed germination is 63%, 64% over two years, 65% over 3 years, 53% over 4 years, 44% over 5 years. So it can be concluded, that the seeds of savory keeps germination up to 5 years, with a maximum value of 65%.

Passiflora variety (*Passiflora incarnata* L.) being of subtropical origin was introduced in culture field in the Republic of Moldova, form mature fruits with viable seeds only if is multiplied by seedling grown on semi-protected land (in greenhouses) providing a total period of vegetation 200-210 days. Passiflora has an indeterminate growth. By October on the creepers of Passiflora persist fruits with different degrees of maturity, many with baked crumb, but with white, brown and black seeds. It was established that the black or dark brown seeds are better matured and the mass of 1000 grain is almost 32-35 g and germination of 86% in the first year after the harvest; 79% in the 2nd; 43% in the 3rd, 39% in the 4th year, and 34% in the 5th year of storage.

Brown seeds of Passiflora have MMB 27-28 g; germination of 50% over 1 year after harvest; 49% over 2 years; 39% over 3 years; 35% over 4 years and 31% over 5 years after harvest.

White seeds have MMB of 18,5 to 19,5 g; germination of 34% over 1 year after harvest; 30% over 2 years; 5% over 3 years; 0% over 4 years of storage.

Weight of 1000 grains correlates with the degree of maturation of seeds, black is being the best. If the mature fruits froze in early wintering seeds fully lose their germination.

So good seed germination at Passiflora begins in the first year after harvest and maintain during 3 years. Acceptable for use are black and dark brown seeds. Passiflora seed productivity under the conditions of our country is 300-400 kg/ha.

Dragonhead Moldavian (*Dracocephalum moldavica* L.) semi-early Aroma-1 variety has a vegetation period (from germination to seed maturation) of 120-125 days.

The plants bloom and form seeds staggered and at the time of seed maturation in the basal part of the inflorescence but the tip persist blossoms.

Being harvested at the mature time of seeds mass production Dragonhead provide a yield of 400-500 kg/ha of brown seed with MMB 1,4-1,6 g.

The seeds have not a pronounced seminal rest. In the first year after harvesting seed germination is 91%; 86% over 2 years; over 3 years - 82%; over 4 years - 71%; over 5 years - 53%.

It was also found that moisture conservation at Passiflora seed storage is 7,4-7,8%, at the savory from 8,5 to 9,0%, and at Dragonhead from 8,0 to 8,3%.

THE NEW VARIETIES OF AROMATIC PLANTS

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Cultivation of medicinal and aromatic plants in the Republic of Moldova is in the process of recovery, the areas occupied by them reaches to 4500-5000 ha. Along industrial crops – clary sage and lavender, more and more are requested menthol mint, savory, fennel and oregano. Based on market demands and the available scientific achievements over the last years have been created and approved new varieties of mint, mountain savory, and fennel.

There were created and included in the Catalogue of Plant Varieties of the Republic of Moldova new variety of mint *UsIgen* (authors Musteatsa Gr., Ceban V., Pisov M., Baranova N., Timciuc C.).

Variety is created by individual selection as hybrid mutant of *Mentha incana* Willd x *Mentha sachalinensis* (Brig) Kudo. The plant is herbaceous, perennial, forms shrub with branches of the package, but with pronounced dominant central stem, erect, 70-80 cm at flowering, under conditions of irrigation and fertilizer application. It has large leaves (4-6 cm) of green-grey color. Beam roots reach deep into the soil (up to 80 cm). Rhizomes of UsIgen variety plants are well developed, white-green, 5 to 6,5 mm thick, placed deep in the soil (0-8 cm), which makes plants resistant to winter. Yield of rhizomes in irrigation condition is 8-10 t/ha. Variety is multiplied by seedling and rhizomes, and can be grown 2-3 years to irrigation.

Variety is well budded with high content of essential oil in leaves (4,884%) and highly productive: aromatic feedstock production – 9,6 t/ha; pharmaceutical dry leaves 1,49 t/ha; volatile oil 68,2 kg/ha in the first year of vegetation, 92,7 kg/ha in the second year of vegetation. Free menthol content in volatile oil is 50-55%, of total menthol – 58-63%. The production of this variety can be used as pharmaceutical dry leaves and essential oil production.

Variety of Perennial savory (*Satureja montana* L.) *Alfa-14* was created by individual and mass selection of local improved population (authors Timciuc C., Musteatsa Gr., Jelezneac T., Vornicu Z., Dragalin L.).

It is an undergrowth shrub with height 45-60 cm semispherical, semi compact, consisting of 120-250 semi erect stems and arched, with annual shoots 32-36 cm long. Sessile leaves on annual shoots are green and length 12-18 mm. The flowers are small with white corolla color with pink shades, grouped by 4-8. Fruits are small nut, dark-brown, mass range 0,3-0,4 g per 1000 seeds. The seeds productivity is 40-50 kg/ha.

Variety is adopted to environmental conditions of Moldova, has high quality production: production of raw materials - 9,0 t / ha, volatile oil - 54,6 kg / ha, volatile oil content of 0,609% in fresh raw materials, the content of phenolic compounds (carvacrol + thymol) reaches 70-80%. The final output may be used as a volatile oil, and as pharmaceutical herb.

Fennel *Peren-1* is new variety, developed at the Institute of Genetics, Physiology and Plant Protection in collaboration with the Botanical Garden (Institute) of ASM (authors: Musteatsa Gr., Chisnicean L., Teleutza A., Rosca N., Vornicu Z.).

It is a sort of perennial plants with lifespan of sowing of 3 years, vegetation period of 125-128 days to produce raw materials (plants) and 145-148 days to obtain seeds (fruits). Mature plants have size 91±3 cm in the first year of vegetation and 132±7 cm in subsequent years.

The variety is highly productive: the production of raw materials (plants) - 11,8 t/ha, fruits (seeds) – 1,1 t/ha, volatile oil 93,9 kg/ha, content of trans-anethole reaches 79,8 % and content of fenchone -13,9%. The variety can also be used for the production of essential oil and seeds with high pharmaceutical properties.

The mentioned varieties are implemented in production. The raw material (dry herba) of Perennial savory, mint and fennel fruits of the mentioned varieties as well as raw material of Dragonhead of Aroma-1 and Passiflora are widely used by "Doctor Farm" firm as the main components in the production of food supplements like Multivitamin Tea, Tea for men, Evening Tea, Energizing Tea, Calmo-plus, Imuno-plus and other natural products.

MOLECULAR MARKER APPLYING FOR SWEET PEPPER BREEDING
(*CAPSICUM ANNUUM* L.)

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Sweet pepper (*Capsicum annuum* L.) – one of the most important natural sources of minerals, vitamins and antioxidants, including pepper fruit pigments as the most important ones. The major carotenoids of the pepper fruits are carotene, capsanthin, capsorubin, lycopene, lutein, zeaxanthin, violaxanthin, xanthophylls and other. Carotenoids are the most widespread pigments in photosynthetic organisms. They act as photo-protectors and accessory pigments in photosynthesis, and play an important role for pollinator attraction and seed spread. Carotenoids also can exhibit antioxidant activity, and some of them are the precursors of vitamin A.

Ripe fruit of *Capsicum* can be classified into eight classes by colors, ranging from white to deep red. Color variation seems to be determined by three independent gene loci, *Y*, *C1*, and *C2* (Hurtado-Hernandez and Smith model). The gene capsanthin-capsorubin synthase (*Ccs*), which is involved in antheraxanthin to capsanthin and violaxanthin to capsorubin conversion, is considered the candidate gene for the *Y* locus, at the same time the *C2* locus corresponds to the *Psy* gene. The identity of the *C1* locus remains unknown. *C2* and *C1* regulate the amount of carotenoids rather than type. *CrtL-b* gene encodes lycopene β -cyclase that converts lycopene into β -carotene and δ -carotene, therefore decreased activity *CrtL-b* causes reduction in the amount of β -carotene, xanthophyll or other carotenoids. Mutation of gene *chlorophyll retainer* (*cl*) of pepper inhibits the ability to degrade chlorophyll during ripening. It leads to the production of ripe fruits characterized by both chlorophyll and carotenoids accumulation. Brown fruits color is determined by combination of allele Y^+ and *cl* mutation, and yellow-green fruit color is determined by combination of homozygous recessive allele Y^- .

High biological value of pepper fruit is determines by development and introduction of hybrids with a complex of fruit quality traits. So, it is of great practical interest to combine all favorable genes in one genotype. In this case, the traits features are searched in breeding genepool, often by DNA markers that is linked to selected characteristics.

The aim of this study was to screen the collected gene pool of sweet pepper for favorable alleles for determining high carotenoids content. Plant material (27 varieties of fruits varying in shape and color) was obtained from «All Russian Research Institute of Vegetable Breeding and Seed Production of Russian Academy of Agricultural Science» and from our sweet pepper collection.

Specific primers for gene *Ccs* amplified a fragment of 1470 bp in length in red fruit sweet paper varieties (L-25311, Igrok, Pregol', Otello, L-510, Pfiolotoviy Krasavec, Belosnezhka, L-208, L-255-11, Cherniy Krasavec, Mayak, Shokoladnaya Krasavica, L-24, ZongKao and L-160-10), that is consistent with the reported data and confirms the presence of an active form of capsanthin-capsorubin synthase (*Ccs*). This enzyme activity was not detected in sweet paper varieties with yellow and orange fruits. Products 2844 bp and 1534 bp are synthesized during the amplification with primers for genes *Psy* and *CrtL-b* respectively. There fragments were found in all collection genotypes, indicating the presence of phytoene synthase and lycopene β -cyclase genes in their genomes. Y. Borovsky reported dCAPS marker for *cl* gene associated with dark color of pepper fruits. However, the region of the marker contains two restriction sites for *FokI*, one of which is undesirable. Optimization helped to find an effective pair of primers giving a product of about 200 bp. The wild type gene *cl* contains *FokI* restriction site while *cl* mutant is insensitive for digestion, that facilitates identification of favorable alleles. Screening the gene pool for *cl* markers identified four varieties containing *cl* mutant allele (L-236/09, Sladkiy Shokolad, Shokoladnaya Krasavica, L-45-11). Three of the genotypes are brown and L-45-11 is yellow-green when ripe. In three varieties (Otello, Pfiolotoviy Krasavec and Cherniy Krasavec) the dark fruit color corresponds to technical stage and cherry, red and yellow does to biological stage. However, we did not identify *cl* allele in these varieties.

Thus, we optimized the methods of identification of genes involved in carotenoids synthesis regulation and tested our DNA-techniques in 27 sweet pepper varieties with different fruit color. We identified genotypes with favorable carotenoids traits and its combinations. Based on the findings of our study, we propose applying the MAS for development of varieties and hybrids with high fruit quality.

MOLECULAR-GENETIC APPROACHES TO IDENTIFICATION OF MAIZE GENOTYPES WITH IMPROVED QUALITY PROTEINS

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Currently, more than 70% of maize is used for food and feed, therefore grain quality improvement can increase nutrition and energy value of a given culture. Deficiency of two essential amino acids (lysine and tryptophan) significantly reduces the nutritional quality of maize proteins. However, compared to conventional varieties maize *opaque 2* (*o2*) mutants contain greater levels of lysine and tryptophan in the endosperm and this increases maize proteins bioavailability. The aim of the study was an identification of maize accessions with a high quality protein with using of molecular-genetic techniques. *Opaque-2* gene diversity was studied with 3 pairs of fluorescent labeled primers to the *phi112*, *umc1066*, and *phi057* sites. Primers sequence and reaction conditions were taken from Yang et al. 2004. Extraction and urea-PAGE separation of zein proteins were performed according to GOST for maize seeds.

54 maize genotypes with different endosperm phenotypes were selected for diversity analysis of 3 SSR sites. *Phi112* SSR is located in the 3' leader sequence, and its mutation can affect transcription of the *O2* gene. Our results indicate the presence of at least 3 alleles of *phi112* – 145 bp, 151 bp and recessive (*o2*) zero allele with no PCR-product. Favorable recessive *o2* state of *phi112* was revealed only for 8 maize accessions. The *umc1066* and *phi057* SSRs are located in exon 1 and exon 6, respectively, and were reported as hypervariable regions of *O2* gene. Recessive alleles at the *umc1066* and *phi057* SSR sites have a length of 161 bp and 143 bp respectively. Only 22 of 54 tested maize accessions amplified bands of that length. However, the majority of *umc1066* and *phi057* alleles were in heterozygous. Only 8 maize accessions were homozygous recessive (*o2o2*). Results obtained for allelic states (*o2/O2*) of all three SSR sites corresponds each other, suggested some extent of linkage among them.

Quality Protein Maize (QPM) apart of *o2* mutation contain *o2* modifier genes that convert soft, floury endosperm of mutants to vitreous phenotype. Although the genetic basis of the conversion is poorly understood, the degree of endosperm vitreousness in QPM is associated with the variations of zein fraction of seed storage protein. It is known that QPM kernels have low levels of α -zein and increased concentration of 27-kDa γ -zein protein. So we used Urea PAGE method for identification of QPM genotypes. In PAGE-patterns of opaque phenotype accessions was observed reduction of 22-kDa α -zein accumulation. QPM patterns exhibited both 22-kDa α -zein reduction and increases in 27-kDa γ -zein protein band.

In summary, the allelic variations of the *umc1066*, the *phi057* and the *phi112* sites within *O2* gene include the variations of the repeat number of SSR motifs (different fragment length for all primers) and variations in the regions flanking SSR (absence of product for *phi112*). It is best to use the 3 markers together in molecular marker-assisted selection for high-lysine maize materials. Urea PAGE of zein protein fraction is effective for segregating of QPM genotypes. Selected QPM accessions are of interest for maize breeding programs aimed to grain quality improvement. The use of the markers to *o2* and modifier genes reduces time for developing of QPM maize varieties, as well as significantly reduces the labor and financial costs of their production.

CURRENT STRATEGIES IN SUNFLOWER BREEDING

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Sunflower served as a decorative plant, long time after its arrival in Europe (in the 16th century).

Large - scale use of sunflower as an oil seed crop started in Russia in the 1830s.

Sunflower selection begun simultaneously with its expansion as an oil crop. Around the end of the 19th century, farmer's selection was practiced and it was aimed at improving the available sunflower populations.

The beginning of scientifically grounded sunflower breeding date to 1912, when a program of varietal development was established. Breeding of varieties was started in Kruglik, Kharkov and Saratov stations, in the same year. First methods applied in sunflower breeding were mass and individual selection for specific characteristics.

Under the leadership of V.S. Pustovoit, a method was developed for obtaining high-yielding varietal populations based on individual selection and seed reserve that is used through the selection cycle. Using this method, the oil content in seed was increased from 36% to 52% and resistance to broomrape and some diseases was incorporated into sunflower varieties.

Greatly due to the development of Russian high oil sunflower varieties were developed in a short time, many varieties in several research centers in Europe (Romania, Bulgaria, Serbia, France).

Another step in sunflower breeding was made by G. Pustovoit when it was introduced interspecific hybridization (crossing *H. tuberosus* with cultivated sunflower).

Extensive genetic studies on sunflower inbreeding and heterosis were conducted in the second half of the 20th century by a large number of researchers.

Some sources of nuclear male sterility played a brief role in the production of hybrid sunflower but, practical utilization of heterosis in sunflower begun with the discovery of a stable source of cytoplasmic male sterility, discovered by Leclercq in 1969, in the wild sunflower *H. petiolaris*.

After the discovery of the first cms and first restorer gene (by Kinman, 1970), a large number of breeding centers for developing sunflower hybrids were established.

To be able to define optimal breeding activity for developing new sunflower hybrids is needed to have a certain level of knowledge in genetics and breeding, in addition, modern breeding needs an adequate knowledge of allied disciplines of biology and more recently, molecular biology and statistics.

The first objectives in sunflower breeding programs place emphasis on high seed yield and high oil content. The selection must be targeted on genotypes with high oil content in kernels – to obtain a high oil yield per unit. For a successful realization of high yields it is necessary to improve a range of properties and characteristics, such harvest index, resistance to biotic and abiotic stresses, earliness, adaptability.

For special markets, breeding may be slightly different. For confectionery sunflower hybrids is needed large achene and kernel size, special shape and color.

In recent period, introgression of genes for herbicides resistance (imidazolinone and sulfonylurea) from wild *Helianthus* species has become a topical breeding objective for both oil and confectionery sunflower.

Detection of genes for modified oil quality of sunflower and their introduction into productive genotypes are important objectives. The new mutant have a high oleic acid content or palmitic or stearic acid. Developing hybrids with modified tocopherols (alpha, beta, gamma and delta) is an important breeding objective because this feature may increase storage life of oil.

Defining an ideal plant type, for specific agroecological conditions is also important in sunflower breeding.

On the whole, the major sunflower breeding objectives, for all sunflower types, should be high yields and quality of oil, proteins and other products for non-food industries. The target is to produce maximum yield, taking into account the economy of production inputs and to minimize the negative environmental effects.

THE BEHAVIOR OF A SUNFLOWER HYBRIDS SET IN DIFFERENT SOIL AND CLIMATIC CONDITIONS, IN ROMANIA

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Sunflower crop is resistant to dry conditions, but, in case of rainfall missing or high air temperature, specially in fill up seed time, the seed yield could be affected.

The oil content in sunflower seed can be influenced by climatic conditions, registering different values in different years.

Our study with 12 sunflower hybrids in 3 locations and two years has demonstrated the influence of soil and climatic conditions on seed yield and oil content.

The rainfall and air temperature have been different in these two years and three locations, so, the seed yield for the twelve hybrids was higher in 2013 year, for all locations and in one location comparing with other two, in 2012 year. The oil content for these 12 hybrids was higher in the same location comparing with other two, in both years, this being influenced by soil type, too.

The seed yield was much influenced by plant height, head diameter and one thousand seeds weight, in all locations.

The statistical analyze has showed that the interaction “year \times location” it was very significant in case of the seed yield as well significant in case of oil content.

RESEARCH ON TETRAPLOID MAIZE WITH *OPAQUE-2* ENDOSPERM

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Maize (*Zea mays* L.) is considered a diploid species ($2x=20$). In the experimental polyploid series of maize, the only forms of practical importance are tetraploids ($4x=40$). The first tetraploid forms of maize were obtained by L. F. Randolph in 1932 by temperature shock. These maize cytotypes have valuable morphological, physiological and biochemical traits but show reduced fertility, slower growth, longer growing season, low productivity, which make them uncompetitive with diploids. However, scientists use them in different research programs regarding genetic variability, inbreeding and heterosis, genomic imprinting, gene expression, dosage effects etc. Also, some researchers from Russia succeeded in obtaining tetraploid maize populations on a broad genetic base, including introgression of teosinte, that have high productivity, protein content and are drought resistant.

At the State Agrarian University of Moldova are carried out experiments with the aim of using polyploidy and kernel mutant genes in improving the quality of grain proteins. In this paper we present the results on induction and study of tetraploid forms of maize which contain the *opaque-2* gene (*o2*) that determines a high content of lysine in the protein of the kernel.

The research was carried out during 2010-2014. The genetic stocks used in the experiments consisted of a single-cross maize hybrid with increased protein and lysine content Chişiniovşchi 307PL with *o2* gene in the endosperm, single-cross hybrid Porumbeni 331Mrf, and the tetraploid synthetic B with normal endosperm. Tetraploidy was induced by colchicine treatment of 0,15% concentration. Newly created tetraploid forms of maize were studied on the morphological, cytological and biochemical levels. Biochemical analysis was performed by infrared spectroscopy. Protein amino acid content was determined by ion exchange chromatography on an automatic aminoacid analyser T339M.

Application of the colchicine allowed obtaining chimeric plants characterized by different degrees of morphosis. Heterogeneous pollen size served as an important criterion in selecting valuable plants. Self-pollination of these plants generated chimera ears with various classes of kernels, distinguished by size and extent of development of the endosperm. Using these criteria we selected tetraploid grains that differed by size, color and weight, which was 30% higher than diploid grains. After selecting tetraploid grains, next step was the verification of chromosomes number to avoid the cases with diploid or aneuploidy grains. Tetraploid plants obtained from selected grains were shorter than the original diploid forms, had a thicker stem, shorter internodes, fewer panicle branches with central branch thicker and longer. Stomata sizes of tetraploid forms were on average 20-25% higher than in the diploid but, less per unit of surface. Pollen from tetraploid forms was larger, but with a low degree of fertility. Tetraploid grain mass is greater than the diploid, but greatly varies.

In order to determine gene heredity in tetraploid forms we applied the hybridological method by crossing the *o2* tetraploids with the synthetic population B, which has vitreous endosperm. As a result of the analysed tetraploid cobs obtained in the second generation (F_2) by self-pollination of a duplex *O2O2o2o2* we obtained an empirical 34,33:1 segregation, which is close to the theoretical 35:1. Application of the χ^2 -test generated a value $\chi^2 = 0.055$, which showed that the *o2* gene inherits at random chromosomal segregation model.

Biochemical analyzes performed on the material under study revealed some essential differences between diploid and tetraploid forms. Protein level in tetraploid *o2* grain was on average 17% higher than in the diploid mutant form Chişiniovşchi 307 PL and 30% higher than in the diploid hybrid Porumbeni 331 MRf. No differences were found between *o2* tetraploids and the tetraploid synthetic B. The content of lipids in tetraploid mutant grains was lower than that of diploid mutant form, practically at the level of normal diploid grains, but no essential differences were noticed as regard to cellulose content. The analysis of amino acids content in the protein of diploid and tetraploid grains containing *o2* gene, revealed that with increasing ploidy level there was a tendency to increase the content of aspartic acid, threonine, serine, glutamic acid, tyrosine, alanine, reducing content of proline, glycine, valine, cysteine, leucine, isoleucine, histidine, arginine. Experiments with different doses of *o2* gene in grain endosperm showed that in the case of six *o2* recessive alleles, the lysine content was higher than all other doses of diploid and tetraploid levels.

PRECOCITATE DE RODIRE A HIBRIZILOR DE PĂR

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Pear has a long juvenile period. In order to accelerate the process of selection is necessary in the process of hybridization include varieties with early fruiting. Cultivars Sokrovishche and Noiabriskaya with an earlier beginning of fruiting showed an earlier entry into fruition theia hybrids. However, majority of own-rooted hybrids begin to bear fruit from 11 year.

Cultura părului se deosebește prin perioada juvenilă lungă. Cu scopul accelerării procesului de ameliorare este necesar a include în procesul de hibridare soiurile cu intrare pe rod timpurie. Soiurile Socrovișce și Noiabriscaia intră pe rod mai devreme decât celelalte soiuri în studiu, la fel și hibrizii lor intre pe rod mai devreme. Totuși, majoritatea hibrizilor studiați intre pe rod începînd cu al 11-lea an.

Unul din indicii de bază a culturii părului este precocitate de fructificare, deci pomii părului se deosebesc prin perioada juvenilă lungă, îndeosebi, pomii pe rădăcini proprii, oscilînd între 7 și 30 ani (1). Altoirea reduce perioada juvenilă pînă la 5-7 ani pe portaltoi franc și 3-5 ani pe portaloii gutui, dar ambii portaltoiuri au neajunsuri. Tendința de intensificare a culturii părului impune amelioratorilor crearea soiurilor noi cu intrarea timpurie pe rod genetic determinată. Intrarea rapidă a pomilor pe rod reduce procesul de ameliorare și răspîndire a soiurilor noi create în producție.

În calitate de obiecte de cercetare au servit 330 hibrizi, creați la ISPHTA, din 6 combinații de hibridare a soiurilor Noiabriscaia, Ciudo, Socrovișce și elitei 8-18-100, obținuți în a. 1994. Hibrizii sunt amplasați în livada de selecție pe rădăcini proprii la Stațiunea Experimentală "Codrul".

Soiurile utilizate în procesul de hibridare au fost selectate după un set de caractere valoroase, în primul rînd calitatea fructelor, productivitatea, rezistența la boli și dăunători. Deși intrarea pomilor pe rod depinde și de condițiile pedoclimatice și de nivelul de tehnologie, totuși este genetic determinată. Studiul particularităților biologice ale soiurilor și elitelor de păr au arătat diferența semnificativă după acest caracter. La fel și soiurile incluse în hibridarea diferă semnificativ. Soiul Socrovișce se deosebește prin cea mai timpurie intrare pe rod a pomilor pe portaltoi păr franc- al 4-lea, apoi soiul Noiabriscaia la al 5-lea an după plantare, iar soiul Ciudo și elita 8-18-100 la al 7-lea an după plantare. Pomii hibrizilor pe rădăcini proprii intră pe rod și mai tîrziu. Începutul fructificării hibrizilor în studiu a oscilat între anii 7 și 20. În combinații de încrucișare a soiurilor Socrovișce și Noiabriscaia au fost obținuți niște hibrizi cu intrare pe rod la al 7-lea an după plantare. În combinație de încrucișare soiului Ciudo intrarea pe rod a pomilor începe cu al 10-lea an după plantare, iar elitei 8-18-100 la al 12-lea an după plantare.

Intrarea pe rod la partea majoră a pomilor hibrizilor în studiu (87%) a oscilat între 7 și 20 de ani. Ponderea hibrizilor cu intrarea pe rod pînă la 11 ani consituie 8,4%. Partea majoră a hibrizilor intră pe rod începînd din anul al 11-lea, iar 8% din hibrizi în studiu intră pe rod foarte tîrziu - după 20 de ani. Pomii hibrizilor cu intrarea timpurie pe rod pe rădăcini proprii va fi și mai precoce și pe portaltoi.

Cu scopul accelerării procesului de ameliorare este necesar a include în procesul hbridării soiuri cu intrare timpurie pe rod, care corelează cu intrarea timpurie pe rod și hibrizilor lor. Din cercetările efectuate cele mai bune rezultate au fost obținute în combinații de hibridare a soiurilor Socrovișce și Noiabriscaia.

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**TRENDS AND RESULTS OF RESEARCHES ON WINTER WHEAT BREEDING IN THE
CONDITIONS OF BALTI STEPPE OF REPUBLIC OF MOLDOVA**

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The winter wheat breeding orientation in the Republic of Moldova is determined by several factors:

- strongly pronounced soil and climatic zonality;
- unfavorable structure of predecessors for this crop;
- obvious tendency to climate continentality, especially of hydrothermal regime during the last decade.

Such situation makes it necessary to create new varieties of winter wheat with a wide range of agro-biological characteristics.

Thus in the RIFC "Selectia" was founded and developed a model of varieties of two ecological types:

- varieties of intensive ecotypes, with a straw length of 75-90 cm, highly resistant to lodging, destined for cultivation after early harvested predecessors, high level of soil fertility, including irrigation conditions;
- varieties of semi-intensive ecotypes, with a straw length of 95-110 cm, resistant to lodging, recommended for less fertile soils, late predecessors and eroded soils on slopes.

Varieties of both ecotypes have their own specific characteristics and properties, but they have the following in common:

- high level of productivity in combination with high adaptive capacity in contrasting conditions of their cultivation;
- good indicators of grain quality;
- sufficient level of frost-winter hardiness and drought-heat resistance.

The drought-heat resistance has acquired a special priority during the last period of time because of high air temperatures in the spring and summer - the phase of spike formation and grain filling.

Taking into account this concept, new varieties have been selected at the RIFC "Selectia" and have been included in the State Register of Plant Varieties of the Republic of Moldova with high productivity, adaptivity and competitiveness, such as: Dumbrăvița, Căpriana, Baștina, Lăutar, Talisman, Meleag, Vestitor, Vatra and others.

A number of new varieties are at the moment in the state trials, such as: Creator, Rod, Fenix, Acord, Numitor and Căpriana Plus.

For the near future in breeding work is planned to enhance drought resistance and heat resistance of winter wheat new varieties.

**PLANT – GREENBUG INTERACTION: SELECTION IN PHYTOPHAGE
POPULATIONS AND RESISTANCE EXPRESSION**

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The study is devoted to investigating regularities of selecting phytophages by aggressivity and virulence under influence of plant genotypes. The possibility of the greenbug (*Schizaphis graminum* Rond.) population genetic structure radical changes during a host vegetation period was revealed. Seasonal variation of greenbug collected on sorghum was revealed for the virulence to barley samples i.e. crop not being the phytophage host at the monitoring period. During reproduction of the insect on barley plants the individuals which did not possess “redundant” genes for virulence to sorghum had the advantages in competition. Change of the host led to rapid accumulation of the clones virulent to sorghum genes for resistance. With the use of molecular markers a high seasonal polymorphism in *S. graminum* genotype composition was found in the clones with dominated phenotypes for virulence. Under alteration of the host a significant shift of genotype groups, similar by RAPD profiles, was observed. Distribution of RAPD markers was independent of alleles for virulence of the aphid. Difference of greenbug populations from Krasnodar and Dagestan by the frequencies of virulence to major and weakly expressing plant genes has been demonstrated. For the first time the heterogeneity and difference of the two phytophage populations by the nucleotide sequences of mitochondrial genome fragments was revealed using pyrosequencing technique. A significant influence of weakly expressed resistance of host plant on *S. graminum* fecundity was found. Barley weakly expressing genes of resistance to *S. graminum* were shown to differentially interact with phytophage genotypes and can not be a basis of durable (horizontal) resistance. A high frequency of resistant to *S. graminum* forms among local barley and oat accessions from Asian countries was revealed. This is probably due to the long history of interactions between insect and plant hosts. Forty nine barley accessions heterogeneous by the resistance to *S. graminum* were identified the resistant components of those possessed resistance genes alleles which are different from alleles of the earlier identified *Rsg1* gene. It is assumed that the oat accessions resistant to Krasnodar population of *S. graminum* are protected by the genes differing from the known *Grb1* – *Grb3* genes. In the natural populations of greenbug the selection of insect virulence phenotypes was observed which was conditioned by weakly expressed resistance of plant host. The phytophage clones with a more broad virulence spectrum are more competitive at the reproduction on susceptible genotypes. Reproduction on the relatively resistant forms can result in decreasing frequency of genes for virulence to major genes for host resistance in the insect population. During reproduction on the plant genotypes with different resistance level the alteration of nonspecific aggressiveness of greenbug is observed. In compatible combinations of the phytophage – plant host interaction the increase of nonspecific aggressiveness of *S. graminum* is usually detected. The conditions of PCR and 9 primers to six genes involving in sorghum immune response at greenbug colonization of plants have been elaborated. It was shown on the example of interaction between variety Sorgogradskoe protected with the resistance gene *Sgr5* and virulent and avirulent greenbug clones that the amount of mRNA for key defensive proteins depends on virulence and duration of the pytophage feeding.

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INFORMATION STANDARDS FOR PLANT GENE BANK

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Developing standards for documentation and for exchanging information is essential for ensuring that the vast amount of data on crop species and varieties is available to countries to improve their capacity to store, manage and share information about the plant genetic resources. In order to exchange data, it is necessary to have compatible documentation systems. This can only be achieved through common standards for information exchange. In this context, the lists of standardized descriptors have been developed by Bioversity International (formerly the International Plant Genetic Resources Institute, IPGRI) since 1976.

Descriptor lists include key attributes, characteristics or traits of a crop, and set out the method used to measure and document them, along with the relevant registration data. Descriptor lists therefore aim to include information and data that are relevant for different types of genebank operations for a specific crop or gene pool, from initial registration, through characterization, evaluation and management, to their eventual use. Included in a descriptors list are passport data, characterization and evaluation.

Generally, three types of standard were adopted: crop descriptors, Multi-crop passport descriptors (MCPD) (FAO/IPGRI), and Descriptors for genetic marker technologies.

Each crop descriptor list represents an important tool for data exchange by providing an internationally agreed format and universally understood language for PGR information, particularly with regards to characterization and evaluation data. Descriptors have been developed by Bioversity International in collaboration with scientists and international research organizations for almost 100 crops.

The MCPD standard represents a minimum set of passport descriptors which are contained in each crop-specific descriptor list. These descriptors aim to be compatible with IPGRI crop descriptor lists and with the descriptors used for the FAO World Information and Early Warning System (WIEWS) on plant genetic resources (PGR).

This list of descriptors for genetic marker technologies defines a minimum set of data needed to describe accessions using biochemical and molecular markers, and defines community standards for documenting information about genetic markers.

The system for the documentation of plant genetic resources in Republic of Moldova named *ReGen* represents unified information system that includes three basic functional blocks: ex situ, in situ/on farm conservation. *ReGen* contain the passport data, evaluation and characterization data. For the standardization of data are used common international descriptors, developed by the Bioversity International with the participation of FAO. In case of passport data are used the List of Multi-Crop Passport Descriptors (MCPD), and for evaluation and characterization – IPGRI Crop descriptors.

For elaboration of *ReGen* system was used programming language Visual Fox Pro 9.0. The system was set up for operation system Microsoft Windows 2000 and XP. *ReGen* is a relational information system. Databases consist of many tables connected by special fields. Between tables through indexation may establish certain relationships that allow fast search of necessary information.

Passport data includes twenty-eight fields such as: institute code, accession number, collecting number, genus, species, accession name, acquisition date, country of origin, geographical description, biological status, type of germplasm storage, collecting source, ancestral data, donor institution etc (Alercia, A., 2012). At present about 2 000 plant genetic resources are described in passport database. Characterization and evaluation of PGR is made using IPGRI (Bioiversity International) crop descriptors. A lot of information is documented manually and only a small part of these data has been computerized.

Standardization of information is necessary that allow an easy exchange of information between different genebank at the European level. In this context, EURISCO - European Internet Search Catalogue was created. A set of data (about 2000 accessions) on plant genetic resources from Moldova has been loaded to EURISCO web catalogue (<http://eurisco.ecpgr.org>)

The comprehensive and standardized description of a crop provides for better compatibility between documentation systems and facilitates information exchange thus encouraging and supporting collaboration among scientists working in various countries.

STUDIUL LEGĂTURILOR FUNCȚIONALE ÎNTRE INDICELE DE PLUTIRE A BOABELOR CU PARAMETRII BIOCHIMICI AI CALITĂȚII ȘI VIABILITĂȚII SEMINȚELOR DE PORUMB

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Apariția necesității de reorientare la cerințele Europene față de calitatea materialului semincier, care se crează și se reproduce în Republica Moldova a determinat oportunitatea de identificare a indicilor noi de calitate a boabelor de porumb. În special, analiza detaliată a posibilităților metodice ale colegilor români, care se ocupă cu studierea porumbului alimentar, arată utilizarea efectivă a parametrului "indicele de plutire a boabelor" (IPB). De aceea, prezintă interes problema de cercetare a posibilităților folosirii a acestui indice a calității boabelor în procesul ameliorării porumbului în Republica Moldova și pe tot spațiul post-sovietic.

În calitate de material pentru cercetările efectuate au fost folosite 70 linii de porumb (indurat și dentat), și 70 combinații hibride (inclusiv hibrizi de tip indurat). Cercetările au fost efectuate în decurs de două sezoane de vegetație (2013 și 2014). Capacitățile de germinare și vigoarea germinării s-a determinat după GOST R 5255-2005; procentul de umiditate - după GOST 1396.4-80 cu utilizarea metodelor gravimetrice; conținutul proteinei crude - GOST 13496.4-92 (metoda Kjeldahl); uleiul după metoda Rucovski (GOST 13496.15-97); amidonul după metoda Ewers; carotinoizii (β -carotina) – după Ermacov. Totalitatea acestor metode a creat baza de calculare a ecuațiilor de regresie a analizatorului cu unde infraroșii (modelul 4250, SUA), care lucrează în regiunea apropiată a spectrului - după standardele: P50817-95 și P50852-95 ce a permis să diagnosticăm accelerat calitatea boabelor la 33 hibrizi de porumb de tipul indurat. Pentru studierea "indicei de plutire" în laboratorul de biochimie și fiziologie a Institutului de Fitotehnie "Porumbeni" a fost aprobat standardul român STAS 5447, n.4.7 cu câteva modificări specifice, bazat pe diferența de densitate care există între stratul cornos și cel făinos al endospermului. Rezultatele obținute au fost prelucrate după analiza corelativă cu ajutorul programelor Microsoft Word, Microsoft Office, Excell 2007, Point Adobe Photoshop 7,0 prin sistemul de operare Windows XP.

Rezultatele obținute au dat posibilitate să stabilim că indicele de plutire a boabelor de porumb se caracterizează cu valoarea medie de corelație cu indicii de productivitate a acestei culturi: după masa 1000 de boabe și după recolta boabelor. Sunt delimitate intervalele de variație a indicelui de plutire, permițând alocarea cu precizie a liniilor de porumb indurat și dentat în corespundere cu două intervale a indicilor de plutire: de la 3 până la 65% (indurat) și de la 65 până la 100% (dentat). La hibrizii de porumb indurat este relevantă corelația negativă esențială cu conținutul de β -carotină în boabele genotipurilor studiate. În procesul de manipulare experimentale privind determinarea indicelui de plutire este demonstrată și eficiența folosirii parametrului "numărul boabelor cu calitate de plutire" - (N_1): ieșirea boabelor la suprafața soluției de NaNO_3 , cu densitatea 1,25 g/cm³, - pentru genotipul corespunzător studiat. Anume grație de acest parametru sunt stabilite relațiile corelative de semnificație medie cu indicii fiziologici (capacitățile de germinare și de vigoare germinării) la porumb dentat.

Pe baza analizei rezultatelor obținute autorii consideră rezonabil să recomande parametrul "indicele de plutire a boabelor" (IPB):

a) ca marker selectiv și pentru liniile de porumb indurat (IPB < 65%), și pentru liniile de porumb dentat (IPB > 65%);

b) ca un marker selectiv a mostrelor de porumb cu conținutul înalt de carotină pentru diagnosticarea rapidă a materialului inițial pentru procesul de ameliorare la calitate a formelor indurate de porumb alimentar.

De asemenea se recomandă folosirea indicelui N_1 ("numărul boabelor cu calitate de plutire" - în soluție NaNO_3) pentru studii fiziologice mai aprofundate la prima etapă de germinare a porumbului dentat cu scopul de a identifica posibilitățile diagnosticării rapide a capacităților de germinare și de vigoare a germinării după parametrul N_1 .

AMELIORAREA GRĂULUI DURUM DE TOAMNĂ ÎN REPUBLICA MOLDOVA

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Grăul durum de toamnă, întrunind în mod reușit caractere și însușiri valoroase ale grăului comun de toamnă și a grăului durum de primăvară, s-a impus ca o cultură importantă în industria mondială a gramineelor, evidențiindu-se prin însușiri de calitate deosebite. Având boabe cu o sticlozitate înaltă și conținut sporit de proteină și gluten, este de neînlocuit în industria de paste făinoase.

În natură grăul durum este ca cultură de primăvară sau umblatoare. În anii cu ierni blânde formele umblatoare dădeau roade bogate. De aceea grăul durum de toamnă demult a atras atenția amelioratorilor deoarece productivitatea lor era cu mult mai mare decât a celor de primăvară. În legătură cu aceasta înaintea amelioratorilor s-a pus problema de a crea o cultură nouă – grău durum de toamnă. Ameliorarea grăului durum de toamnă în republica noastră a fost începută în anii 1960 de către savanții Buicli, Covarschii, Sulima și Gheorghiev. În rezultatul hibridărilor interspecifice dintre grăul comun de toamnă și grăul durum de primăvară au obținut forme prețioase după particularitățile biologice și indicii agronomici. A doua etapă a creării acestei culturi a fost îmbunătățirea ei prin hibridările intraspecifice.

În Moldova această cultură este cultivată pe suprafețe mici, deoarece persistă pericolul de îngheț. Rezistența la iernat este unul din criteriile de bază în ameliorarea grăului durum de toamnă, deoarece în Moldova temperatura minimală la adâncimea așezării nodului de înfrățire scade până la -15(-20°C). De aceea în unii ani se urmărește peirea plantelor acestei culturi, ceea ce duce la un grad înalt de rărire a semănăturilor. Prin urmare principala problemă a amelioratorilor acestei culturi este crearea formelor și soiurilor noi de grău durum de toamnă cu o rezistență înaltă la iernare. Primele soiuri de grău durum de toamnă, create în republica noastră Meleanopus 276, Hordeiforme 16 și al. după rezistența la iernare erau la nivelul soiurilor Miciurina și Novomicurina (grău durum de toamnă). Soiurile Chișinevscaia ranea, Chișinevscaia nepolegemaia și al. [Buiucli 1969,1972] după rezistența la iernare se aflau la nivelul soiurilor Odesskaia iubileinaia și Rubej (grău durum de toamnă). Ele se caracterizau prin spic mare, cu număr mare de boabe, conținut înalt de proteină (16-17%) și gluten (28-32%). Cu părere de rău, aceste soiuri n-au fost omologate și nu s-au răspândit, de oarece înfrățirea plantelor a fost mică și talia plantelor înaltă. În anii cu o cantitate mare de precipitații aceste soiuri poligneau și complica strânsul recoltei, ceea ce duce la micșorarea productivității și înrăutățirii calității boabelor. De aceea, începând cu anii 1980 lucrul de ameliorare s-a dus în direcția micșorării înălțimei plantelor, măririi rezistenței plantelor la iernare și cădere, sporirea înfrățirii plantelor.

În ultimii ani la Institutul de Genetică, Fiziologie și Protecție a Plantelor al AȘM în rezultatul efectuării unui număr mare de hibridări interspecifice și intraspecifice au fost create și omologate trei soiuri de grău durum cu talie scurtă (Auriu 273, Hordeiforme 335 și Hordeiforme 333) cu un potențial de productivitate de 6 t la hectar, cu rezistență înaltă la factorii abiotici și biotici ai mediului ambiant și cu un conținut înalt de proteină (15-18%) și glutenă (26-32%), care actualmente pot fi sămănite în condițiile climaterice ale republicii noastre. Soiurile și formele create de noi prin încrucișările interspecifice și intraspecifice dintre cele mai bune forme de grău durum de toamnă și cei mai buni genitori de grău comun, deschid perspective noi pentru viitorul acestei culturi în țara noastră. S-ar părea că odată cu crearea soiurilor înalt productive, care sunt adaptate la condițiile de mediu pot rezolva pe deplin problema producerii boabelor de grău durum în țara noastră. Însă, cu părere de rău ca și cu 20 de ani în urmă această cultură nu poate fi cultivată pe suprafețe mari.

Principalele pricini a nerezolvării acestei probleme după părerea noastră sunt următoarele: - lipsa cerinței grăului durum de toamnă pe piață, în legătură cu faptul, că întreprinderile de fabricare a crupelor și a productiei de macaroane lucrează în baza boabelor de grău comun; - lipsa întreprinderilor de prelucrare a boabelor de grău durum și a liniilor de producție a spaghetelor, la care ca materie primă va servi numai grăul durum. Cu toate acestea amelioratorii continuă lucrul de cercetare în direcția îmbunătățirii acestei culturi în Republica Moldova.

THE MODIFICATION OF SINGLE CROSS HYBRIDS OF EARLY MAIZE

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In the report are presented the results of modification of single cross hybrids of early maize for export in northern areas by substitution of poorest parent and use of crosses between sister/ related inbred lines as seed parent. In the investigations were included 8 inbred lines, 16 related crosses A x A1 as seed/ female parents from Reid Iodent germplasm group, 8 inbred lines from BSSS-B37 and Euroflint alternative heterotic groups as pollen/ male parents. This biological material were evaluated in 2010 – 2014 years for such agronomical important traits: seed cold tolerance, seedling vigor, flowering, silking and physiological maturity, resistance to *Ustilago maydis* and *Sorosporium reilianum*, grain yield and moisture, general (GCA) and specific (SCA) combining ability. The female parents additionally were appreciated for 1000 kernel weight (TKW) and the sizing of the seed into different fractions based on the width and length. The studies of seed germination under suboptimal temperatures in field plots indicate that 10 inbred lines possessed middle tolerance – under 70% of germination. In the group of low tolerance genotypes (less than 60%) were included 3 inbred lines and 3 inbreds showed more than 70% of seed germination classified as high tolerance. The highest percentage of emerged seeds (83,2%) were observed at flint inbred line MKP 20. The dent inbred lines from Reid Iodent and BSSS-B37 germplasm groups achieved the physiological maturity, marked by appearance of the black lower on the base of kernels, for 105,5 days from emergence with a variation from 102,5 to 108,8 days. For flint genotypes were characteristic early flowering, silking and maturity – 2 days early than dent inbreds. In 5 years period the average production of grain was 3,91 t/ha for Reid Iodent inbred lines, 3,63 t/ha for BSSS-B37 and 2,88 t/ha for flint inbreds with Euroflint germplasm. These results indicated that Reid Iodent germplasm were better adapted to the natural conditions of Moldova and is more preferable as seed parent. Related crosses formed in average 5,68 t/ha grain with the fluctuation of grain heterosis from 20,5% until 89,6%. The evaluation of 16 inbred lines and 16 related (sister) crosses for general and specific combining ability permitted to select inbreds MKP 61, MKP 63, MKP 64, MKP 70, MKP 71, MKP 711 and related crosses MKP 61 x MKP62, MKP 63 x MKP 62 with high values of CGA effects. In preliminary testing trials were evaluated 60 single cross and 112 modified single cross hybrids from maturity group FAO 210 – 300. Advanced level of evaluation testing reached 21 hybrids (28.7%) from heterotic pattern Reid Iodent x BSSS-B37 and 23 hybrids (21.9%) realized in heterotic pattern Reid Iodent x Euroflint. In generally hybrids with male parent from Euroflint germplasm group were more sensitive under drought and longer growing season of Moldova but in cold and shorter season areas of Belarus performed better than dent genotypes for grain and silage. The testing program in one location of Moldova and two locations in Belarus of modified single crosses A1 x B, A x B1, A1 x B1 from heterotic pattern Reid Iodent x BSSS-B37 resulted in identification of 7 hybrids with significant performance for grain production and lower moisture, including Bemo 235 registered in Belarus and Porumbeni 310 in Moldova. In heterotic pattern Reid Iodent x Euroflint were selected only one competitive single cross hybrid that performed well in all ecological locations. From related crosses were selected 3 female parents with 25.8 – 56.9% of heterosis for grain yield and uniformity of plant and ear traits in modified hybrids (A x A1) x B. The modified combinations in generally were closely to single crosses for grain yield and acceptable uniformity. Related crosses that serve as female parent in a production field contribute to the stable yield, larger quantities of seeds, better TKW with separation of kernel fractions with greatest interest and increase the profitability of hybrids multiplication. Analysis of correlations among grain yield of related crosses per se and hybrid combinations show a low negative relationships ($r=-0.110$). A middle significant, positive correlation ($r=0.654$) exists between moisture of related crosses per se and moisture of hybrids. Experimental dates of this study suggest that for substitution of poor parent may be useful inbred lines with higher general and specific combining ability, better grain yield per se. For related crosses as seed parents important are combining ability, index of heterosis no more than 60% and uniformity of plant traits in hybrids. The results of researches are new modified single crosses from heterotic pattern Reid Iodent x Euroflint registered in Russia (Rosmold 202MRF) and Belarus (Bemo 203).

APPLICATION OF METHODS OF POLLEN ANALYSIS FOR THE EVALUATION OF RESISTANCE OF TOMATO REGENERANTS

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It is well known that high temperatures and lack of moisture significantly reduce growth and productivity of most cultivars including tomato. In connection with this the selection for resistance to these factors is an actual breeding and genetic problem. The successful realization of this problem suggests the use of genotypes adapted to the stressful abiotic environmental factors in the process of selection. Thus, studies dealing with the developing of methods of adaptive selection become most important, being based on the latest achievements of genetics and biotechnology. These methods allow in a short time to improve the efficiency of traditional breeding approaches on the base of creation of new genotypes - donors of resistance to stress factors. In order to identify new sources of adaptability to abiotic environmental factors the collection of tomato regenerants was tested for the thermostability and resistance to lack of moisture of male gametophyte. As a result, it was found that most genotypes in field conditions formed pollen with a high viability (64,5 ... 76,0%), that exceeded the value of control in 1,3 ... 1,4 times. In the experiment, combined action of temperature and low moisture reduced pollen viability and pollen tube length in 1,7 ... 7,7 times depending on the genotype. The processing of the obtained results by the method of multivariate dispersion analysis revealed that the main contribution to the identified variability of viability and resistance of male gametophyte includes the temperature, the impact of the genotype is less significant. At the same time the main role in variability of the spectrum of length and stability of pollen tubes belongs to lack of moisture. The obtained results showed that the most genotypes are characterized with the high level of gametophyte resistance (65,3 ... 81,1%), that exceeds the control value in 1,5 ... 1,8 times. Among the studied genotypes, the most high level of thermostability was demonstrated by regenerants 52/1 and 69 (81,9% and 77,6%). At the same time from the total number of studied genotypes only one demonstrated the level of resistance of gametophyte to lack of moisture more than 30%. As a result of studies carried out there were identified three genotypes: 52/1, 69, and 50, which combined resistance to both: temperature and lack of moisture. Processing of the obtained results by the method of correlative analysis allows to reveal the significant positive correlation between pollen viability and resistance in conditions of high temperature and low moisture ($r=0,78^{**}$), as well as between the length of pollen tubes and their resistance to stress conditions ($r=0,84^{***}$), that may indicate the effectiveness of selection for these characteristics.

Thus, as a result of studies some genotypes were revealed that demonstrated high values of the functional parameters of male gametophyte and combined resistances to high temperatures and lack of moisture. These genotypes may be valuable original material for selection. Summarizing the data of three years of researches and based on the study of collection of tomato regenerants there were identified 12 new sources of resistance to the temperature factor, as well as 7 regenerants resistant to lack of moisture. Values of resistance of these genotypes differ in ranges 79,8 ... 59,7%, exceeding the control value in 1,5 ... 1,8 times. Also, 2 genotypes were identified which combined pollen thermostability (77,9 and 82,2%) and high productivity of 48,9 and 60,2 t/ha. Thus, this evaluation allows to characterize the reaction of gametophyte regenerants to the action of stressful abiotic factors, to make their differentiation and to identify resistant genotypes which can be recommended for cultivation in conditions of high temperature and drought, as well as for selection of parental pairs for hybridization. The combination of methods of embryoculture and gametophyte selection can be an effective approach to create and detect at early stages new high resistant and productive genotypes.

IMPORTANT QUANTITATIVE CHARACTERS OF MATERNAL HAPLOID INDUCER LINES IN MAIZE (*ZEA MAYS* L.)

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One of the ways to accelerate the production of improved maize genotypes is to reduce the time of homozygous lines creation. This objective has become achievable due to the application of doubled haploid technology (DH) [Röber et al., 2005; Geiger, Gordillo, 2009; Prigge, 2012]. The process of DH lines production from heterozygous germplasm includes two generations. In first generation the haploidy is induced from diploid plants, their chromosomes number is reduced to half. In second generation, haploid set of chromosomes are doubled, each chromosome get an identical copy and diploid number is restored. The widely production of haploids is possible due to the creation of special lines with genetic capacity to induce the haploidy. For identifying the haploids, in inducers were integrated certain marker genes that determining anthocyanin pigmentation in specific organs at different stages of development. In the process of haploid-inducing lines creation, is also important to improve quantitative traits of agronomic interest.

In our research, were crossed Stock 6, ZMS and MHI inducers which differ considerably according by plant height and tasseling length. Even the rate of induction and expression of marker genes were the main criteria of selection, it is important that some families F_6 were morphological more advantageous than the best of genitors – MHI. Through correlational analysis (r) was found that dependence between plant height and length of tasseling of F_3 inducers was $0.77 * (p \leq 0.05)$, while of F_6 inducers – $0.72 * (p \leq 0.05)$. This demonstrates that plants with higher height have a longer tasseling, the regression equation of dependence is: $y = (-8,6528 + 0,1949) \times x$ and $y = (3,5508 + 0,1182) \times x$, respectively, for F_3 and F_6 inducers. Based on the equation were y - tasseling length, and x - the plant height can be calculated accurately tasseling length which is a significant ameliorative character, because it is probable that such tassels produce a greater amount of pollen. Be mentioned that inducers that have deviated from the general trend - the theoretical curve and confidence interval reveals that although in general there is a positive correlation between *plant height* and *panicle length* among tested genotypes are inducers that have relatively small height, but tasseling length - great. This refers to the inducers $F_6 26$ and $F_6 28$.

For haploid-inducing lines, from quantitative characters is important the pollen quantity and the duration of flowering tasseling. Thus in F_6 progeny, inducers plants daily produced 0,188 to 0,318 g of pollen with flowering duration of 3-5 days. Compared with the most performant original lines - MHI, all improved inducers families produced more pollen. The most improved family – $F_6 27$; $F_6 28$; $F_6 32$; $F_6 29$; $F_6 33$ exceeded the level of the MHI (1,142 g), the average of analyzed character ranging in limits from 0,256 to 0,318 g. The dendrogram of distribution F_6 inducers, developed on the base of *plant height*, *tasseling length*, *amount of pollen* and *duration of flowering tasseling* characters, showed that the highest similarity with the ZMS and Stock 6 original lines manifested: $F_6 26$, $F_6 30$, $F_6 32$, $F_6 27$, $F_6 28$, $F_6 29$, $F_6 33$ and with MHI: $F_6 36$, $F_6 37$, $F_6 63$, $F_6 38$, $F_6 39$ which showed high values of the parameters included in the study. Thus, the average values of these families varied within: 165,5 to 176,5 cm; 21,3 to 24,5 cm; 0,188 to 0,234 g; 4-5 days, respectively for plant height, length of tasseling, amount of pollen, duration of tasseling flowering.

Through the analysis of clusters based on *k-means* method was found that the highest capacity of differentiation of clusters showed *plant height*, followed by *tasseling length* and *duration of flowering*. The amount of pollen did not present a relevant index for differentiation of clusters the values were similar in all genotypes studied. Along with mentioned characteristics, the created inducers showed resistance to the fall and diseases.

IDENTIFICAREA POTENȚIALULUI AMELIORATIV AL RESURSELOR GENETICE ALE VÎTEI DE VIE

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Rezolvarea cardinală a problemei privind protecția vîtei de vie contra factorilor abiotici și biotici stresanți poate fi realizată prin crearea soiurilor noi, a căror rezistență este asigurată de constituția genetică a plantei. Lucrările de ameliorare genetică a vîtei de vie în Republica Moldova și practica explorării generațiilor de hibrizi interspecifici F_1 a permis stabilirea caracterului corelării calității strugurilor și a rezistenței la iernare și patogeni. Valorile coeficienților de corelație liniară între calitatea boabelor și rezistența la iernare, între rezistența la iernare și rezistența la mană pentru populațiile de descendenți sunt mici și ne semnificative ($r = -0,08 \pm 0,19$), ceea ce determină că descendenții au moștenit independent aceste însușiri. În cadrul materialului biologic însușirile de calitate înaltă a boabelor, rezistența la mană și la ger sunt întâlnite în diferite combinații și sunt o confirmare practică a ipotezei posibilității combinării libere a acestor însușiri. Aceasta denotă lipsa linkajului între genele sau complexul de gene ce determină caracterele studiate sau, chiar dacă genele sunt prezente în același cromozom, amplasarea lor este departe una de alta, astfel că linkajul nu se manifestă. Prin urmare, bariere genetice în transmiterea prin ereditate la descendenții hibrizi a calității și rezistenței nu sunt.

Astfel, pentru prima dată s-a argumentat posibilitatea creării unui astfel de sortiment pentru nișa geografică carpato-danubiano-pontică și s-a reușit crearea unor soiuri cu însușiri de pionierat – calitate, inclusiv apirenție, productivitate, maturare timpurie, utilizare diversă, rezistență la factorii abiotici și biotici nefavorabili: Moldova, Codreanca, Pamiati Negrulea, Struguraș, Decabrischii, Urojanăi, Apiren alb, Apiren negru de Grozești, Apiren roz, Apiren roz Basarabean și Apiren roz timpuriu.

În general, practica ameliorării genetice la plantele de cultură a dovedit că progresul semnificativ este determinat de prezența unei diversități genetice favorabile și de aplicarea unor (bio)tehnologii eficiente de prebreeding și breeding. Astfel, programul prebreeding, conceput de noi, este orientat la crearea populației de descendenți cu grad diferit de apirenție, utilizare diversă, inclusiv procesare industrială, cu rezistență avansată la factorii defavorabili ai mediului ambiant, și constituie preocuparea cardinală în utilizarea biodiversității vîtei de vie. Primele rezultate de pionierat fiind o serie de genotipuri omologate deja, unele din ele brevetate, care, concomitent servesc și material biologic inițial pentru actualele și viitoarele programe prebreeding și breeding. „Explozia” de soiuri de struguri pentru masă, obținute recent în Ucraina și Rusia cu utilizarea în calitate de componente parentale a soiurilor noi create în Republica Moldova confirmată valoarea și potențialul lor în calitate de material prebreeding.

Și în continuare, în programele de ameliorare, dezideratele principale rămân soiurile cu bob mare, apiren, strugure atractiv, maturare de consum extratimpurie și timpurie. Diversificarea materialului biologic inițial se efectuează urmând schemele de încrucișări: genotip rezistent x genotip rezistent apiren, genotip rezistent x genotip sensibil apiren și genotip sensibil x genotip rezistent. În scopul accelerării și sporirii randamentului procesului de ameliorarea cu obiectivul apirenție s-au inițiat încrucișări după schema genotip apiren x genotip apiren. Astfel, în programul de ameliorare au fost incluse, în calitate de componente materne, soiurile introduse din alte areale geografice Prezental, Doci Nimranga, Madlen Angevin, Ceauș alb ș.a., soiurile vechi autohtone Coarnă neagră, Coarnă roșie, Damașin galben, Negru de Căușeni ș.a. Componentele paterne au inclus soiurile apirene Centennial seedless, Beauty seedless, Calina, Bessemeană Magaracia, Romulus, cât și soiurile noi create în Republica Moldova - Apiren alb, Apiren roz, Apiren extratimpuriu, Apiren negru de Grozești, Apiren roz Basarabean, Kișmiş lucistai. În consecință, din multitudinea de genitori evidențiați, în programul de hibridări la etapa inițială a fost implicat un segment din întreaga gamă de resurse genetice, variate după originea geografică, genetică, forma și culoarea bobului, direcția de utilizare, rezistența la factorii nefavorabili ai mediului.

**CREATION OF HIGHLY PRODUCTIVE EARLY RIPENING LINES OF SUNFLOWER
(*Helianthus annuus* L.) FOR CONDITIONS OF BELARUS**

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Sunflower (*Helianthus annuus* L.) is the new, very actual cultivated crop for Belarus especially as the source of high quality vegetable oils.

The creation of early ripening and ultraearly hybrids which will provide maturing of seeds by the first decade of September is necessary for our conditions. Thus the main problem in breeding of this culture in Belarus is the obtaining of high productivity at the reduced period of vegetation of plants, because early ripeness and oil content are in negative correlation and also there is a close correlation between productivity and duration of the vegetative period. This problem is partially solved by the selection of highly productive lines with the short interphase periods, especially with the shortest seedlings-flowering period in early inbreeding generations.

The purpose of our work was the creation and the assessment (evaluation) of new early ripening lines of sterility maintainer and restorers of fertility. For creation of new initial material convergent crossings between three self-pollinated M685 (1)/07B, M603 (6)/09B and M605/04B lines were carried out in 2009-2011, with the subsequent self-pollination and selection of plants with the earliest flowering.

As the result of breeding we obtained sunflower plants varying in height (50-160 cm), length of the interphase period seedlings-flowering (40-63days), oil content of seeds (36,7-55,3%) and another important agricultural characters. All studied maternal lines and lines of fertility restorers demonstrate good tolerance to diseases under natural conditions of cultivation, in particular, to Sclerotinia sclerotiorum.

The earliest forms were characterized by the short interphase period of seedlings- flowering: 40-45 days, however, they had a low oil content of seeds (36,7-39,5%). Also, they had few leaves on a stalk (8-10 pcs). And this character along with the weight of 1000 seeds and quantity of seeds from a basket are the main components of seed productivity and criterion of an assessment of selection material. Plants of this group of genotypes were short-growing (65-80 cm).

Also we obtained plants with longer interphase period of seedlings-flowering: 49-55 days. For these maintainer sterility plants rather high rates of oil content of absolutely dry seeds (47-55,3%) and, in most cases, the weight of 1000 seeds (55-65 g) were observed. Height of plants was 120-130 cm. Plants had an optimal quantity of leaves at the stalk (16-20 pcs).

Maintainer sterility lines with the best agricultural characters were ed on *Helianthus petiolaris* cytoplasm.

Molecular genetic studies using SSR-markers for identification and differentiation of lines and hybrids of oilseed sunflower were initiated.

The breeding program of sunflower carried out in the laboratory of cytoplasmic inheritance of the Institute of genetics and cytology NASB allowed to create new, industrial hybrids – F₁ Poisk (2008), F₁ Agat (2010), F₁ Belorusski ranni (2015), and also a number of new advanced lines.

THE STUDY OF ORGANELLAR GENE EXPRESSION IN BARLEY

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The nuclear-cytoplasmic interactions greatly define the future of developing plant organism, especially different types of hybrids. Usually we can see the consequences of disturbances in nuclear-cytoplasmic interactions in such phenotype characters of plant as vigour, sterility, grain yield and etc. But what happens at the level of gene expression?

In order to evaluate the expression of genes in cytoplasmic organelles and nucleus in barley we elaborated special macroarray (microarray) system consisting main chloroplast, mitochondrial and some nuclear related genes. Totally the level of transcripts from each of 65 genes was studied. The obtained data were verified by PCR with cDNA. Particularly, Real Time PCR was performed for mitochondrial –cox1, cob, chloroplast – psbA, psbD, psaA genes. RLI was taken as control gene. Also we checked the level of hsp17 gene transcript as the marker of sensitivity to stress in different nuclear-cytoplasmic combinations of alloplasmic barley lines.

Initially the expression profiles of various mitochondrial and chloroplast genes were evaluated in alloplasmic barley lines and their euplasmic analogs in optimal and temperature stress conditions. We studied the way of changing of each gene from total RNA pool. The level of transcripts of the same gene depended on precise nuclear-cytoplasmic combination – in some cases it was stable, in another it reduced or increased.

The level of psbA transcript in some lines was reduced after stress, but it didn't changed in others. It coincided with the data about D1 protein (psbA gene product), that it was extremely sensitive to unfavourable conditions, in our case- temperature. We also investigated the level of transcripts of nuclear hsp 17 gene encoding heat shock protein. It was shown that heat shock dramatically enhanced the expression in all lines, but the range of changes depended on lines.

Consequently, the plant genes expression depends on precise nuclear-cytoplasmic combination and environmental factors.

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CARACTERISTICA SOIURILOR DE TOMATE DUPĂ CARACTERELE ECONOMIC VALOROASE

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The variety Anatolie was created as a result of intraspecific hybridisation, (variety Potoc x variety Nota). This hybrid has been reproduced by *in vitro* culture, as a result of which were obtained regenerants, reducing the process of improvement with 2 years, (2 generations). The individual selection was made from F₄ generation. Plants with growth type determinant, well developed with 4-6 ramifications. The main length of sprig 55-65 cm. The length of internodes at I-IV inflorescence (4,4, 6,8)cm. The leaves are ordinary type 1, 24-26 cm. length, 16,5-19,0 cm., width. The colour of leaf is dark green. Leaf veins are pinnate. The position of the leaves in relation to the central axis - horizontally oriented. Inflorescence type (2nd and 3rd legate) multipara prevails. The length of inflorescence is 8 cm. The first inflorescence appears after 4-5th node, following ones after 1-2. Yellow flower with diameter of 1,7-2 cm, opened with 5-6 petals, normal sepals. Number of flowers on 1-st panicle 6,5 - fruits 5,0. Second panicle 6- flowers, 5-fruits, third panicle 6,7 flowers – 6 fruits. Number of flowers on main stem 19,5 - fruits 15,6. On plant, flowers - 100 - fruits 85. The pubescent of style is missing. The pedicel of fruit is without knees. The length of pedicel from the point of abscission till the calyx 1,3-1,5cm. The fruit with weight of 65-90 grams, rounds – little oblong, uniform, wrinkle-free at peduncle. The colour of unripe fruit is green, at maturity dark red. The height of fruit is 4,84cm, diameter of 4,90 cm. At the base of the fruit with medium dent, top of fruit surface plane, at some fruits little sharp. Pericarp thickness 0,50-0,60cm, the thickness of the pulp 3,80-3,95 cm. Number of seminal lodge - 3. Number of seeds in the fruit - more than 100.

Vegetative period 118-125 day, tardy variety. The fruits contain dry substance 5,3-7,0, sugars 4,9-5,1%, ascorbic acid 35,6-59,5%, mg/%. Titrated acidity 0,32-0,35%. The general harvest 43,7-47,3t/ha, the percentage of control group 97 %. The share of fruit products 94,07%. The variety is resistant to low temperatures, cultivation by seedling and seed culture.

The variety Iulihirsutian has been obtained by application of intraspecific hybridisation between spontaneous species *Solanum Lycopersicum hirsutum* var *glabratum* C.H.Mill. with small green fruits, with glandular hairs pronounced and the variety of culture *Solanum Lycopersicum* Prizior, with the fruits of medium size, colour red, through embryo culture and ova undeveloped *in vitro*. Individual selection of the F₆ generation. In variety prevails determined type plant, with normal healthy aspect, well developed with 4-6 branches of rod on main stem. The length of main sprig is 55-70cm. The length of internodes between I-IV inflorescence is of (6, 8, 8, 10) cm. Number of nodes 8-9.

The usual type 2 of leaves, de 27-30cm. length, 24-26cm width. The colour of leaf is dark green , embossed. Leaf veins are pinnate. The position of the leaves in relation to the central axis - horizontally oriented. Inflorescence - intermediate (2nd and 3rd legate) type. Flower is yellow, with a diameter of 2-2,5cm, completely open with 5-6 petals, sepals at the same level with petals. Simple inflorescence of 5-7 flowers. The first inflorescence appears after 4-5th node, following ones after 1-3. The pedicel of fruit is without knee joint. The length of pedicel from the point of abscission till the calyx (sepls) 2,5-3cm. The fruit: crimping of the peduncle is absent. The unripe fruit is light green, at maturity the colour of fruit is dark red. Number of seminal lodge in fruit - 3. Number of seeds in the fruit - more than 100. The seeds are oval - rounded, with hairs with gives them silver colour or more grey according to extraction. The number of seeds in a gram range from 300-370.

The fruit has a weight of 95-100 grams, round, uniform. Fruits with pericarp and the inner pulp fleshy. At the base of the fruit with medium dent. Top of fruit surface plane. The duration of the period of vegetation from emergence of green plants till maturity 76 days, early variety.

The content of dry substances in the fruit 6,24% - the percentage of control group 6,02%, sugar 4,72-5,75%, vitamin C 45,6 - 57,5 mg/%, titrated acidity 0,35-0,41mg/%. Demonstrates a general harvest of 43- 47t/ha, with 10 % more major as the standard form, commodity production 86,9%. The variety is productive, with high taste qualities, resistant to drought. It is recommended, for seedling growth, fresh and consumption and industrialization. It is more resistant tol Stolbur virus than the standard variety Elvira.

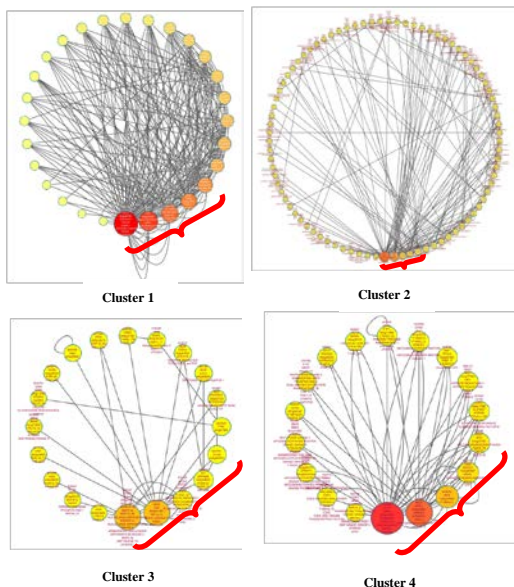
THE INTERACTION NETWORK BETWEEN GENES INVOLVED IN DEFENSIVE RESPONSE OF SUNFLOWER (*HELIANTHUS ANNUUS*) TO BROOMRAPE (*OROBANCHE CUMANA*)

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Sunflower is one of the main crops in Moldova and regarding area of cultivation it occupies third place after corn and wheat. Broomrape is a sunflower parasite which can cause up to 90% of production losses that's why is very important to know more about genes involved in sunflower defensive response and especially about connections between these genes. The goal of our research was to create a functional interaction network between 81 genes involved in defensive response of sunflower to broomrape taking into consideration their first neighboring nodes. The network has been created in *Cytoscape* using sequenced *Arabidopsis thaliana* genome as a model. It has been organized in clusters by using *clusterMaker* app, *Markov* clustering algorithm and 1,5 granularity parameter. The results highlighted four important clusters. In each cluster have been identified and functional annotated the first 5 most important genes (hubs). According to our analysis *Cluster 1* and *Cluster 4* with 28 nodes, 248 edges, respectively 18 nodes, 55 edges are involved in protein kinase signaling which triggers fundamental cellular processes such as growth, differentiation, stress response, apoptosis, and survival. *Cluster 2* with 85 nodes and 161 edges mediates such pathways as protein catabolism, trans - membrane transport, metabolism of reactive oxygen species (ROS), regulation of hypersensitive response, defensive response to biotic stress. *Cluster 3* (20 nodes, 28 edges) is involved in cellulose, starch and galactolipids biosynthesis, photo - and gravitropisms and also in the metabolism of ROS.

In conclusion we can say that two clusters (1, 4) mediate especially the detection of biotic stimulus and trigger the kinase signaling cascade which in turn probably regulates the proteins involved in defense response. Other two clusters (2, 3) are mostly involved in ROS metabolism and is well known that plants use ROS toxic properties to fight off invading pathogens (O'Brien et al., 2012). In the case of parasitic attack, polysaccharide biosynthesis such as cellulose and lignin may be used by sunflower to develop a mechanical barrier against aggressor in order to prevent the spread of its haustorium.



Four clusters resulted after network clustering

**GENETIC VARIABILITY OF OIL QUALITY COMPONENTS IN SUNFLOWER AS A
FUNCTION OF DEVELOPING HYBRIDS WITH NOVEL OIL**

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Sunflower is one of the most important oil crops in the world, and its oil is one of the highest quality vegetable oils. Oil quality is determined by the fatty acid composition, tocopherol content and type, phytosterols, carotenoids, and some other compounds. Standard sunflower oil contains linoleic, oleic, palmitic, and stearic fatty acids as well as several other fatty acids that are found in traces.

The objective of this paper was to make a review of the genetic variability oil quality components in sunflower using own results and those of other authors.

Standard sunflower oil is linoleic in type, but using induced mutations genotypes have been developed that have high levels of oleic, palmitic, and stearic acids, and the mode of inheritance of these traits (gene number and type) has been determined. Also, some results have been achieved in the study of the mode of inheritance at the molecular level using marker genes.

Standard sunflower oil contains predominantly alpha tocopherols (>95%). Using spontaneous mutations the genes *tph1*, *tph2*, and *tph1tph2* have been discovered that control different levels of alpha, beta, gamma, and delta tocopherols. Also, Spanish researchers used induced mutations to obtain mutants with high levels of beta and delta tocopherols. In one of our own studies, the restorer line RHA-S-59 was found to contain only gamma tocopherol (100%), while 20 other restorer lines had only alpha tocopherol (100%).

It has been scientifically proven that phytosterols (campesterol, stigmasterol, and beta-sitosterol) also play an important role in determining oil quality and that different genotypes have different levels of these substances.

It has been shown that in genotypes that have high levels of oleic acid coupled with high levels of beta, gamma, or delta tocopherol a certain synergy occurs that increases oxi stability up to 15 times compared to standard sunflower oil.

Using the existing genetic variability of components that determine oil quality it is possible to develop sunflower hybrids with novel oil. Thus far, the most has been done in the development of high-oleic hybrids.

ASSESSING THE IMPACT OF COMMON ACTION OR LONELY OF BARLEY STRIPE MOSAIC VIRUS AND GAMMA RAYS ON WHEAT PRODUCTIVITY INDICES

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Classical methods of genetics and breeding include the use of experimental mutagenesis to obtain new forms, which allows widening the range of variations to the original forms. Phytopathogenic viruses represent a recombinogenic factor that ensures induction of genetic variability in a number of crops. In this paper, we have proposed the evaluation of joint action and / or separate of Barley Stripe Mosaic Virus (BSMV) and gamma rays on variation of productivity in progeny populations of spring wheat.

As biological material were used two genotypes of spring wheat (Arnaut 7 and Israel). The seeds were irradiated with gamma irradiation appliance RXM - V -20 source - 60Co in doses of 50, 100, 150 Gy. Plants of experimental variants were infected mechanically in 2-3 leaves stage with BSMV. Uninfected healthy plants and free of viral germs served as control variants. Determined was done by the negative contrasting by electron microscopy method. The plants were grown under field conditions according to standard techniques. Evaluation of the role of viral infection and / or gamma rays on variations of quantitative indices was performed under trifactorial dispersion analysis (ANOVA), the power of influence (PI, %) and coefficient of variation (CV, %). Statistical analysis was performed using Statgraphics Plus 2.1 software package.

The results show that development of wheat plant is affected by the action of both factors and their interaction. In most cases, the differences between control and experimental variations were significant. The impact of sources of variation was positive in the case of low-dose irradiation 50-100 Gy and negative for 150 Gy dose, as well as in the case of solitary action of viral infection. Seed germination was significantly influenced by both factors, and the growth energy turned out to be several times higher in the experimental variants, but over time this difference has subsided and germination percentage was significantly higher in the control group. Statistical analysis for most analyzed biomorphological indexes (plant size, last internode length, number of tillers / plant, number of spikelet / spike etc.) in the first generation, certify that the share action of BSMV is higher for plants treated with gamma rays. Based on the ANOVA test, we find that the influence of the viral infection on the trait „fertile tillers per plant” varied by genotype: from 39,22% (Arnaut 7) to 52,87% (Israel), and the interaction of factors gamma rays - BSMV had a 24,21% share of influence only for one of studied genotype (Israel), the differences being significant at 99,9%. At that time, the lonely action of the gamma ray certified non-significant differences and included a share of 1,56% (Israel) to 5,27% (Arnaut 7). We note that previous research conducted in our laboratory found that barley stripe mosaic virus causes repercussions with a maximum power in the first generation for plant size, last internode length, keeping major variation amplitude in the third generation as well for offspring of spring barley and power of influence is genotype specific.

The descendent populations from variety Arnaut 7 following the action of lonely or common action of viral infection and gamma rays were subjected to individual selection. Thus were selected plants that are characterized by a greater number of spikelet in the spike (≥ 20) and were evaluated by determining productivity indices: spike size, spikelet number / spike, grain number / spike, main spike weight, 1000 grains weight and healthy grain number / spike. Variance analysis of productivity indices of population in fourth generation shows that the action of BSMV has not a significant impact on the traits spikelet / spike and 1000 grain weight, but on the height on the spike has a significant influence power at 99 % ($F = 7,95$). Meanwhile lonely effect of gamma ray was significant at 99 % for 1000 grain weight ($F = 5,73$), and not significant for the other two traits (spike height and spikelet / spike). Note that the interaction of factors viral infection - gamma radiation resulted in significant rates of change for all analyzed indices: spike size and weight of 1000 grains at 99,9 % ($F = 15,03$ and $F = 8,02$, respectively) and number spikelet / spike at 95 % ($F = 2,66$).

In conclusion, the variability productivity traits in wheat populations descended from plants subject to common action and / or separate BSMV and gamma rays is influenced primarily by viral infection, interaction of factors BSMV – gamma ray and is specific for productivity index and generation progeny analyzed.

**MOLECULAR TOOLS FOR TRACING BIOTROPHIC PATHOGENS
IN CROP PLANTS**

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Biotrophic oomycetes such as downy mildews and white blister rusts are major diseases of many crops worldwide. Symptomless phases in their endophytic life cycle, resting spores in seeds and soil, and fast developing genotypes of diverse pathogenicity or fungicide resistance impede the employment of effective control measures in agriculture. Recent genetic studies afforded DNA sequence information suitable to develop PCR-based tests which allow screening of plant material for contamination with pathogens and enable monitoring of the diversity in pathogen populations. The usefulness of such assays will be illustrated with examples from sunflower, grapevine and tobacco cultivation.

ENTORNOPATHOGENIC VIRUSES DIVERSITY AND USE AS BIOINSECTICIDES

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Insects constitute the most divers group of living things on the world. They can be found practically in every terrestrial and fresh-water habitat. Some have been associated with humans throughout their evolution and play an important role since the beginning of the agriculture, by becoming natural competitors for the food humans grow. Many insect species are called pests because they cause damage to humans, most frequently as competitors for the food that humans grow. Although many measures have been used for this purpose, an efficient technique was developed by the middle of the 20th century, which made obsolete and impractical any other kind of control: the use of chemical insecticides.

Consequences of the practically exclusive use of this technique soon were apparent. New control alternatives have been developed, especially those environment-friendly, such as biological control. Three main groups of organisms constitute the backbone of biological control: parasitoids, predators, and pathogens. The first two groups are mostly represented by other type of insects, while the third one is constituted by infectious microorganisms causing lethal or deleterious effects on susceptible individuals. These can be viruses, fungi, protozoa, and nematodes, and are frequently used as "bioinsecticides", which are sprayed on pest populations. Nowadays, the market for bioinsecticides is about 2.5% of the total insecticide market and it is estimated that will rise to 4.2% by 2020. Although viruses constitute an important component of this type of agents, especially the baculoviruses. These constitute the most diverse group of entomopathogenic viruses which have been found practically exclusively on insect populations, mainly within the orders Lepidoptera, Hymenoptera, and Coleoptera.

As pest control agents, baculoviruses do not leave harmful residues in the environment and resistance against the viruses is not observed in the field. Another advantage of these viruses is that they can persist in susceptible insect populations leading to a dynamic control of the pest. The baculovirus used in this project, is commercially available in many European countries as a bio-pesticide for the control of *Hypantria cunea*, *Agrotis segetum*, *Mamestra brassicae*. The environmental and ecological impacts of baculovirus products are mirrored by characteristics of their pathogenesis and host range. In essence, every study that is required to assess their potential for environmental toxicity will be influenced by their limited host range and lack of infectivity to non-target animals. Baculoviruses have recently been included in the qualified presumption of safety (QPS) list authorized by the European Food Safety Authority (EFSA) (Leuschner et al., 2010) panel on Biological Hazards (BIOHAZ) (EFSA, 2009). Following a review of literature, EFSA concluded that baculoviruses are safe for animal and human consumption and are, therefore, acceptable for use in the control of insects that cause damage to plants (EFSA, 2010). Given that all published reviews unequivocally state that baculoviruses are safe and support their use as low-risk biological control agents for the control of insect pests, we propose that human and environmental toxicity.

Commercial production of baculoviruses for use as biological control agents of insect pests is carried out worldwide at different scales depending on the market. In 2014, there are more than 430 registered biopesticide active ingredients and 1320 active product registrations. Over 50 baculovirus products have been used worldwide as microbial insecticides. At Institute of Genetics, Physiology and Plant Protection of the Academy of Science of Moldova are prepared the bioinsecticides for use in Republic Moldova, mostly for the control of insect pests. In order to reduce the population of insect it is recommended utilization of the ecologically inoffensive preparations Virin-ABB-3, Virin-OS, Virin-HS-P, which is an efficient preparation for combating this pest in agricultural, ornamental and forest biocenosis.

EXPERIMENTAL EVALUATION OF THE MUTAGENIC ACTIVITY OF PLANT GROWTH REGULATOR REGLALG-1

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REGLALG-1 – is a preparation of the natural origin, registered in Moldova as a plant growth regulator for seeds treatment (wheat, soybeans, corn) and for use on table and vine grapes.

REGLALG-1 represents 20% and 50% solutions in ethanol or butanol of dried extract of freshwater green algae *Spirogyra spp.* The active ingredient of the product – is mixture of polyunsaturated fatty acids, aldehydes, ketones, aldehydoketonov.

The purpose of the study – is evaluation of the mutagenic activity of the preparation REGLALG-1 in *in vitro* mammalian chromosomal aberration test.

The *in vitro* chromosomal aberration test was performed on primary cell cultures - human peripheral blood lymphocytes. Human peripheral blood lymphocytes were obtained from adult (30-35 years of age), non-smoking donors. Whole blood treated with an anti-coagulant was cultured in Eagle's medium supplemented with 10% horse serum and streptomycin (10 EA/1 ml medium). Chromosome preparation involves hypotonic treatment of the cells, fixation and staining (OECD/TG 473).

In our study were evaluated the next three test concentrations: 0.5, 1.0 and 2.0 µg/ml (the highest test concentration that produced cell death and reduction the mitotic index not higher than 50%). The test concentrations were diluted in Eagle's medium and added to the test culture in the volume - 0.2 ml per vial, after three hours of incubation. Exposure conditions: 69 hours at 37°C. Cell cultures were treated with colchicine (1 µg/ml medium) for 2.5 hours prior to the end of incubation.

Results. The 200 well-spread metaphases were scored per concentration and control. The results are presented in the table.

Table1. Number of cells with CA and type of CA for treated and control cultures

Type aberrations	Test concentration (µg / ml)			
	0 (control)	2	1	0,5
Chromosomal aberrations				
Total	2	2	0	1
on 100 metaphases	1,0	1,0	0	0,5
including: paired fragment				
Total	2	2	0	1
on 100 metaphases	1,0	1,0	0	0,5
Chromosomal exchanges				
Total	0	0	0	0
on 100 metaphases	0	0	0	0
Chromatidal aberrations				
Total	3	2	1	1
on 100 metaphases	1,5	1,0	0,5	0,5
including: point fragments	0,5	0	0	0
breakage of single chromatids	1,0	1,0	0,5	0,5
Total number of aberrations	5	4	1	2

The results of the research showed follow:

- the mean frequency of aberrant metaphases per group was – 2; 0.5 and 1.0%, respectively, at doses of 2, 1 and 0.5 µg/ml. In control the spontaneous frequency of chromosomal aberrations was not high than 2.5%.
- both in the control and experimental groups the aberrations were mainly represented by point and single fragments, i.e. chromatid-type aberrations. They accounted from the total number of CA 75% - in the control and 50, 50 and 100%, respectively, in doses.

Statistical analysis of the level of CA in blood lymphocytes showed no significant differences in these parameters in the experimental and control groups. It must be mentioned that even the most studied concentrations of REGLALG-1 reduced the number of CA compared with the control.

Thereby, REGLALG-1 does not cause structural chromosomal aberrations in cultured mammalian cells.

EST-SSR GENOTYPING OF YELLOW LUPINE (*LUPINUS LUTEUS*) IN BELARUS

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Yellow lupine (*Lupinus luteus* L.) is a high-protein crop capable of symbiotic nitrogen fixation, better than any other species of lupine grows in sandy soils.

Progress in improving the efficiency of lupine breeding programs is largely dependent on the improvement and application of modern molecular techniques and their appropriate combination with traditional methods. However, there is little information concerning genetic markers of *L. luteus* agronomic traits. Currently the method of assessing genetic diversity of this crop based on polymorphism of intragenic microsatellite repeats (EST-SSR) is known. The application of this method for searching genetic determinants of yellow lupine valuable properties seems reasonable.

The objects of this study were 19 samples of yellow lupine: Maculosus, Maculatus, BSKHA 433, BSKHA 365, BSKHA 555, BSKHA 556, Magikan, Demidovski, CWN-K-1, CWN-K-2, CWN-K-3, Zhemchug gray, BSKHA 658, P-13 (PN), B-19 (PN), Rozant, Myth, Prestige, Nadezhny, provided by the Department of Breeding and Genetics BAA.

To determine the genetic polymorphism eight EST-SSR markers (2itg03938, 2itg27515, 2itg14694, 2itg45631, 2itg26293, 2itg13638, 2itg50945, 2itg20349) were selected. The index of the polymorphism information content (PIC) was calculated.

Polymorphism has been detected in seven of the eight used EST-SSR markers. Markers 2itg03938, 2itg14694, 2itg50945 revealed the presence of 3 alleles, markers 2itg27515, 2itg45631, 2itg26293, 2itg20349 - 2 alleles. Marker 2itg13638 revealed no polymorphism in the given collection. The highest polymorphism level was found in the markers 2itg14694, 2itg50945 (PIC value - 0,66 and 0,62 respectively). PIC values for markers 2itg03938, 2itg27515, 2itg45631, 2itg20349 were within 0,36-0,48. That corresponds to a moderate polymorphism level, sufficient to differentiate the studied forms. The marker 2itg26293 had low PIC value, that indicates its poor suitability for typing the given samples collection.

Based on these results we can conclude that EST-SSR markers 2itg14694, 2itg50945, 2itg03938, 2itg27515, 2itg45631, 2itg20349 can be used to differentiate various yellow lupine samples in our collection.

TRANSCRIPTIONAL ACTIVITY OF PATHOGENESIS-RELATED GENES DURING SUNFLOWER-DOWNY MILDEW INTERACTION

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Pathogenesis-related proteins are essential component of plant defense responses, which could prevent various pathogen invasions and are accumulated locally in the infected and surrounding tissues, and also in distant uninfected tissues (mechanism of prevention of further infection). Currently, PR-proteins were classified into 17 families according to their biochemical properties and functions.

PR5 proteins are the family of thaumatin-like PR proteins, which expression is induced in plants under several pathogen attacks. Accumulation of these proteins correlates with development of systemic acquired resistance (SAR), thus, these are considered markers of SAR.

Another important group of PR proteins is defensins. Plant defensins are small, rich in cysteine, high-stable peptides, which take part from the innate immunity directed towards ensuring the protection against fungal pathogens. The biological activity of defensins not limited only to the antifungal and, antibacterial activity, they also possess insect amylase- and protease-inhibitor activity. Transgenic plants with overexpression of defensins are highly resistant to fungal pathogens. Moreover, defensin is considered marker of JA mediated defense response.

Thus, the aim of our study was estimation of *PR5* and *defensin* genes expression levels in leaves of five sunflower genotypes (Drofa Rf, Drofa CMS, Drofa F₁ hybrid and two isogenic lines – 393A / 393B) in normal and infected with downy mildew plants grown in field.

Total RNA was isolated using TRI Reagent (Ambion), 0,6 µg of total RNA was treated with DNase (*Promega*) and first-strand cDNA was synthesized with RevertAid Reverse Transcriptase and Random hexamer and Oligo-dT18 primers (*Thermo Scientific*) according to the manufacturer's instructions. The primers for *PR5* and *defensin* genes were designed using Primer3Web v. 3.0.0. software from the *Helianthus annuus* mRNA complete coding sequences (GenBank: AF364865.1 and AF364864.1 respectively). Gene expression was estimated with RT-qPCR using Maxima SYBR Green/ROX qPCR Master Mix (*Thermo Scientific*).

Evaluation of transcript levels of *PR5* gene showed a significant up-regulation of expression in strongly and medium infected plants of all investigated genotypes. The most evident increase was observed in systemically infected Drofa CMS line (32,67-fold higher than the control group). Also, a 2-fold up-regulation was recorded in weakly infected plants of this genotype. Similar reaction was observed in non-symptomatic parts of infected plants. Exception presented Drofa F₁ genotype, which was characterized by 8,5-fold down-regulation of expression.

Thus, *PR5* gene transcriptional activity was up-regulated in strongly and medium infected plants, ensuring protection of uninfected tissues towards the spread of infection.

Unlike *PR5*, *defensin* gene transcriptional activity was characterized by down-regulation in non-symptomatic parts of infected plants and by up-regulation in weakly infected plants of all studied genotypes and those strongly and medium infected of Drofa CMS and F₁ genotypes. However, the most pronounced down-regulation of *defensin* expression was observed in strongly infected isogenic lines 393A and 393B – 38- and 97,5-fold respectively. Drofa CMS line was characterized by maximal expression values that exceeded those of the control group 3,73-fold in weakly, 12,8-fold in medium and 1,56-fold in systemically infected plants.

Obtained results revealed that *defensin* gene expression is up-regulated in early stages of infection and only in tissues invaded by pathogen, when contact between plant tissue and pathogen take place.

Comparative analysis of *PR5* and *defensin* gene expression showed up-regulation of their expression: *PR5* in systemically and medium infected plants and non-symptomatic leaves of weakly infected plants and *defensin* in weakly infected plant, which could be explained by participation of these genes in different signaling pathways. Thus, at early stages of infection the immune response is provided by *defensin* gene activated in JA-signalling pathway, but in late stages and in healthy tissues (distant from the site of infection) it is mediated by *PR5*, activated in SA-signalling pathway.

PAL – KEY ENZYME IN *HELIANTHUS ANNUUS* L. DEFENSIVE RESPONSE TO *OROBANCHE CUMANA* WALLR.

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Phenylalanine ammonia-lyase (PAL) catalyzes the nonoxidative deamination of L-phenylalanine to *trans*-cinnamic acid, which is the first step in the phenylpropanoid pathway and an important regulation point between primary and secondary metabolism. *Trans*-cinnamate is the precursor of numerous phenylpropanoid compounds that fulfil various functions as mechanical supports (lignins), as protectants against biotic and abiotic stress (phytoalexins, antioxidants, and UV-absorbing compounds) (Wang and Zhang, 2010) and biosynthesis of SA (Dehghan et al., 2014). Lignin, which is synthesized from phenylpropanoid compounds, is a major structural component of secondary thickened cell walls in the plant vascular system essential for stem rigidity and for conducting water, minerals, and photosynthetic products through the plant. Cell wall reinforcement is the first barrier of defense at contact with the pathogen and haustorium penetration in the host plant roots. Lignin has involved in a cell wall integrity maintenance mechanism (Denness et al., 2011). PAL has been extensively studied because of its role in plant development and its synthesis in response to a wide variety of environmental stimuli.

The presence of PAL has been reported in diverse plants including certain algae, fungi and a few prokaryotic organisms *Streptomyces*. In most plants, PAL is encoded by a small gene family of 3-5 genes. Exceptions to this are the potato PAL gene family, which is comprised up to 40-50 genes and the loblolly pine PAL, which is encoded by a single gene. The PAL gene family in tomato consists of 26 family members, in soybean – 8 genes, maize – 20, rice – 12, grapevine – 16 and *A. thaliana* – 4 (Rawal et al. 2013). Currently in sunflower is identified one PAL gene (Y12461, GeneBank).

PAL enzyme has been isolated and characterized from a number of plant species. Mostly, studied PALs range in size from 300 to 340 kDa in native molecular mass. Some examples of exceptions are -showed masses of 152 kDa in *Ocimum basilicum*, 250 kDa in *Helianthus annuus* (Jorrín et al., 1988), 266 kDa in *Fragaria ananassa*. PAL is normally a homo-tetrameric protein consisting of four identical subunits. Hetero-tetrameric PAL as a complex of two hetero-dimers has been described for *H. annuus* (2 × 58 kDa and 2 × 68 kDa) (Jorrín et al., 1988).

There are many studies which demonstrate PAL involvement in defensive response to biotic stimuli in crops. Accordingly to Letousey et al. (2007), the transcript levels of PAL and other lignin synthesis genes (*c4h* and *chs*) were rapidly and strongly increased under broomrape infection in sunflower. In inoculated roots of the resistant genotype (LR1), the phenylpropanoid metabolic pathway, leading to synthesis of SA, phytoalexin coumarins or lignins were induced. High levels of lignins synthesis were also linked to the degree of sunflower resistance to *Orobanche cumana*.

The same results were obtained for another studied pathosystem (*Helianthus annuus* - *Plasmopara halstedii*), where the strong induction of PAL in sunflower hypocotyl showed significant increase of PAL transcript levels during incompatible interactions but not in the compatible ones, suggesting that regulation of this mRNA is an important component of the resistance mechanisms in sunflower (Mazeyrat et al. 1999).

The assessment of transcription levels of PAL genes evaluated in sunflower hybrids allow evaluation of quantitative resistance to *O. cumana* artificial infection. RT-qPCR revealed the changes in gene expression to roots of host a three phases after parasite attachment. Correlation between these data, PAL enzyme activity and histochemical analyses for determination of lignin content in infected and uninfected plants enable understanding and explanation of the defense response activation.

AGROBIOLOGICAL PECULIARITIES AND FODDER VALUE OF THE NEW
PERENNIAL FORAGE LEGUME SPECIES IN MOLDOVA

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The concerns for both protection of the environment and achieving high quality food production have encouraged an increase in organic farming. Forage legumes provide the basis for organic farming through much of the world. Their importance has arisen principally because of atmospheric nitrogen (N) fixation by rhizobial bacteria growing symbiotically with the legumes. With reduction in the real prices for animal products and the prospects of further future reductions with moves towards more liberalized world trade, it has become more important to seek ways of reducing the unit cost of producing milk and meat. Forage legumes have much potential for reducing costs with the opportunity to reduce inputs of fertilizers, because of N fixation and concentrate feeds, because of high nutritive value.

After over six decades of scientific research on plant resource mobilization, the Collection of non-traditional fodder plants of the Botanical Garden (Institute) of the ASM, which includes over 300 species and cultivars, including 70 fodder leguminous plants, has been founded. The seed material was collected as a result of expeditions and international exchange of seeds.

As a result of the research on agro-biological peculiarities, it has been established that investigated perennial leguminous species of the genera: *Astragalus* L., *Galega* L., *Lotus* L., *Lathyrus* L., *Coronilla* L., *Medicago* L. (*M. polychroa* Grossh., *M. agropyretorium* Vass., *M. cancellata* M.B., *M. glutinosa* M.B. and *M. trautvetteri* Sumn.) form seeds with dense tegument (hard seeds) and need seed scarification and adequate soil moisture for a uniform germination. In the first year of vegetation, the growth and development rate of these species is slower in comparison with *Medicago sativa* L., some of them reach the flowering stage and their green mass can be harvested only once. The species of the genus *Onobrychis* Mill are characterized by the fact that they require a 2-5 day longer period before the emergence of seedlings on the soil surface and, during the growing season, the growth and development rate is more intense in comparison with *Medicago sativa*, ensuring a good yield of green mass (50 t/ha).

In the next years, the vegetation resumption of the studied species was observed during March-April. As compared with *Medicago sativa*, the species *Astragalus galegiformis* L., *Coronilla varia* L., *Galega orientalis* Lam., *Medicago tianschanica* Vass., *Medicago varia* Mart., *Onobrychis inermis* Steven, *Onobrychis arenaria* (Kit.) DC., *Lathyrus grandiflorus* Sibth. start growing 3-7 days earlier and the species *Lathyrus sylvestris* L., *Medicago falcata* L., *Lotus uliginosus* Schkuhr. start growing 5-14 days later. The species *Astragalus galegiformis*, *Galega orientalis*, *Onobrychis inermis* are characterized by an intensive growth and development rate, which makes possible to harvest the plants in the first half of May or 10-15 days earlier than *Medicago sativa*, the yield achieves during this period 35-43 t/ha green mass. The species *Onobrychis arenaria*, *Medicago tianschanica*, *Medicago varia*, *Lotus corniculatus*, *Coronilla varia* reach harvest time in late May simultaneously with *Medicago sativa* and some species of the genus *Medicago* (*M. polychroa*, *M. agropyretorium*, *M. cancellata*, *M. glutinosa*, *M. falcata*, *M. trautvetteri*), *Lathyrus sylvestris*, *Lathyrus grandiflorus* reach the stage of flower bud formation in the second half of June, which would ensure a permanent provision of green fodder for animals.

The harvested fodder of the studied species has a high content of raw protein (6-7 %), nitrogen-free extractive substances (10-12 %) and a moderate content of cellulose (8-9%), calcium (5,0-10,2 g/kg) and phosphor (1,5-2,2 g/kg), a nutritive value of 0,22-0,27 nutritive/kg units and 2,42-3,26 MJ/kg metabolizable energy. A nutritive unit is provided with 175-280 g digestible protein.

The species of the genus *Astragalus*, *Lotus*, *Onobrychis* during the grazing process don't cause intoxication (flatulence) to ruminant animals and can be used to restore degraded pastures.

By individual selection of the introduced population *Galega orientalis*, were created and registered in the Catalogue of plant varieties of the Republic of Moldova the local varieties *Speranta*, the green mass is used for the preparation of hay; leaves remain on the stem, which helps ensure higher forage value. 100 kg of hay contain 71-74 nutritive units, 750-793 MJ/kg metabolizable energy for cattle and 9,16-12,2 kg digestible protein.

The studied forage legumes species are an important nectar source for bees and entomophagous insects. The species *Astragalus galegiformis*, *Coronilla varia*, *Lathyrus grandiflorus*, *Lathyrus sylvestris*, *Medicago agropyretorium*, *Medicago tianschanica*, *Medicago varia*, *Onobrychis arenaria* can serve as initial material for creating and implementing new varieties of leguminous species for fodder production.

BIOLOGICAL CHARACTERISTICS OF SAFFLOWER TINCTORIAL IN THE MOSCOW REGION.

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As a result at the Center of reservation, maintenance and gene pool of the Selection and Technological Institute of Horticulture and Nursery Sciences Academy, previously named as Moscow branch of N.I. Vavilov Research Institute of Plant Industry (Mikhnevo, Stupino district, Moscow region) was created a cultivar of safflower tinctorial named "Krasa Stupinskaya". The authors: Temirbekova S.K., Kulikov I.M., Kurylo A.A., Norov M.S., Metlina G.V., Postnikov D.A., Ionova N.E. This cultivar is included in the State register of breeding achievements since 01.01.2013. The patent number - 6930.

This cultivar is used as the sideral, phytosanitary, phytomeliorative, stern, decorative and oilseed crop.

These studies were performed at the Center of Plant genetic resources and biological resources (Mikhnevo, Stupino district, Moscow region) in 2010-2014.

Phenological and biometric observations were made during the growing season in accordance with the Methodology of State variety testing of agricultural Cultures (1983).

Oil content and fatty acid composition in seeds was determined in accordance with GOST 10857 «Oilseeds. Methods for determination of oil content".

The results showed that vegetation period from germination to maturity of safflower seeds was 96 days (compared to 110-115 days in previous years). Weight of 1000 seeds – 41,1 g (average dates for 5 years).

Safflower seed productivity averaged over years of research was 0,7 t/ha. It was noted the formation of a higher 1000 seeds weight and total productivity in 2010-2012, 2014 than in 2013.

Analysis of 4 safflower reproductions grown in the Moscow region showed a negative effect of weather conditions on the accumulation of fat in the safflower seeds. In 2010, characterized by higher temperature (5-7 ° C above normal for the entire vegetative period) and low rainfall (186 mm below normal rate throughout the growing season) seed's oil content was 31,2% , that is on 27,6 % higher than in 2013 (3,6 %), which was characterized by excessive precipitation norms at the maturity period (74,8 mm above normal rate).

Determination of fatty acid composition in such cultivars as Krasa Stupinskaya, VIR2933 and Mahallm 260 (Tajikistan) revealed identical fatty acid composition (content of myristic acid – 0,1 %) in the cultivars Krasa Stupinskaya and Mahallm 260.

However, in comparison with other accessions this cultivars contain a little less of diunsaturated linoleic acid – 75,7 % and 75,6 % respectively, in the VIR 2933 – 80,1 %, and the higher content of monounsaturated oleic acid – 13,6 – 13,2 % accordingly in Krasa Stupinskaya and Mahallm 260, VIR 2933 – 10,7%.

It should be noted that Krasa Stupinskaya and Mahallm 260 were characterized by a high content of saturated fatty acids - stearic and palmitic (7,7 – 2,0 % and 7,6 – 2,6 %, respectively), and in the VIR 2933 – 6,9 and 1,5% .

It was determined the effect of wet weather conditions during flowering and seed maturation of safflower tinctorial on the weight of 1000 seeds, yield and oil content.

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NATURAL BIOLOGICALLY ACTIVE PREPARATIONS AS STIMULANTS PHOTOSYNTHETIC ACTIVITY OF PLANTS APRICOT

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The productivity of the plant organism - an indicator caused by a complex series of integrated fixed genetically and phenotypic factors. Here pride of place belongs to the photosynthetic function leaf, the implementation of which depends on the relationship of photosynthesis with all other physiological processes, such as photosynthesis, growth and integration as the basis of a common production process. Growth and photosynthesis determine the nature of ontogeny plants, crop formation is determined ultimately by epigenetic events: neoplasm and development, the physiological condition of the donor and acceptor. Search and study of new and environmentally friendly natural substances capable of regulating these processes underlying the formation of a crop, offers the prospect of managing productivity of plants and has theoretical and practical value.

Which compounds are natural bioregulators of steroidal class glycosides: trigonellozid, capsicozid, melongozid, linarozid isolated in the Institute from plants of the genus *Trigonella*, *Capsicum*, *Solanum* and *Linaria*. Over the years, we studied the effect of these compounds on the rootstock seedlings and grafted plants of different varieties of apricot. In the industrial nursery and fruiting garden Moldovan Institute for Fruit Growing and under lysimeters IGPPP studied features of growth and photosynthetic characteristics and Production processes rootstock seedlings of different genotypes: almonds, zherdel, apricot, peach seedlings of different ages and different varieties of apricot.

The high responsiveness of rootstocks and varieties of annuals Kishinevsky rannii, Kostujensky, Shalah and Nadejda zoned in Moldova, and promising new varieties NJA -42 and Traian by trigonellozid and moldstim (capsicozid) treatment. These natural compounds stimulate the growth of the trunk, shoots, leaves and root system, increase the leaf surface, optimize the accumulation of photosynthetic pigments and metabolic processes that alter the ratio of the mass of the aerial organs and roots towards strengthening the role of atragiryushey root system. Bioregulators increases the importance of the components of photosynthetic productivity, especially net photosynthesis and that is especially important for the nursery, seed quality by 6-7%.

Revealed that young non-bearing and comes into bearing plants apricot and peach 3 - 5 years of age are highly sensitivity to the action of steroidal glycosides capsicozid and melongozid, especially in its combination with microelements zinc and manganese. Response associated with stimulation of photosynthetic activity, that promotes increasing the yield on the test plants. It is shown that the values of the photosynthesis, respiration and transpiration experienced apricot plants usually exceed these values than in the control in the control 1,3 - 1,5 times. The impact of biologically active compounds determined by the age of plants, fruit load and weather conditions during the growing season. Capsicozid regulatory role and its mixture with microelemens persisted until the end of the growing season. There was a positive correlation the intensity of photosynthesis with the processes fruiting. Accounting of crop showed that the number of test plants apricot fruit were similar to the control, at the same time weight per fruit in the control 1,2-1,4 times lower. The yield per plant apricot in variants moldstim, moldstim + Zn and moldstim+ Mn exceeded harvest in control by 16%, 18% and 43% respectively.

The peculiarities of the sink – source relations in three promising varieties of three-year plants apricot plants with different ripening fruits: early variety NJA-32, Olymp grade middle and late ripening variety Sirena. Investigation of the influence of natural prepartate linarozid showed the possibility of regulation of these relations at the level of leaf - shoot - fruit - root. High responsiveness of the studied plants for processing linarozid manifested in enhancing the growth of the aerial and root system, stimulating the accumulation of pigments, the intensity of photosynthetic and metabolic processes, that contribute to a fuller realization of photosynthetic capacity and improve overall productivity.

Thus, the research resulted in detection a high responsiveness of rootstock seedlings and different varieties of apricot on the foliar treatment by natural growth substances trigonellozid, capsicozid (moldstim), melongozid and linarozid, as well as combinations moldstim and melongozid with microelements zinc and manganese. This was manifested in stimulating the growth of aboveground and root system, accumulation of pigments, photosynthesis, increase the values of the components of the production process, that promotes increasing productivity. Based on the data developed and patented recommendations for growing plants apricot.

RAPID PROPAGATION OF MINIROSES IN MOLDOVA

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The history of miniroses is relatively unknown. The only known facts are that a miniroses variety was introduced to England that was a form of *Rosa chinensis minima* (R. Rouletti Correv). By the 1850s it was a popular form of rose (Genders, 1965). A quarter of a century after miniatures were introduced in England and Europe, they lost popularity and disappeared. A century later they were rediscovered by Major Roulet who saw them in a Swiss Alpine village. Henri Correvon propagated the ones he received from Roulet and created the variety Rouletti (Browne, 1974).

In the early 1900s, this species was introduced to the west. Genetically, dwarf miniroses originated from Chinese breeding efforts. The large selection of miniroses we have today is due to propagation and hybridizing of these plants (Dole and Wilkins, 2005).

Many minirose cultivars were patented in Canada, the United States, the Netherlands, and Denmark. To this day, Ralph Moore has done more breeding of miniroses to bring them to their present state of development than any other breeder (Genders, 1965).

In Moldova for the first time were studied, tested and multiplied varieties of miniroses of different heterogeneous groups (*Floribundas*, *Miniature* and *Patio* roses) in different cultivation conditions (*in vitro*, *ex vitro*, in pots and in the open field), identifying resistant varieties and decorative qualities. The miniroses multiply through different ways: division of the bush, grafting, cutting, microcloning. Tissue culture of miniroses is a propagation technique widely used in modern floriculture, because it allows production of many clonal plants from relatively little starting material. *In vitro* clonal propagation of roses is of great commercial value. Using this technique, many pathogen-free rose plants can be produced. It was determined the optimum temperature for rooting, callus formation period, the terms of rootedness "*in vitro*" and miniroses vitrocultivare classic medium base (MB) Murashige-Skoog (MS), as amended by us. Miniroses grown in the open and protected where selected and used appropriate growing media. A liquid culture system using nodal segments was used for shoot proliferation and root induction in *Avon*, *Starina* and *Cri-Cri*, *Baby Masquerade*, commercially-important species of miniroses. For efficient and large scale induction of roots in microshoots, a rooting vessel was designed and developed to facilitate the micropropagation protocol. Micropropagation of miniroses has been reported by many workers, but there is a need to standardize the micropropagation protocol for every cultivars because response of rose under *in vitro* condition is dependent upon genotype. The present work highlights the significance of osmotic potential in relation to enhanced growth and development in liquid cultures, vis-à-vis agar-gelled cultures, especially in relation to root induction during micropropagation. Explants were washed in running tap water for 15 min and for another 5 min in 5% liquid detergent, then washed again in tap water. They were disinfected with 70% ethanol for 30s followed by surface sterilization 2 min and then washed thoroughly in sterile distilled water. Explants were cultured on Murashige and Skoog's medium⁵ supplemented with 3% sucrose, 0.8% agar and growth regulators. The pH of the medium was adjusted to 5.6. Isolated shoots about 2-3 cm long, were rooted on half-strength MS medium with 1,5% sucrose, 0,8% agar and 0,5 mg/l (NAA). After 3 weeks, well rooted plantlets were transferred to plastic pots containing sand and soil (1:1) mixture and kept covered to maintain high humidity for the first one week. After 3 weeks, plants were transferred to the field conditions.

The results argue morfolobiological change of varieties of miniroses and serve as a genetic source for deepening efforts to introduce further new results obtained in this field. Key milestones have been developed on determining the biomorfolobological character of rose (systematic classification, requirements for environmental factors); phytopathogens index; pests and diseases, parasitic, and viral infections, especially with regard to symptoms, pathogen characteristics and that prevention and control; records of evaluation for assessing the decoration.

Important scientific problem addressed in this research work is to determine the morphological research on developing miniroses for the first time we performed in Moldova have emerged from the need to highlight a suitable assortment of 32 varieties of the gene pool used in world practice, to be introduced in Moldova. Generalizing the work of introducing and studying the peculiarities of miniroses under Botanical Garden (Institute) of the ASM and some localities in the Republic, we believe that they will be important both theoretical and practical perspective that will enhance their cultivation in Moldova: *Cri-Cri*, *Avon*, *Starina*, *Little Opal*, *Cinderella*, *Mandarin*, *Red Sunblaze*, *Baby Bush*, *Baby Masquerade*, *Little Buckaroo*, *Yellow Doll*, *Pandora*, *Cinderella*, *Easter Morning*, etc.

ASPECTE PRIVIND STUDIILE TEHNOLOGIEI DE OBTINERE A SEMINTELOR DE ELITĂ ÎN REPUBLICA MOLDOVA

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Floarea-soarelui (*Helianthus annuus* L.) a câștigat un loc de seamă în agricultura de pe glob, datorită avantajelor sale economice de necontestat: producții mari de ulei pe unitatea de suprafață, ușurința de extragere a acestuiași calitatea înaltă din punct de vedere nutritiv. Cultivarea hibrizilor de floarea-soarelui a deschis un mare viitor pentru cultura acestei specii. Dintre factorii esențiali care contribuie la creșterea producției agricole, cel mai eficace este utilizarea unei semințe de calitate, liberă de boli și de dăunători. Producerea de sămânță este considerate etapa finală a creării de soiuri (hibridi) și etapa inițială a tehnologiei de cultură [Ceapoiu N., 1993].

Savanții din Republica Moldova menționează, că tehnologia de cultivare a semințelor de elită și superelită a formelor parentale, precum și a celor hibride (F_1) de floarea-soarelui în mare parte este similară cu tehnologia intensivă de cultivare a semințelor-marfă. Însă lucrările de semenologie necesitatea și respectarea unor condiții speciale.

Astfel, pentru obținerea formelor parentale și a semințelor F_1 biologic pure, care să respecte structura genetică originală, este necesar izolarea spațială a cîmpului de alte semănături de floarea-soarelui. Fiecare populație trebuie semănată la o distanță de 1500 m față de alt hibrid sau populație de floarea-soarelui și la cel puțin 500 m față de cereale. Dacă nu se respectă aceste distanțe de izolare, vor rezulta biotipuri deosebite de populația originală, iar în decurs de 2-3 ani se va obține o nouă populație care va fi total diferită de cea originală. Cercetările realizate au pus în evidență de asemenea necesitatea creării unor zone specializate în producerea semințelor F_1 [Вронских М.Д., 1979] cu rotația de cultivare a florii-soarelui de 8 – 10 ani [Perju T., 2004] și doar în cazuri excepționale – minimum de 6 ani.

În calitate de culturi premergătoare pot servi cerealiere păioase, porumbul și chiar prășitoarele recoltate timpuriu.

Sub controlul metodic al colaboratorilor Institutului de cercetări pentru culturile de cîmp, gospodăriile semincere din Republica Moldova au produs semințe de clasa super-elită, iar în caz de necesitate – și de elită ale liniilor parentale pentru hibrizii omologați și de perspectivă, iar gospodăriile specializate în producerea semințelor au cultivat pe sectoarele de hibridare semințe de prima generație conform recomandărilor elaborate [Знаменский А.Н., 1968] care au fost în permanență optimizate la condițiile de producere și la tipul hibrizilor implementați.

Astfel, pe parcursul a mai bine de 15 ani, suprafețele sectoarelor de hibridare au variat de între 5672 ha (1996) și 10543 ha (1991), asigurându-se o recoltă de la 2000 (1998) pînă la 9382 (1992) tone de semințe de prima generație, care integral au asigurat înșămînțarea terenurilor agricole de floarea-soarelui-marfă. Pentru asigurarea eficienței și calității lucrărilor de producere semincere toate gospodăriile semincere au dispus de terenuri, tehnică agricolă și utilajele respective. În zona de producere a seminței hibride de prima generație au fost executate lucrările finale de prelucrare a semințelor.

Începînd cu anul 1999 aceste tehnologii au fost parțial preluate de companiile Magroselect, iar mai apoi (2004) și de compania AMG-Agroselect.

Deși au trecut doar circa 200 de ani de la introducerea florii-soarelui în agricultură în general, iar în Republica Moldova doar puțin peste 150 de ani, putem constata cu certitudine că ramura acestei culturi are deja acumulată o tradiție bogată de cultivare la scară industrială intensivă a semințelor marfă, ameliorarea și multiplicarea materialului semincer. Cunoștințele acumulate constituie un fundament trainic și durabil pentru dezvoltarea ramurii în ascensiune, cu un randament economic pe potriva așteptărilor savanților, oamenilor de afaceri din agricultură și industrie, dar și a consumatorilor.

AMELIORAREA PORUMBULUI PRIN SPORIREA CALITĂȚII PROTEINEI ÎN BOB

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Ameliorarea calității bobului la porumb, în primul rând al complexului proteic la etapa actuală se realizează prin utilizarea efectului biochimic al mutației endospermului o2, mo2, recombinanților genice și altor sisteme noi, inductoare ale caracterelor valoroase. Este bine cunoscut că porumbul o2 se deosebește prin conținutul sporit de lizină cu 60-70% și capacitatea nutritivă al boabelor cu 25-30% mai înaltă față de porumbul obișnuit. În același timp porumbul o2, concomitent bogat în lizină și proteină asigură un efect nutritiv superior (la 50%) față și de porumbul o2 dar cu nivelul obișnuit al proteinei în bob.

Datorită valorii nutritive excepționale a bobului o2, suprafețele mondiale însămânțate cu porumb bogat în lizină, triptofană, treonină și alți aminoacizi esențiali a crescut vertiginos și ocupă milioane de hectare în multe țări situate pe continentele American, Asiatic, African fiind folosit, mai cu seamă, în hrana oamenilor adulți și copiilor. Este evident că porumbul bogat în lizină necesită a fi prețuit la justa valoare și în Republica Moldova. Suprafețele semănate cu acest porumb în țară trebuie să fie cel puțin de 30-50 mii ha și amplasate, în primul rând, în gospodăriile crescătoare de porcești și păsări.

În publicația dată prezentăm unele rezultate obținute de noi, privind crearea, evaluarea și utilizarea materialului inițial, a hibridilor de porumb o2 bogați în lizină.

Cercetările științifice efectuate de noi pe o durată îndelungată de timp (anii 1976-2014), privind ameliorarea calității bobului la porumb, sunt bazate pe utilizarea acțiunii biochimice a mutației endospermului opaque-2 (o2) și mo2, unor recombinanți genice de tip o2su2, o2wx1 etc. Prin eforturile depuse s-a creat o colecție valoroasă de linii mutante, mai mult de 550 la număr, dintre care peste 70 sunt transmise și incluse în Colecția Mondială de gene din or. Sanct Petersburg și Centre de Resurse Genetice Vegetale ale unor instituții din țările CSI și Republica Moldova. De asemenea s-au creat, studiat și evidențiat numeroși hibridi o2, dintre care 2 hibridi cu valoarea nutritivă înaltă a bobului sunt omologați în țară și la momentul dat se mențin în Catalogul Soiurilor de plante al Republicii Moldova. Aceștia sunt hibridii de porumb special Chișinău-307 PL cu bobul bogat în proteină și lizină și Chișinău-401 L mo2 cu endospermul modificat bogat în lizină. Potențialul producției boabe obținut a constituit 14,9 t/ha pentru hibridul Chișinău-307 PL și 15,9 t/ha pentru hibridul Chișinău-401 L mo2 și corespunzător producția de lizină de 34 kg și 30kg la 1 ha.. Sunt pregătiți pentru a fi transmiși la testarea de stat 2 hibridi noi de porumb o2 cu producția boabe la nivelul matorului cu bobul obișnuit, rezistenți la condițiile mediului mereu schimbătoare.

Rezultatele cercetărilor noastre au demonstrat că problema productivității porumbului bogat în lizină este posibil de rezolvat doar în baza creării hibridilor noi și a materialului inițial cu capacitate combinativă înaltă. Deoarece metoda analogilor aplicată în acest scop și-a epuizat posibilitățile și trebuie recunoscută ca etapă ce ține de trecut. Prin cercetările efectuate se contribuie permanent la completarea colecției de lucru cu noi linii o2 și mo2 utilizate pe larg în diferite programe de cercetări a calității porumbului atât în țară cât și peste hotarele ei. La rând cu crearea liniilor noi se sintetizează hibridi competitivi după producția boabe, rezistenți la secetă, boli, dăunători și alte caractere valoroase. Anual se testează peste 100 hibridi noi și se selectează cei mai buni, care se studiază în următorii ani în câmpurile corespunzătoare a procesului de ameliorare.

Valoarea nutritivă înaltă a boabelor bogate în proteină și lizină a hibridului Chișinău-307 PL a fost demonstrată în hrana purcelor cu vârsta de înțarcare. Rezultatele experiențelor au demonstrat că sporul în greutate al animalelor hrănite cu boabe bogate în lizină a depășit semnificativ grupa purcelor hrăniți cu boabe de porumb obișnuit în medie cu 49,5%.

Prin urmare, hibridii performanți de porumb cu conținutul înalt de proteină și lizină în bob Chișinău 307 PL și Chișinău 401L omologați în Republica Moldova pentru boabe pot fi cultivați în toate raioanele și tipurile de gospodărie din țară.

După producția boabe și rezistența la condițiile nefavorabile ale mediului nu cedează sau depășesc cei mai valoroși hibridi de porumb obișnuit.

Producția boabe a porumbului special poate fi folosită în scopuri alimentare cât și în calitate de materie primă pentru industria producătoare de furaje combinate.

Valorificarea potențialului nutritiv al porumbului bogat în lizină contribuie la sporirea eficacității întregii economii naționale a Republicii Moldova.

HIBRID NOU DE PORUMB CEROS CU BOBUL ALB CHIȘINĂU 295 WX1;Y1 OMOLOGAT ÎN REPUBLICA MOLDOVA

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Îmbunătățirea complexului glucid al bobului de porumb, mai cu seamă, al calității amidonului prin sporirea conținutului amilopectinei din contul suprimării totale al sintezei amilozei, se utilizează prin acțiunea mutației wx1 (waxy-1). Amidonul bobului wx1 constă practic 100% doar din amilopectină, se deosebește avantajos prin un șir de însușiri fizice, biochimice și tehnologice valoroase în comparație cu amidonul obținut din porumbul tradițional. De aceea porumbul wx1 sau amilopectinic este larg întrebuințat în diverse ramuri ale economiei contemporane așa ca industria alimentară, farmaceutică, hârtiei, producerii zahărului inofensiv pentru sănătatea umană, a cleiului, furajelor combinate și a. La rând cu multe alte criterii înaintate față de calitatea amidonului amilopectinic este și asigurarea transparenței soluțiilor sale cu apa și alți solvenți, o necesitate obligatorie și apreciată înalt în industria producătoare de amidon. La momentul actual, hibridii de porumb wx1 omologați în Republica Moldova (Chișinău 297 wx1, Chișinău 297 wx1, Chișinău 333 wx1, Chișinău 403 wx1, Stalker, omologat în Ucraina), practic aparțin de varietatea cu bobul galben cu un anumit conținut de carotinoizi. Amidonul extras din acest porumb nu asigură transparența necesară soluțiilor sale. Doar amidonul amilopectinic obținut din boabele albe, lipsit de pigmenții coloranți este chimic pur și întrunește caracteristica cerută. Prin aplicarea metodelor de ameliorare și acțiunii mutației recesive y1 (white color), se înlocuiește complet alela sa dominantă Y1 (Yellow) din endospermul galben. În așa mod s-au creat linii noi și hibridi de porumb cu bobul alb, inclusiv de porumb ceros. Colecția de lucru creată de noi, doar a porumbului ceros, alcătuiește peste 175 linii cinsangvinizate, inclusiv cu bobul alb. Majoritatea din ele sunt studiate la capacitatea generală de combinare. Multe din aceste linii urmează a fi studiate și la capacitatea specifică de combinare. Anual în baza lor se sintetizează, testează, studiază și evidențiază după productivitate, rezistență la boli, dăunători și alte caractere valoroase peste 250-300 de hibridi în culturi comparative a câmpurilor de control, preventiv, de concurs. Menționăm că colecția de linii wx1 este sistematizată și inclusă în patru varietăți botanice, dintre care, două varietăți sunt propuse de noi. Formele de porumb wx1 cu bobul galben și rahisul roș aparțin de varietatea *luteorubraceratina* iar cele cu bobul alb și rahisul roș – de varietatea *alborubraceratina*.

De asemenea, în lucrarea dată punem în evidență și alte realizări obținute de noi prin crearea materialului inițial și omologarea pentru prima dată în Republica Moldova a hibridului de porumb ceros cu bobul alb, denumit Chișinău 295 wx1;y1 cu următoarea caracteristică.

Chișinău 295 wx1;y1 – hibrid simplu de porumb ceros (sau amilopectinic) cu bobul alb, convarietatea *ceratina*, varietatea *alborubraceratina*, care este omologat din anul 2015 în toate zonele climatice ale Republicii Moldova. *Autorii: V. Țiganaș, D. Țiganaș.*

În condițiile ecologice ale țării, hibridul se coace timp de 101-106 zile și aderă la grupa semiprecoce de maturitate. Planta cu înălțimea de 200-220 cm, de culoare verde pronunțată. Înălțimea de inserție a știuletelui principal este de 70-75 cm. Numărul de știuleți pe plantă 1,1-1,2. Numărul de frunze pe plantă 15-16. Lungimea frunzei de 70cm și lățimea de 8 cm. Știuletele are forma aproape cilindrică, lungimea 20-23 cm, cu diametrul de 4,2-4,5 cm și rahisul alb. Formează 14-16 rânduri boabe, câte 40-45 boabe în rând. Masa știuletelui constituie 250-260 g. Randamentul boabe înalt, de 84-85%. Bobul wx1;y1, are consistența dură și cornoasă a endospermului, cu aspect ceros, culoarea albă. Densitatea babelor constituie în medie 1,3 g/ml. Masa a 1000 boabe este de 250-280 g. Conținutul în bob de proteine constituie 10,0-11,0% și grăsimi 4,5-5,0%. Conținutul de amidon în bob este de 71-72%. Amilopectină în amidon se conține 98,8 - 99,8% și nu mai mult de 0,2-1,2% amioză.

Potențialul de producție constituie în medie de la 65-75 q/ha până la 134,0 q/ha boabe, iar de amidon amilopectinic-40-90 q/ha. Însușiri specifice. Hibridul este rezistent la secetă, frângere și cădere, boli și dăunători. Semințele hibride F₁ se produc în baza fertilă cu respectarea strictă a izolației spațiale. Se cultivă cu densitatea de 55-60 mii plante la ha. Producția boabe prezintă materia primă valoroasă pentru industria alimentară, producătoare de amidon și melasă, alte produse economic importante. Boabele wx1 utilizate ca hrană furajeră pentru animale, asigură efect nutritiv semnificativ mai înalt în comparație cu boabele obișnuite de porumb. Hibridul este un premergător bun pentru cerealele de toamnă. La moment, se află în testarea de Stat al doilea hibrid de porumb ceros cu bobul alb creat de noi Chișinău 335 wx1;y1.

INGEN 40 – UN SOI NOU DE TRITICALE PENTRU AGRICULTURA MOLDOVEI

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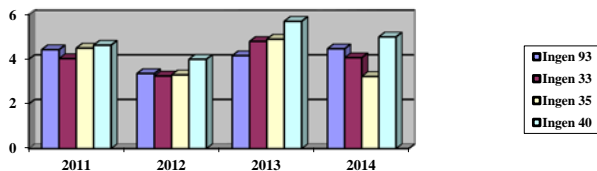
Datorită capacității sale ridicate de producție, însușirilor largi de adaptare, caracteristicilor agronomice și însușirilor de calitate sporite, cultura de triticale în Moldova a căpătat o semnificație economică recunoscută în alimentația umană, industrie și furajarea animalelor. În prezent culturi de triticale î-i revine un rol tot mai important pentru valoarea alimentară, având un potențial agricol considerabil și un conținut ridicat de proteine cu nivel balansat de aminoacizi. Această cultură este rezistentă la ger, secetă, maladii, boabele sunt bogate în proteină, nu este pretențioasă față de sol și are un potențial productiv cu mult mai mare decât formele parentale din care a provenit - grâul și secara

Soiul de triticale de toamnă Ingen 40 a fost obținut prin hibridarea intraspecifică dintre soiul românesc Titan la hibridul intraspecific (Atol x Ingen 93). Producția medie de boabe în ultimii ani variază de la 3,5 t/ha până la 67,0 t/ha, depășind maritorul cu 0,3-0,8 t/ha. Bobul este măscat, masa la 1000 boabe – 47-52 gr, de formă oval. Numărul de boabe în spic ajunge la 65-90, iar în fiecare spiculeț 2-3-4. Lungimea perioadei de vegetație este de 275 - 282 zile și face parte din grupa soiurilor cu precocitate medie. Înălțimea plantei – 95 - 105 cm, cu o înfrățire productivă de 2,8-3,0 frați per plantă. Este rezistent la secetă, ger, pătulire și maladii.

Datorită conținutului balansat de aminoacizi în proteină boabele se pot folosi cu ușurință în producerea pâinii și poate înlocui făina din secară în produsele de panificație. Un rol deosebit se acordă acestui soi cultivarea pe pământuri cu bonitate joasă, demonstrând recolte destul de impunătoare (3,0 – 5,5 t/ha). Cu succes paiea se pot folosi ca nutreț pentru animale și brichete pentru încălzire. Hrănirea animalelor cu concentrate din boabe de triticale se evidențiază un surplus de greutate. Datorită productivității sporite și implementării multilaterale acest soi este mai economic, decât soiurile de grâu comun.

Importanța acestui soi este explicată și prin compoziția chimică a boabelor, care le conferă însușiri deosebite de folosință în alimentație, în furajarea animalelor, precum și în industrie. Calitatea este determinată de complexul însușirilor de morărit și panificație, iar acestea depind în primul rând de conținutul în substanțe proteice, de cantitatea și calitatea glutenului. Din recolta anului 2012 s-a calculat proteina totală și particularitățile tehnologice. Rezultatele obținute confirmă faptul că cultura triticale este intermediară dintre grâu și secară și după acest caracter. Datele experimentale la soiul de triticale Ingen 40 au demonstrat o sticlozitate a bobului de 90-95%, proteină - 14,7%, gluten – 25,5%, volumul pâinii – 530 cm³ și cu porozitatea de 65%. Acești indici sunt aproximativ identici cu cei ai grâului comun Moldova 5.

Soiul Ingen 40 poate fi folosit ca cultură umblătoare. El prezintă interes ca cultură de rezervă, când cultura de grâu de toamnă în condițiile aspre de iarnă pier. De obicei, în condițiile noastre aceste semănături se înșămânțează cu orz și recolta de boabe se poate folosi numai pentru furaj, dar dacă le înșămânțăm cu triticale (Ingen 40) recolta de boabe poate fi folosită cu succes în industria de panificație.



Datorită potențialului mare de productivitate și în anul secetos (2012) cât și rezistenței sporite la factorii condițiilor nefavorabile de cultivare soiul de triticale Ingen 40 a fost omologat pentru recolta anului 2015 pe întreg teritoriul Republicii Moldova pentru boabe.

THE MAIN DIRECTIONS AND ACHIEVEMENTS IN THE BREEDING OF THE LEGUMES FOR GRAINS IN THE PI RIFC "SELECTIA"

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Lately in agriculture of Republic of Moldova was established a trend of increase in areas under leguminous crops. This can be explained by several reasons, but we must mention the main argument – that the cultivation of leguminous plants is profitable primarily because of local varieties of soybeans, peas and beans, adapted to the climatic conditions of the country, created in the PI Research Institute of Field Crops "Selectia" and high biological quality of primary seed produced by this institution.

The pea breeding program, which aim is to create highly productive varieties using mutant gene "afila", that in the homozygous recessive condition causes the transformation of the lamellae in highly developed and branched hitches. It was obtained a serie of new type lines that although a smaller leaf area, provides a more effective penetration of light in th sowing and contributes to higher grain yields compared to other types of plants.

Another important objective is breeding of the resistance of pea beans to shaking. For this purpose are being carried out to include the gene "def" into the plant genome, who controls the character of the resistance to shaking of the beans. As a result of these works in the past 10 years in Laboratory of Breeding and Primary Seed Production of Legumes for Grains of the RIFC "Selectia" was created and approved varieties of peas Gloria, Alisa, Valexa and MZ 7. Production potential of these varieties is of 4500-4800 kg / ha of grain, containing 23-26% crude protein.

The soybean crop breeding out in the high level of production is aims to increasing the levels of the crude protein and vegetabal oil. In the last 10 years have been created and approved the soybean varieties Horboveanca, Indra, Enigma, Deia and Magia , which has a production potential of 4000-4500 kg / ha, a crude protein content of 38-40% and a oil content of 19-21%. Is necessary to mention that variety Deia combines a production potential of about 3200 kg / ha and the growing season of 105 days only, which may be a good precursor for wheat winter and winter barley. From the group of early varieties the highest performance was obtained by the soybean variety Enigma, which in severe drought conditions in 2007, according to the official data of the State Commission for the Testing of Plant Varieties ,exceeded productivity by 70% other varieties of testing. We mention that not only soybeans, but the genetic material of other legumes for grain in relation to environmental tressogenic factors were studied.

The latest achievements in bean breeding may be mentioned varieties Garofița and Nicolina. They are characterized by a potential of 3000-3500 kg / ha grains, containing 24 to 26% of crude protein. In connection with the new requirements of the market the breeding works have started to create new varieties of beans with higher grain as traditional varieties and also suitable for mechanical harvesting. Under this program was created and approved for 2015 the variety Marita, with a potential of 3400 kg / ha, containing 23-25% crude protein and the1000 grain mass 240-270 g, which is 40 -50 g higher compared with currently approved varieties.

For new varieties of legumes for grains after the registration in the Catalogue of Plant Varieties of the Republic of Moldova in the PI RIFC "Selectia" is performed the primary seed production. It is produced annually about 80-100 tons of high quality soybean seeds, about 20-30 tons of peas seeds and about 5-10 tons of bean seeds.

ENVIRONMENT-IMPROVING PHYTOTECNOLOGIES FOR MEGACITIES

A.A. ZHUCHENKO Jr.

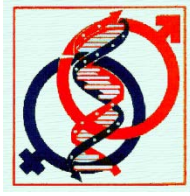
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In megacities (particularly northern ones), fundamentally differing from ordinary cities, people spend most of their time in an indoor environment the conditions of which tend to be detrimental to human health. Therefore, improving life quality lies at the heart of the sustainable development strategy. Clearly, the XXI century calls for a switch from "passive" nature conservation to active development of knowledge-intensive environment-improving phytotechnologies involving mobilization of the world's genetic diversity of plants with environment-improving characteristics.

We have developed environment-improving phytotechnologies that allow harmony between the internal (endogenous) environment of the human organism and its external (exogenous) environment to be achieved taking due account of territorial features of the environment and individual characteristics of a person and their family. Moreover, developed has been a fundamentally new model of the "man-plant" system making it possible, based on prior ecogenetic monitoring, to create a healthy environment in unfavorable local human habitats through employment of environment-improving phytotechnologies calling for:

1. Examining world collections of plant genetic resources with a view to identifying donors of genes responsible for both monogenic and polygenic environment-improving characters.
2. Studying and enhancing environment-improving characteristics of plants (such as antimicrobial, hazardous and noxious substance-absorbing, beneficial substance-excreting, including evaluation of disease-preventive effects of particular concentrations of certain volatile phytoorganic active ingredients, etc.) through employment of advanced plant breeding and genetic techniques, DNA technology, constructing specialized phytocompositions and phytocenoses, as well as significant enhancement of environment-improving features of plants through advanced technologies, and world-standard architectural and design-aesthetic solutions.
3. Conducting ecogenetic monitoring with a view to area zoning according to ecological parameters and identifying individual characteristics of a person, including a person's ecological profile and genetic passport.
4. Developing customized programs for improving the lifestyle and active longevity of a megacity resident.

Therefore, there is a need for identifying, within the system of life-sustaining factors (biological, ecological, social, psychological, ethnic, economic, information, etc.), priorities associated with genetically determined features of man's evolutionary memory of the environment. That is why managing the shaping of a healthy environment for humans spending much of their time in unfavorable conditions is crucial to their active longevity.



ANIMAL GENETICS AND BREEDING

**The Xth International Congress
of Geneticists and Breeders**

THE POSSIBILITY OF CONSERVATION OF GENETIC RESOURCES

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Depletion of genetic resources can cause various negative consequences: firstly, significantly decrease the efficiency of selection; secondly, existing breed will not be able to successfully resist the constantly evolving pathogens; thirdly, will be lost valuable material for studying the evolution of animals and plants. Therefore there is an urgent need to develop methods for long-term preservation of gametes *in vitro*.

The carriers of hereditary information - gametes of animals after receiving may retain functional activity within a few hours. This time is not enough for the effective conduct of large-scale genotypic selection. The latter can be achieved by dilution of the semen. However, in this case, the duration of gametes reaches only a few days.

Inactivations of gametes are possible through the application of chemical compounds which inhibit the metabolism (thiol and phosphate, cyanide). However, this method of preservation causes certain difficulties, as there is a need to find ways reversibility of the inhibition.

Conservation of biological objects can be performed by drying. A change in the intensity of viability of gamete through xeropreservation aims to stop water from cell biological interactions. It is known that during the transition to anabiosis in natural conditions, organisms or their embryos can dry out to air-dry state, when the water content reaches 5-10%. In this case occurs not so deep drying, but it is definitely the offset of the ratio between the condition of the water in favor of her bound form. In such investigations, it is found that the percentage of conceptions of animals is directly dependent on the residual moisture of the seed - the motility of gametes can be recovered only at a humidity of not less than 6%, and only structured water increases the protein stability during freeze-drying, preventing the transformation of protein and oxidation of the hydrophilic sites. Despite significant advances in the field of lyophilization of gametes, speech on the complete removal of the fluid cannot be. The observed effects of freeze drying can be explained by dehydration, in the sense of replacing water with liquid protectors. It should be noted that lyophilization as a method in which cells are first frozen and then the ice sublimates in a vacuum, gives good results in the preservation of viruses, bacteria, fungi and plant seeds. Experiments on the lyophilization of gametes of animals is difficult reproducible. At the lyophilization are possible mutations and chromosome disorder, there is a high possibility of damage to DNA, arising chromosome breakage and mutations are not possible to repair. When developing a method for long-term storage of gametes in the dried state should consider that a form of their adaptation to a gradual increase of osmotic pressure in the environment leads to osmotic suspended animation. Further development and improvement of the method of xero conservation is an urgent task, since this method eliminates the use of refrigerants and expensive equipment for long-term storage of biological objects.

Despite this method of preservation of genetic resources with using of temperature liquefied gas finds wide practical application. This method eliminates any metabolic processes. Under these conditions of the influence on frozen objects genotoxic agents can be eliminated. By calculation, it is established that at -196°C the factor of limiting the duration of preservation of genomes becomes the natural radiation background of the earth. But even in these circumstances, the storage of reproductive cells is permissible within virtually unlimited time. This fact is confirmed by experimental studies by method of scanning densitometry in which determine that the distribution of content of DNA in gametes of native bull semen, and after their storage at a temperature of -196°C for 4 days, 1, 5, 8 and 11 years is of similar nature. The values of the statistical parameters characterizing these distributions also indicate that regardless of the duration of storage of gametes bulls in the frozen state the amount of DNA does not change in comparison with its content in native gametes. The results of the research of content of histones, performs an important role in the specific arrangement and condensation of the DNA of gametes shows the stability of the content of nuclear protein in most cells at different periods of storage. These data confirm the computational studies according to which the DNA and histones do not undergo significant changes in the cryopreservation during prolonged storage of gametes of bull breeders.

Thus, long-term storage of genetic resources at the temperature of liquefied gases has no alternative.

HYPOTHERMAL CONSERVATION OF GENETIC RESOURCES OF BOARS UNDER THE PROTECTION OF L-CARNITINE

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The possibility of dynamic growth in livestock production is determined, among other factors, the intensification of reproduction of the herd with the use of artificial insemination. Zoo Veterinarian advantages of this biotechnological methods of reproduction in comparison with natural reproduction determines its leading role in the technology of production of pigs in farms with different production-economic structure. However, the potential of artificial insemination of pigs is not realized in full.

Based on the above, the purpose of the research was to study the qualitative indicators of reproductive cells of boar stored after dilution with synthetic medium containing the biologically active substance L-carnitine.

As experimental material we used semen of boar breeders of Landrace which were maintained in the conditions of the tribal enterprise "MOLDSUINHIBRID" in accordance with veterinary requirements. For sperm dilution was used a synthetic medium which contained glucose, ethylenediaminetetraacetic acid (EDTA) and sodium citrate. In our experiments we used pharmacological 2% L-carnitine and ferric sulfate. Dilution of sperm was performed 1:1. Synthetic medium for dilution and storage of sperm of farm animals at 16 - 18°C, as a rule, are not complex. They are designed to maintain osmotic pressure, pH and the viability of spermatozoa. However, the problem of improving of the functional status and increasing life expectancy of the cells continues to be relevant for practitioners involved in the reproduction of pigs. Therefore, we consider it appropriate to introduce into the composition of mediums the components that contribute to homeostasis of metabolic processes. For this purpose, we have studied the protective properties of L-carnitine in the composition of the base medium for dilution and storage of boar semen. Experimental evidence suggests that L-carnitine has a dose-dependent effect. Its use in the composition of the medium for dilution of boar semen at a concentration of 0,04-0,08 mg/ml has a positive effect on the functional indices of reproductive cells. In the best experimental variants the motility after dilution and absolute index of survival after 12 hours of storage of the sperm at 16-18°C respectively amounted to $7,6 \pm 0,27$ and $91,2 \pm 3,29$, which indicates an increase of the studied parameters by 22,6 and 22,1% compared to the control variant. L-carnitine (3-Hydroxy-4-(trimethylazaniumyl)butanoate) refers to indispensable substance because it perform a basic role in the transport of fatty acids across the mitochondrial membrane. However, L-carnitine is synthesized in the animal body in the liver and kidneys where by blood flow is transported to other tissues and organs. Great interest in L-carnitine is due to its role in metabolic processes. Among them, it should be noted: transport of long-chain fatty acids into the mitochondrial matrix, where they are included in the process of formation of acetyl-CoA; stabilization of the content of the CoA and the removal of short-chain fatty acids from mitochondria; regulation of the content of the CoASH, which is required for detoxification of metabolic products; maintaining of the optimal ratio of acetyl-CoA / CoASH for stimulate the anabolic processes; maintaining of cell activity through the involvement of L-carnitine in energy metabolism with the participation of phospholipids. It is assumed that the enhancement of mitochondrial energy metabolism may indirectly prevent the formation of free radicals. In this regard, it is recommended to be used as the most widely applicable antioxidant for the regulation of metabolic processes of spermatozoa. From the analysis of the submitted information suggests that the protective effect of L-carnitine in the composition of the medium for dilution and hypothermal storage of boar semen, mainly can be attributed to the regulation of energy metabolism and the detoxification of the products of this process.

Thus, in the maintenance of the functional status of boar semen after it was received a significant role acquires the regulation of metabolic processes at the different technological stages; improving the quality of boar semen stored at hypothermal conditions can be realized by including in the composition of medium for hypothermal storage of boar semen is preferable to use a dense fraction, the volume of which will be increased after the first and repeated dilution.

**ALBINISM - IMPORTANT FACTOR OF GRAY RAT METAMORPHOSIS IN A
LABORATORY ANIMAL**

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White rats represent the result of two substantial experiences. The first one lasted up to several million years and was carried out by nature through natural selection which accomplished the transformation of rats from natural ecosystems in commensal animals, then into synanthropic and synurban animals with specific accommodation qualities necessary for existence in anthropogenic zoocenosis. The second experience was realized by humans in a very short term and was based on the artificial selection resulting in a new type of domestic animal – laboratory animal.

The existence of albino rats in genetically varied heterogeneous populations had a particular role in metamorphosis of wild rats into domestic animals. Hence the first source of transformation was mutation, the following evolution happened under the artificial selection during the domestication of albino rats genetically and ecologically isolated from wild ancestors.

Artificial selection of albino rats has considerably lowered their aggressive behavior specific to emotional stress caused by deprivation of liberty and contact of wild animals with humans. Genetic reorganization was accompanied by the following morphophysiological modifications: 1) diminution of brain mass; 2) diminution of freedom reflex; 3) reduction of sensory perception, 4) minimization of circadian rhythm influence on reproduction; 5) weakening of progeny defence reflex. A major role in domestication had handling, especially at the early postnatal age. This operation induced not only multiple changes in the neurotransmitter systems of the brain, but also in neuroendocrine systems and has significantly reduced the aversive emotional reactions caused by the contact with humans.

In our opinion, one of the prerequisites for successful domestication of rats consists in peculiar reaction to stress of albino rats. In the similar situations of conflict animals have demonstrated different reactions of neuroendocrine systems, in particular this phenomenon was observed in albino animals. Albinism is a typical mutation with pleiotropic effect, which determines several modifications: change of integument and iris colour, reduced locomotory activity, eyesight deterioration, decreased reactivity to the action of stress factors, specific for domestication processes. The above mentioned peculiarities contribute to the diminution of excessive stress typical for initial stages of domestication. The impact of stress reduction can be demonstrated through the maintenance of reproduction during the artificial selection of albino rats.

Taking into consideration the very low frequency of occurrence of albinos in nature, it should be mentioned that their reproduction in very small artificial populations became possible only in case when stress of animals, who were in contact with humans in a limited space did not negatively affect the function of reproductive system and the process of reproduction. Thus, the appearance of white rat (*Rattus norvegicus* var. *alba*), which is used in a number of physiological experiments for more than 150 years, was determined by the change of reactivity of neurohormonal systems responsible for the stress reaction, providing the organism accommodation for existence in new ecological conditions and contributing to the ontogenetic development and survival of the species.

BREEDING AND REPRODUCTION OF LOCAL CARPATHIAN BEES

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Beekeeping success largely depends on the organization and the development of breeding base, improving of queen bees growing technology and exploitation of bee families. Valuable breeding material can be quickly reproduced by queen rearing. The development and productivity of bee families very much depend on the queens' value, their quality and genetic potential.

In breeding work and assessing the quality of individuals the morph-metric sizes indices are taken into account. Appreciation of morph-metric indexes or exterior allows evaluation of genetic peculiarities of the group of individuals, identify the population change at various levels, restoring of population microfilogenesis, which ultimately facilitates keeping control of phenotypic and genotypic structures of honeybee population.

The research aim is to identify, strengthen and reproduction of the most valuable genotypes suited to the local climate and natural honey harvest.

The result of research of morph- metric indices of working bees from apiary "Albinarie" showed that bees of the maternal families had an average of tube length- 6,37 mm with variation between 6,31 to 6,44 mm. The size between projections of sternum-3 ranged from 4,29 to 4,57 mm, the length of tergite-3 - 1,93-2,07 mm, the length of sternum-3 - from 2,51 to 2,65 mm, the width - 3,45 to 3,60 mm, the length of beeswax mirrors - 2,09-2,16 mm, width - 1,39 to 1,55 mm, the length of large right wing - from 8,25 to 8,54 mm and width - 2,97 to 3,33 mm, cubit index - 2,19-2,31 and discoid positive displacement - from 84,6 to 100%, the average being - 92,8% and 7,2% neutral. The body mass of working bees in March averaged 110,35 mg and the mass without digestive system - 72,12 mg.

As a result of the study of morph - productive indices were selected the most valuable families and were created a group of breeding queen bees for reproduction of maternal and paternal families. Results of morph-metric study of working bees from apiary "Dancenii" showed that working bees from maternal families had on average of tube length - 6,24 mm and variation between 6,32 and 6,59 mm, length of tergite-3 - 1,98 mm, length of sternum - 3-2,56 mm and width - 3,65 mm, length of beeswax mirrors - 2,20 mm and width - 1,46 mm, length of large right wing - 8,34 mm and width - 2,98 mm, discoid positive displacement - 82,2% and neutral - 17,8%. To grow up the drones there were selected paternal families. The body mass of working bees averaged 112,23 mg and their mass without digestive system - 79,44 mg.

At apiary from vil. Hirtopul Mic, were selected the most valuable productive, resistance to winter and suitable to pure breed *Carpathian* standard. It was found that worker bees from maternal families have an average of tube length of 6,44 mm with variation between 6,32 and 6,59 mm, size between the projections of tergite-3 - 4,59 mm (from 4,55 to 4,64 mm), the length of tergite-3-2,10 mm (from 2,09 to 2,13), the length of sternum -3-3,74 mm (from 3,69 to 3,82 mm) and width - 2,66 mm (2,63-2,70mm). The length of beeswax mirror - 2,22 mm (2,18-2,31 mm) and width - 1,51 mm (1,45 to 1,59 mm), large right wing length - 8 62 mm (8,52 to 8,73 mm) and width - 3,02 mm (3,0 to 3,04 mm), cubit index - 2,49 (2,2 to 2,9), discoid positive displacement - 98,3% and neutral - 1,7%, the length of tarsus - 1,99 mm (1,96 to 2,03 mm) and width of 1,11 mm (1,07 to 1,15 mm).

Working bees from paternal families had an average of tube length of 6.54 mm, the length of large right wing - 8,54 mm and width - 2,99 mm.

Based on research conducted on of morph - productive indices there were selected and created breeding groups of queen bees for reproduction from maternal and paternal families.

Production and breeding qualities of bee families causes hereditary peculiarities and conditions of breeding of queen bees. Using breeding queen is the most effective method of economically, to increase productivity of bee families. In this case, the costs are determined solely from cost of queen procured for mass reproduction of queen-daughters.

The study found that the most valuable queens were obtained in June with an average body mass of 252,06 mg, and during the active autumn season their mass decreased, in early September constituted - 213,7 mg.

Therefore, the development and productivity of bee families, largely depends on the biological potential and particularities of queens growth technology, providing valuable apiaries with young queens, which is of great scientific and practical interest in breeding selection and queens breeding.

INFLUENCE OF FEED ADDITIVE VITACORM AD-1 TO STIMULATE RESISTANCE TO WINTERING OF BEE FAMILIES

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Economic and social importance of beekeeping in Moldovan agriculture is estimated that bees provide high biological value products, such as honey, wax, pollen, bee bread, propolis, royal jelly. Simultaneously, the bees contribute to the pollination of crops significantly increases the quantity and quality of seeds and fruits.

The success depends on the quality of bees wintering in the family. Best overwinter and live longer life young bees bred in the second half of August - early September, not involved in harvesting and processing nectar, feeding larvae and remain physiologically younger, which is important for increasing the spring brood.

Therefore, during this period must follow the hives are glutted with food protein (pollen or bee bread) and bees reared to form reserves of protein, nutrients in their body, which extends their longevity, especially in the winter.

Preparations to winter rest begin in August. With autumn control is necessary to complete hives with food, food supply is properly arranged, new combs without brood are removed, also are removed black old combs and those with drone cells.

The bees are fed with sugar syrup in autumn, when the amount of food reserves is insufficient in the family, at the change of honeydew or honey that crystallize quickly (as sunflower) and for the prophylaxis.

Late feeding of autumn generation with sugar syrup requires bees to eat during the processing of the syrup of a number of essential nutrients stored in the body for winter. The pharyngeal glands attest degeneration, reduce body fat and longevity of bees.

In the spring, sometimes *Nosema* appears and as a result, we have a number of essential losses, families of dead bees, weak with few bees of low resistance.

Proceeding from the above, the purpose of the investigations is to stimulate bees during the winter resistance against *Nosema* disease, survival after winter rest and increase the productivity of bee families.

To carry out research during the preparation and completion of food reserves for the winter rest in August were formed four groups of hives. Families of bees in group I (control) received pure sugar syrup; in the experimental group II - sugar syrup with 1.5 ml / l Vitacorm AD-1; in group III - 3.0 ml / l; in group V - 4.5 ml / l.

Study of wintering resistance of bee families from the apiary "Albinarie" demonstrated that the autumn revision, 7 August 2013, had power on average from 7.33 to 7.67 spaces populated with bees between the honeycombs, 4.33-5.0 combs with capped brood from 6.67 to 7.33 kg of honey.

In autumn overhaul after replenishing food, on 21 October 2013 bee families had the power an average from 5.67 to 6.67 spaces between combs with bees and honey reserves - from 12.63 to 14.0 kg. At the overhaul in spring (22 March 2014) was found that bee families were out of winter, with an average of power from 5.33 to 5.67 spaces between combs populated with bees, and 9.33 to 11.07 kg of honey.

Better have borne the winter, bee families that were fed to replenishment of reserve with sugar syrup and feed additive Vitacorm AD-1 as 3.0 ml / l, the resistance was 94.43% or with 8.73 % better than in the control group. Increasing the dose of a feed additive per liter of syrup has not led to increased resistance to wintering of bee families.

It is therefore reasonable stimulation of bee families in the autumn to replenishment of reserves food with the use of feed Vitacorm AD-1 3 ml / l, from the third decade of August to mid-September, which ensures better resistance compared with control group.

APPLICATION OF ENVIRONMENTAL DNA (EDNA) METHODS FOR ASSESSING BIODIVERSITY AND BIOMONITORING ENDANGERED SPECIES: A CASE STUDY OF JEFFERSON SALAMANDER (*AMBYSTOMA JEFFERSONIANUM*) IN SOUTHERN ONTARIO, CANADA

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Use of environmental DNA (eDNA) has the potential to be revolutionary in improving our knowledge of biodiversity and in reducing threats to it. Environmental DNA includes DNA that can be obtained from an environmental sample, such as soil or water. This DNA can be of various origins, such as animals, plants, fungi, or bacteria. In the case of animals, it can come from various bodily fluids or skin cells that have sloughed off an individual.

Much of the impetus for the development of eDNA methods has been the need to detect and monitor endangered organisms. Given the importance of protecting endangered species or populations, it is crucial to expedite to standardization of eDNA monitoring protocols. One of the main advantages of targeting eDNA is that its collection is a form of non-destructive sampling - this is especially important in situations where endangered or "at risk" species are being monitored.

We conducted a case study on using eDNA to improve monitoring of salamanders in the genus *Ambystoma*, including the endangered Jefferson Salamander (*Ambystoma jeffersonianum*), in Ontario, Canada. Current monitoring methods for salamanders rely on observation of individuals or egg masses, and identification to species can be difficult. This problem is compounded by the complex genetic system of *Ambystoma*, including the presence of unisexual individuals.

For our research, we collected water samples from various ponds in southern Ontario where Jefferson salamanders had been previously observed. Using tissue samples from specimens of Jefferson Salamander, two other species of *Ambystoma*, and multiple polyploids that are commonly found in the geographic range of Jefferson Salamander, we extracted DNA, which was then barcode-sequenced using standard sequencing primers. The DNA sequences that were generated through barcoding were compiled into a database that served as an *Ambystoma* sequence library. We designed three primer sets based on the sequences in this library. Environmental DNA extracted from the pond water samples was sequenced on a high-throughput sequencing platform (Illumina MiSeq) using these primer sets. We then compared the eDNA sequences generated to a sequence library that we produced by sequencing DNA from tissue of identified *Ambystoma* and unisexual individuals. Our results included positive matches for the DNA sequences from the pond water to those DNA sequences in the library, thereby confirming the successful detection of targeted salamanders from water samples via eDNA. Based on these results, our method will allow for more accurate detection, and for a longer time window than is currently possible.

**HOLOCENTRIC CHROMOSOMES AND THEIR APPLICATION FOR THE SYSTEMATICS
AND PHYLOGENY OF HETEROPTERA.**

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Holocentric chromosomes are characteristic of different invertebrate groups, such as all Nematoda; insects: Dermaptera (earwigs), Heteroptera (stink bugs), homopterans Sternorrhyncha (aphids, mealybugs, white flies), and Auchenorrhyncha (cicads, spittle bugs), Lepidoptera (butterflies and moths), Odonata (dragonflies and damselflies), Phthiraptera (Mallophaga and Anoplura) (lice), Psocoptera (book lice), Trichoptera (cloth moths) and Zoraptera; few species of Onychophora (velvet worms); Arachnida: scorpions of the family *Buthidae*, spider families *Dysderidae* and *Segestridae*, mites – all Astigmata and some species of Prostigmata; Chilopoda (centipedes) – few species of *Scutigridae* and *Henicopidae*. They also occur in some plants and protists.

Distinctive features of holocentric chromosomes are: absence of a primary constriction in mitotic metaphase; migration of sister chromatids parallel to the equatorial plane in mitotic anaphase; persistence of chromosome fragments after irradiation; presence of kinetochore plates covering a large surface of each chromatid in mitosis. An important characteristic of this chromosomal structure is that it may allow for more rapid karyotype evolution through both fragmentation and fusion of the chromosomes.

We studied the karyotypes of 9 species from 8 genera of the *Pentatomidae* family (Hemiptera: Heteroptera), which is a highly derived family from an evolutionary point of view. As the result of this study, we propose a dendrogram reflecting phylogenetic relationships among the investigated genera. We have confirmed the hypothesis that macroevolutionary changes of the karyotypes within the *Pentatomidae* family occurred in the direction of chromosome number increase (*Raphigaster*, *Pentatoma*), as well as chromosome number decrease (*Aelia*, *Eurydema*, *Carpocoris*, *Graphosoma* and *Dolycoris*), compared to the ancestral modal chromosome number $2n=16$ (*Eysarcoris*).

In another study, we used karyological analysis to investigate the chromosome number, mechanism of sex determination, and relative size of X chromosome for males of the water strider *Aquarius remigis* (Heteroptera: *Gerridae*). For this analysis, we used adult males of *A. remigis* from laboratory culture. Their testes were dissected and immediately fixed, after which they were stained with aceto-orcein. Squashed preparations were prepared in order to obtain metaphase plates, which were studied under the microscope and photographed. The results obtained regarding chromosome number and sex determination mechanism for *A. remigis* suggest that phylogenetically, it should be placed as a sister group to *Gerris lateralis*.

**GENETIC CHARACTERISTIC HEIFERS MOLDOVAN TYPE OF BLACK-MOTLEY
CATTLE - DESCENDANTS VARIOUS OF BULLS**

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In the article are the results of research and monitoring of changes in the gene pool of the Moldovan type black-Motley cattle in the herd STE "Maximovca." Identified differences in antigenic spectrum of blood groups in the offspring of 11 bulls used in the herd during the period 2003 to 2013 years.

Among the identified 6 antigens (B_2 , G_2 , O_2 , Y_2 , E'_2 , Q') the descendants of all the bulls there is a high frequency of occurrence of Antigen Y_2 , which varied from 0.3250 (Dichii 788) to 1.0 (Abkhazian 835, Meteor 376).

On AEB-locus Antigen B" identified only by descendants of bulls Captain 2354 and Svet 732, as evidenced by the earlier assessment of the genetic structure of the used lines of bulls in the herd STE "Maximovca" and is a characteristic of the descendants of the bulls line Vis Back Ajdiala and Pavni Farm Arlinda Chief.

A study of relationship between the descendants of various bulls set the closest were the descendants of the bulls Academician 767 and Signor 7415, the genetic distance is 0,1291, and most distant descendants of the bulls Abkhazian 835 and Dichii 788.

Results of study of relationship between the descendants of various bulls revealed the closest were the descendants of the bulls Academician 767 and Signor 7415, the genetic distance is 0,1291. The most distant - descendants of the bulls Abkhazian 835 and Dichii 788, the genetic distance is 0,3095.

Key-words: cattle, the descendants of, frequency antigens, the antigenic spectrum, genetic distance, dendrogram.

MANIFESTATIONS OF ENDOTOXICOSIS MARKER INDICES AND OF THE HISTIDINE-DIPEPTIDES IN QUAILS TREATED WITH AN AUTOCHTHONUS REMEDY

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The economic and social importance of aviculture in the Republic of Moldova's economy is appreciated by high biological volume value products such as: meat and eggs. At the same time, aviculture contributes to improving and enhancing the quality of human life due to the price and availability of poultry products. Quail rearing is justified by the fact that quail meat and eggs are nowadays widely requested by consumers for their curative and dietary qualities (Alexadru A. 2001). Despite the fact that quails, in terms of the volume of production achieved, are incomparable with hens, they are very significant due to the avalanche of investigations in decoding the genome, thus enriching and even confirming the obtained results on hens (F. Minvielle, 2009). One important aspect is that quails are sensitive enough to the stressogenic factors that may negatively affect egg production and meat quality. Given the biological specificity of this species, but for other reasons as well, more often are being used biologically active products with adaptive, anti-stress and high growth stimulators properties. In aviculture there are used with no doubt growth stimulators, priority being given to the ones of natural origin, harmless and of plant origin. Part of the ecological and biologically active remedies category, obtained through modern technologies from cyanobacteria *Spirulina platensis* is also the BioR product, nationally and internationally recognized (Macari V. et al, 2014).

The possible impact of this remedy on quails' marker indices of endotoxiosis and histidine-dipeptides has not been elucidated yet. In order to elucidate the influence of the BioR remedy a study was conducted on five lots, 40 quails each, in a poultry factory, under physiological conditions. The tested remedy was administered intramuscularly, twice to 4 experimental lots at the beginning of the study, in doses of 0,25-1,5 ml/head, and the second time, with an interval of 7-10 days, in the same doses, to the same experimental lots, while birds in the control group received 0,5 ml of 0,9% NaCl according to the same administration scheme. During the study the quails were monitored to determine body temperature and the number of respiratory movements per minute. Blood samples were taken for laboratory testing, at the beginning of the study, from 5 birds, and then twice during the research, from 5 quails each lot.

Following a thorough study of quails, over a period of about 40 days, has been established that the tested product doesn't cause any side effects or health deviations in birds, and the bird treated with BioR were quieter. Further it was established that the quails that benefited from the tested product had lower body temperature, recording lower values which at the end of the study were by 0,22-0,44°C compared to the control group, data which show the adaptive and anti-stress actions of the tested remedy. The biochemical investigations have shown that under physiological conditions the BioR remedy truthfully reduces the necrotic substances level, at the first sampling by 4,7-20,7%, while at the second sampling by 11,9-24,1% compared to the control group values. Similar results have been obtained while dosing the concentration of average molecular weight substances, which at the end of the study, in the experimental groups, were lower by 2,3-15,6% compared to the control group. At the end of the study was evident the positive impact of the tested product, reflected in lower values of the investigated parameter by 7,9-28,3% compared with the control lot, which may be a consequence of the decrease of the catabolic processes. In addition the tested remedy on adult reconditioned quails, clinically healthy induces a truthful increase of histidine – dipeptides – Carnosine in blood serum, determining the activation of the anabolic processes.

In conclusion we can say that this remedy manifests positive effect on birds, induces the decrease of the endotoxiosis marker parameters: average molecular weight substances, necrotic substances, and simultaneously the increase of histidine-dipeptides – serum Carnosine.

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THE STUDY OF GENETIC STRUCTURE OF HOLSTEIN COWS POPULATION USING LACTOPOLYMORPHICS LOCI

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In recent years in Moldova taking into account local climatic conditions is working on creation of new types of black and white breed by crossing domestic cattle with the best representatives of specialized dairy breeds, mainly with Holstein, with the aim of increasing the potential for milk production and technological qualities of animals. Successful implementation of this work is impossible without the use of modern methods of genetic analysis. Of particular interest in this area is the study of blood and milk polymorphic protein systems for which allelic variants are inherited considerably simpler than their quantitative traits.

Identification of indicators of biochemical polymorphism of blood and milk proteins for Holstein and Black Pied breeds in the climatic conditions of Moldova is helping to improve methods of selection for animals of different genotypes identifying their productive qualities.

The samples were taken for research on the breeding farm of "Doksankom" CW Gagauzia, Chadyr-Lunga district, village Thomas in a quantity of 180 animals. Experiments were conducted in The Scientific Laboratory for Determining the Quality of Animal Origin Products at The SAUM, The Department of Biotechnology in Animal Science.

The of this work was to determine the polymorphism of milk protein loci in α S1Cn, β Cn, κ Cn, β Lg.

At a population of "Doksankom" HO in locus α S1Cn were identified three alleles A, B, and C. The highest frequency of allele is B = 0,911; followed by allele C with a frequency of 0,052 and allele A = 0,041. The presence of the above alleles indicates a biodiversity study population. The greatest number of individuals had a genotype BB – 81,1% (146 animals), followed after individuals with genotype AC – 10,56% (19 animals) and AB – 8,3% (15 animals). The presence of the above alleles indicates the biodiversity of the study population. The greatest number of individuals had a genotype BB – 81,1% (146 animals), followed after individuals with genotype BC – 10,56% (19 goals) and AB – 8,3% (15 animals). In locus β Cn we observe the presence of two alleles A and B. We have observed frequency of allele A = 0,891 and frequency of allele B = 0,102. The greatest number of individuals have the genotype AA – 81,6% or (147 animals), followed by individuals with genotype BB – 2,7% or (5 animals) and AB – 15,5% or (28 animals). In locus κ Cn were Identified 2 alleles A and B. The frequency of allele A is 0.819 and the frequency of allele B = 0,180. Presence of A and B alleles in the locus κ Cn makes it possible to form a three genotypes AA, BB, and AB. The greatest percentage – 74,4% or (134 animals) is for genotype AA, individuals with genotype BB and AB was much smaller: AB - 15% or (27 animals) and BB - or 10,5% (19 animals).

In locus β Lg, as well as in loci κ Cn and β Cn are observed the presence of A and B alleles with frequencies A = 0,769; B = 0,230. The greatest number of individuals with genotype AA has 67,2% or (121 animals) followed by individuals with genotype AB and BB: AB = 19,4% or (35 animals) and BB = 13,3% or (24 heads).

Population in all studied loci was in genetic equilibrium according to the law of Hardy- Weinberg.

As a result of these experiments it can be concluded: in studied population at all loci is present clearly marked polymorphism. It has a positive effect on the degree of variability and consequently on the level of productivity.

INFLUENCE OF MILK PROTEIN POLYMORPHISMS ON SOME INDICATORS OF GOAT'S MILK PRODUCTION

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At the present time in the world is very relevant topic related to the increasing production of livestock products. In countries where livestock is the development of the industry the marker selection is very promising in the breeding of farm animals. In particular if it is done with the use of molecular genetic techniques for identifying the loci of polymorphic signs. This allows determining the relationship of polymorphic signs with different productive qualities of animals of their vitality and others.

The experiments were conducted at Scientific Laboratory for Determining the Quality of Animal Origin Products at SAUM, Department of Biotechnology in Animal Science. For experiments were taken 30 Saanen goats exported from Austria in the summer of 2013 which are grown in the village Rezeni, District Ialoveni. We have attempted to determine the effect of polymorphic systems of milk proteins on the productivity of goats, namely on fat content and on protein content in the milk. From the experimental data it can be seen that the locus $\alpha_S Cn$ with 93,3% (or 28 animals) had genotype BB, and only 6,7% (or 2 animals) were heterozygous AB. This can be explained by the fact that the study population saanen goats were brought to Moldova from Austria, where breeders give their preference to allele B which is associated with high productivity. In βCn locus in the studied breed goats were revealed two alleles (A, B). The presence of these alleles has led to the formation of three genotypes: BB genotype occurs at 24 goals which is 80% of the herd; Heterozygous genotype AB is showed at 5 goals that are 16.6% of the herd; Homozygous for the A allele with the AA genotype is found out only at one goal out of the flock which is 0.03% of the herd. In βLg locus we found out two alleles (A, B), the frequency of which was: βLg^B - 0,8800; βLg^A - 0,1200. The presence of these alleles has not led to the formation of the expected three genotypes but only two. Homozygote for allele A is absent. It can be noted that 83,3% (or 25 animals) have genotype BB. And 16,7% (or 5 animals) were heterozygous and had genotype AB. Homozygous for the A allele are absent. As a result of our study it was found that individuals with genotype βCn^{AB} have milk fat content 4.11%, this is 0.05% higher than the individuals with the homozygous genotype for allele B locus βCn , i.e. βCn^{BB} having a fat content of 4.06%. As for milk protein content locus βCn has the opposite phenomenon observed here. If the fat content was higher for goats with genotypes βCn^{AB} then milk protein content is higher for goats with genotypes βCn^{BB} . It is 3,74%, which is 0,28% higher than that for individuals with genotypes βCn^{AB} . Hence the conclusion that allele B correlates with the percentage of protein in milk. The highest milk fat content is observed for goat's genotype kCn^{BB} with 0,21% higher than for the individuals with the genotype kCn^{AB} and with 0,25% higher than for the individuals with the genotype kCn^{AA} . From the experimental data it can be seen that the alleles B at a locus kCn correlate with the percentage of milk fat. With regard to locus βLg , from the experimental data can be seen that the percentage of fat in milk for individuals with genotype βLg^{AB} and for individuals homozygous for the allele B βLg^{BB} is the same equal to 4,07%. The percentage of protein for individuals with both the above genotypes did not differ from each other and is equal to 3,45%.

The principal results of our experiments lead to the following conclusions: In studying the influence of different loci on the fat and protein content of goat's milk it was found that the allele B in locus kCn affect the fat content. These individuals increase with 0,27% the fat content in milk compared with peers having homozygous genotype AA. Polymorphic system does not seem to affect much on goat's milk protein content.

IMPROVEMENT OF TSIGAIE SHEEP BREED OF MOLDOVAN TYPE

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This paper deals with appreciation of the herd of sheep of Tsigai race of Moldovan type which specializes in wool-meat-and-milk direction of production, at birth, at 3-3,5 months of age, 12-14 months of age. There is presented productivity of animals of different age groups selected in the selection nucleus in relation to the minimum requirements of the standard.

According to the animals' body mass and wool production, the animals of selection nucleus are more superior to the official percentage requirements for breeding Rams by 0,7% to the body mass, and by 15,8% to production of wool; while the sheep are superior according to the body weight by 15.4% and according to the wool production - by 14,8%; breeding lambs - by 13,9% and 2,5% according to the body mass and wool production respectively; breeding ewes - by 5,6% according to body mass and by 8,7% according to the production of wool.



PLANT AND ANIMAL BIOTECHNOLOGY

**The Xth International Congress
of Geneticists and Breeders**

**MACRO AND MICROBIOTA AS LIMITING FACTORS IN PROPAGATION OF
OROBANCHE CUMANA WALLR.**

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Orobanchaceae family includes more than a hundred species of parasites plants predominantly (about 90%), originating from temperate areas of Europe. Not looking at factors associated with the globalization process, very close trading links, exchange of seed material, biological, etc. between the continents of Europe and America this plant is lacking sunflower plantations in the New World countries. Orobanche fall within 90% of plants that fail to invade a new ecosystem, due to limitations imposed by biotic and abiotic components of habitat. Temperature is an important abiotic factor that limits the development of this species. Fluctuations in the temperature of the soil during a dry and hot hormones are necessary for the activation of germination. Only after this process of preconditioning, root exudates removed from the host plant parasite can induce seed germination. Some environmental factors such as temperature and soil might explain the success of an invasion of *O.cumana*. Soil biota favored by existing plant community may limit the degree of attack. Another factor may be the texture of the soil can also affect the structure of the vegetation. Orobanche generally occurs on sandy soils where nitrogen sources are poorly available. It was also found that soil salinity may limit the scope of the attack *O. cernua* in tomato. To elucidate the mechanisms related to plant parasitic invasions, it is necessary to investigate several biogeographical factors.

A comparative study between infected and uninfected habitats would provide important insights for management to limit the impact of the invasion of broomrape in R.Modova conditions. The objective of this study was to highlight the macro factors and microbiota and some soil factors of farmland flooded and noninvadate of broomrape in different biogeographical zones of Moldova.

Exploratory study was conducted at two levels distinguished as macrobiotic factors (geographic location, latitude, longitude, height from sea level, climate - min and max air temperature, rainfall) and microbiota (physico-chemical peculiarities of soil).

Environmental parameters of soils infested and non-infested were taken from hydrometeorological stations in the region sunflower plantations (online access).

Soil samples (30 cm) of sites infested and non-infested were used in the experiment. As soil physical and chemical characters were determined: pH; calcar - CaCO₃; Total N, nitrate, ammonia; P-total; K-total, humus and soil texture. These parameters were determined by standard methods.

The research results revealed a correlation between physical and chemical soil component with the degree of infection and distribution of *O.cumana*.

BIOLOGY, MANAGEMENT AND CONTROL MEASURES OF *OROBANCHE CUMANA* IN R.MOLDOVA

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Orobanche or broomrape (*Orobanche cumana* Wallr.) locally known as lupoaie, or Țfoșniță is a phanerogamic, obligate, troublesome root parasite that lack chlorophyll and obtain carbon, nutrients, and water through haustoria which connect the parasites with the host vascular system. The attached parasite functions as a strong metabolic sink, often named “super-sink”, strongly competing with the host plant for water, mineral nutrition and assimilate absorption and translocation. Depending upon the extent of infestation, environmental factors, soil fertility, and the crops’ response damage from Orobanche can range from zero to complete crop failure.

Broomrape is a pathogen of sunflower considered of medium importance regarding damage caused culture conditions of our country, but in recent years has expanded the area of the attack and the impact on production and quality. Thus, *O. cumana*, as required parasite has developed and will develop new physiological races, all pathogenic on especially for sunflower.

Because nutrition and high capacity of resistance to dry summers, Moldova, sunflower crops is one of the basic and most important source of vegetable oil. But the sunflower crop harvest, suffering considerable losses due to adverse action of abiotic and biotic factors such.

There were studied 27 sunflowers plantations infected by broomrape on Moldovan territory. The research was conducted in the second half of August 2014, when broomrape plants were in full ripe seeds.

For the first time farmers have reported this parasite in the 70s in Cahul (village Manta). Most cases of detection of this parasite dates back to the early 2000s sunflower crops, tomatoes, tobacco. The main method of combat of broomrape is mechanical damage by digging.

Plantations were analyzed surfaces between 0.04 ha and 142 ha. Favorite genotypes are hybrids: Pioneer, Paraiso, Subtle, Performer, LE 75, Arena, Donscoi, Drofa.

Repeated cultivation of sunflower is performed in culture medium prior to 3-5 years in most cases as wheat and corn.

Artificial irrigation systems are not used. 20% of the lands analyzed were fertilized with N and P fertilizers, and only in one case were used organic fertilizers. 1/3 of lands were treated with herbicides (Chlororganic). The average yield per hectare being from 0,4 to 2,5 tons. Losses caused by parasite attack reach up to 25-30%.

In the 75% of the cases analyzed was heterogeneous distribution of infection and is manifested by local outbreaks. This is due, in our opinion the phenomenon of seed dispersal mostly confined to contaminated crop seeds owing to poor quarantine services, however, animal grazing, unfermented contaminated manures, wool, fur and farm machinery could be the other sources of seed dissemination.

Wind and flowing water contributes negligible to seed dispersal as the seeds are heavy enough to be dispersed away. The seeds do not float in water because of their high specific gravity and once surface tension is broken they sink in water. Thus, wind and flood water is a low risk vector.

Field investigations highlighted that the parasite the attack level is in dependence of population. The hardest hits were in plantations from southern Moldova region. Here was found a greater uniformity of distribution of infection. Because broomrape in the last 10-15 years has become very damaging parasite, especially in the southern growing areas because of the development of new breeds of virulent parasite requires mapping the current geographical spread of phytopathogenic farmland Moldova, crucial for organizing effective programs to improve plant resistance. Moreover, data obtained from a rigorous assessment of land cultivated sunflower broomrape infected, would contribute to proper training of farmers on the use of resistant hybrids, crop rotation observance and application of modern technologies to prevent and combat the pest.

This parasitic weed has the tendency to proliferate well in coarse textured soils with high pH, low in nitrogen status having poor water holding capacity where the crop cultivation is either rain fed or dependent on sprinkler systems for irrigation.

MORPHO-ANATOMICAL ASPECTS OF TWO BASIL CULTIVARS PLANTLETS UNDER EXPERIMENTAL HEAVY METAL CONTAMINATION

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Recently the use of plants in substrate remediation emerged as a promising alternative for elimination of heavy metals excess in soil and water. Among species able to develop in polluted substrates, plants producing medicinal bioactive compounds are important when their composition is not modified as result of contamination, such as in essential oils (1). The basil is largely used in alimentation or as essential oils in perfumery and medicine. The specie has a high morphological and chemical variability, various chemo types based on essential oil composition existing, with known antimicrobial, insecticidal, antioxidant activities etc. (2). Cerium ($^{58}\text{Ce}_{140,116}$) is one of the most abundant rare Earth elements (REE), up to 50 ppm in the crust, used in the composition of some metal alloys, in glass polishing, semiconductors etc. (3). The present paper investigates the ability of two basil cultivars to germinate and develop in experimental Ce pollution.

Seeds of Sweet Genovese and Red Rubin basil cultivars were germinated in Petri dishes, on filter paper (25 seeds/dish, 3 replications per treatment) using a growth chamber with 12:12 h photoperiod, 22-24 °C, 60% humidity. The treatments consisted in 10, 20, 50, 200, 400 mg/l Ce in a volume of 4 ml/dish. Anatomical structures were observed on colored razor sections, under a Novex microscope. The number of germinated seeds was recorded until the 8th day. The root and shoot lengths and fresh weight were recorded after 8 days. Anova and Tukey tests were used to assess differences at $p < 0,05$.

Anatomical observations of roots indicate reduced number of absorbent hairs depending on the Ce concentration. The hypocotyls present, in higher concentration treatments, an increased number of tectory hairs, and reduced numbers of secretory hairs. Apart from these differences, the anatomical organization of investigated organs was the same for controls and treatments. The treatments with various Ce concentrations did not significantly influence the germination process. Root length was reduced at higher concentrations (20 – 400 mg/l Ce). Hypocotyle length was reduced at 200-400 mg/l Ce, but it developed normally or was stimulated at lower concentrations. The relatively normal germination and growth at lower concentrations suggest the presence of physico-chemical or physiological mechanisms allowing the reduction of Ce toxicity. The presence of mucilages at germination may contribute to the absorption of this element from the solutions, as shown for copper (4).

Sweet Genovese and Red Rubin cultivars present relatively normal germination and initial development under 10, 20 and 50 mg/l Ce. The results justify further evaluation of the phytoremediation potential of the two cultivars by elemental analysis and analysis of essential oil composition for Ce presence.

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BIOLOGICAL AND PHYTOCHEMICAL STUDY OF SOME NEW CULTIVARS OF BLACKBERRY, MULTIPLIED BY BIOTECHNOLOGY IN VITRO

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Blackberry present particular interest as valuable raw material in herbal medicine and healthy food in contemporary nutrition. Geneticists and breeders trying to get new blackberry cultivars, varieties and forms with special features: frost resistance; phytopathogens action; different terms of fruit ripening with longer storage; new qualities of taste and resistant to mechanical actions. These new created genetic forms of blackberry can ensure consumers with fresh, rich in various vitamins and other chemical constituents with invaluable therapeutic and alimentary value for a longer period.

To highlight the biological, technical, phytochemical (qualitative and quantitative analysis of ascorbic acid, flavonoids and tannins) characteristics were taken for scientific research three blackberry new cultivars of American origin: *Thornless Evergreen* obtained from species *Rubus laciniatus* L.; *Cester* and *Arapaho* – from species *R. fruticosus* L., Rosaceae family, multiplied *in vitro* and acclimatized to the climatic conditions of our region by Laboratory of Embryology and Biotechnology of the Botanical Garden (Institute) of Academy of Science of Moldova.

The new cultivars are characterized by different fruit maturation period: *Arapaho* – early, the other two *Thornless Evergreen* and *Cester* – less late. Cultivars differ on the biological characteristics of plant: repent stems – *Thornless Evergreen*, semierect twig – *Cester*, and erect growth – *Arapaho*; greater vigourity – *Cester* and *Thornless Evergreen* cultivars and semivigourity – *Arapaho* cultivar; *Cester* and *Evergreen* cultivars are more resistant to frost specific to Moldova than *Arapaho* cultivar, but cultivar *Evergreen* supports easiest insufficient humidity and the highest temperatures of summer period; all cultivars were demonstrated the moderate resistance to specific pathologies of blackberry, but *Cester* cultivar was mentioned as very resistant to foliar pathologies.

The fruit, which is plant organ exploited both for food and therapeutic, is distinguished by the following morphological and technical indicators: fruit shape – oval-spherical for *Cester* and *Thornless Evergreen* cultivars, cylindrical – for *Arapaho*; fruit weight – 4,5 g for *Thornless Evergreen* and *Cester* cultivars and 7,5 g for *Arapaho*; *Thornless Evergreen* and *Arapaho* cultivars have sweet taste, but *Cester* cultivar has sweet-sour taste.

Comparative analysis of ascorbic acid in fruit cultivar investigated reveals its presence as a white spot on pink background in all analyzed samples (thin layer chromatography). Qualitative study of ascorbic acid in fruit cultivars analyzed by titrimetric method demonstrates that the cultivar *Thornless Evergreen* highlighted with the highest content (0,30%), followed by cultivar *Cester* (0,26%) and the lowest content in cultivar *Arapaho* (0,23%).

Qualitative and quantitative study of tannins show differences that correlate with blackberry analyzed cultivar. Qualitative analysis based on specific color and sedimentation reactions highlights the presence of hydrolysable tannins in the fruit of all three cultivars; only the result intensity of reactions is different, more intense in the fruit extracts of *Thornless Evergreen* cultivar, being compared to the other two cultivars. The analysis results obtained by the titrimetric method for the determination of the content of tannins shows that the richest fruit in this content are *Thornless Evergreen* cultivar with 11,08%, then the descent – *Cester* with 9,23% and *Arapaho* – 8,31%.

For qualitative study of flavonoids applied to complex of specific chemical reactions (staining and sedimentation), and the results analyzed revealed the presence of following flavonoid constituents: anthocyanins, leucoanthocyanins, flavanone, flavone and flavonol in fruit analyzed cultivars. Quantitative study of flavonoids by spectrophotometric method showed that fruit of *Arapaho* cultivar is the richest – 3,459%, slightly lower amount in fruit of *Cester* (3,385%) and *Thornless Evergreen* (3,314%) cultivars.

The results of biological and phytochemical studies show that all new cultivars of blackberry (*Thornless Evergreen*, *Cester* and *Arapaho*) were well acclimatized to the climatic conditions of Moldova and possess resistance to phytopathogens. Biological characteristics of the plants, technical features and content of active chemical principles of the fruits will serve as support to the management of blackberry plantation on larger areas.

PHYLOGENETICAL APPROACH FOR THE SEARCH OF VALUABLE METABOLIC PRODUCTS IN CYANOBACTERIA

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Cyanobacteria are an important group of microorganism that plays a crucial role in both terrestrial and aquatic ecosystems. Species are beneficial as sources of food, as well as producers of carotenoids, antioxidants and other secondary metabolites. Mass development of cyanobacteria in natural and artificial reservoirs can also cause negative consequences due to the accumulation of toxins. We propose using a phylogenetic approach to detect strains of potential value for the production of various secondary metabolites. Existing phylogenetic methods lack precision and only make use of rDNA or single gene approaches, thus preventing a more detailed understanding of the evolutionary trajectory of cyanobacteria. Advances in molecular and genome studies have revealed the main genes involved in different stages of secondary metabolites production. Several genes have been identified in *de novo* TAG biosynthesis pathway and are known to be involved in lipid synthesis (rbsL, me g6562, accA, accD, dgat g2354, dgat g3280, gat g7063), production of malic enzyme subunits (ACCase and diacylglycerol acyltransferase), and carotenoid synthesis (crtB, crtP, crtQ-1, crtQ-2, crtL, crtL-2, crtO, crtR). The first group of genes is well studied in relation to biofuel production, while the second group has been widely employed in the pharmaceutical, cosmetics, and food industry for natural dyes and antioxidants. This paper will demonstrate how phylogenetic techniques can be used to search for novel strains of carotenoid producing cyanobacteria by elucidating the evolutionary relationship between genes involved in carotenoid synthesis. Many species of carotenoid producing cyanobacteria are already known from the genera *Anabaena*, *Arthrospira*, *Calothrix*, *Lyngbya*, *Microcystis*, *Nostoc*, *Phormidium*, *Scytonema*, *Synechococcus*, *Tolypothrix* and others. *Arthrospira platensis* (*Spirulina*) and *Nostoc commune* are already used in commercial scale production of biomass and metabolites, and several taxa (i.e. *Anabaena cylindrica*, *Calothrix desertica*, *Nostoc sp.*) have sequenced, assembled and annotated genomes. Data are publicly available from the National Center for Biotechnology Information (USA), the National Library of Medicine (NCBI of NLM), Joint Genome Institute of Department of Energy (USA) (JGI of DoE) and other research institutions. A phylogenetic analysis revealed which species of cyanobacteria are closely related to known carotenoid producing species. Phylogenetic results were compared with empirical data of secondary metabolite production to compare carotenoid production with total biomass per strain. Our approach also proved useful in detecting producers of dangerous toxins. It was determined that genes *mcyA*, *mcyB*, *sxtI*, *cyrA* are responsible for toxin production and may be used as early indicators of potential toxicity of biomass or biomass-derived products. A phylogenetic approach is shown to be an indispensable tool in phycobiotechnology that can greatly facilitate identification of novel strains, as well as insure the quality and safety of biological products.

INFLUENCE OF MILLIMETER RADIATION ON VIABILITY OF SEEDS OF VARIOUS PLANT SPECIES AFTER *EX SITU* CONSERVATION

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Laboratory of Plant Genetic Resources of Moldova in the Institute of Genetics, Physiology and Protection of Plants conducts years-long investigations on the use of low electromagnetic field of the millimeter range or millimeter radiation (MMR) for increasing viability of seeds of various plant species (vegetables, grain crops, cereals, pulses, technical and medicinal plants) after *ex situ* conservation. Principal attention is paid to seeds with different periods of storage in gene bank. It is known that irreversible physiological and biochemical changes occur in seeds during their aging and result in drastic decrease of seed viability that should be restored for further reproduction and their use in plant breeding. This approach is one of the most important problems of gene banks.

Influence of the following parameters of MMR was studied: wavelength (4.9 mm; 5.6 mm and 7.1 mm), radiation exposures of seeds (2 to 30 min) and power density (2 to 10 mW/cm²). Seed viability was assessed by various parameters including morphological (sign of bioisomerism and seedling size), physiological (germinability and seed germinating power, seedling growth rate), biochemical (enzyme activity and protein synthesis in seedlings, pigment content in leaves of seedlings, etc.) and genetic (number and type of chromosome aberrations in cells of rootlets) parameters.

It was revealed that millimeter radiation is characterized by predominantly uniform type of effect on seeds regardless of their species and genotype. Stimulating regimens of MMR influence on seeds are often similar and characterized by the following parameters: exposure (2 to 10 min), power density (6 to 10 mW/cm²), and wavelength (5.6 mm). Stimulation of morphological and physiological parameters was more than 20%.

A hypothesis was made about water as primary receptor of MMR shared by all objects (seeds) and present both inside the object and outside but in contact with the object. This hypothesis agrees with the literature data.

Millimeter radiation caused reduction of chromosome aberrations in meristem of embryonic roots of seedlings, and this allows to revive seed and planting material, i.e. to correct chromosomal breakages and eliminate undesirable mutations that appear in case of long-term storage of seeds.

Radioprotective and radioreparative effect of millimeter radiation was discovered during its influence on seeds before and after their treatment with γ -radiation, i.e. with the help of MMR we can both prevent seeds from harmful consequences of γ -radiation and mitigate its negative effect thus obtaining non-trivial mutations.

Stimulating effect of millimeter radiation manifests itself in: 1) decreased enzyme activity of IAA oxidase, 2) increased content of pigments in leaves of seedlings; 3) increased total amount of freely-soluble proteins in seedlings; 4) reduced incidence of chromosome aberrations in cells of embryonic roots of seedlings; 5) changes in spectrum of chromosomal abnormalities primarily due to decreased number of separate chromosome bridges.

Water that was radiated with millimeter waves stimulates germinability of seeds. Stimulating effect in case of water radiation was at least equivalent to stimulating effect obtained with radiation of seeds. Water radiated with MMR acquires "memory" of this influence (it retains ability to exert stimulating effect on seeds). Persistence of water memory increases in case of storage of seeds at low temperatures.

Results of 3-year studies demonstrated that influence of millimeter radiation with the wavelength of 5.6 mm, power density of 10 mW/cm² and radiation exposures of 2 and 8 min on tomato seeds with extended storage period (6 to 12 years) led to significant increase of plant productivity in the field conditions (up to 30%). The point to be emphasized is that stimulating effect of millimeter radiation depends on period of seed storage, on cultivar and on climatic conditions during the year of testing.

Method of pre-sowing treatment of seeds with millimeter radiation is more preferable as compared to γ -radiation of seeds in terms of its safety, economic efficiency and technological feasibility.

Thus summing up the aforesaid, it seems appropriate to recommend the use of millimeter radiation for increasing viability of seeds of various plant species after *ex situ* conservation as well as for increasing productivity of plants obtained from these seeds in the field conditions.

GROWTH REGULATORS OF STEROIDAL NATURE IN TECHNOLOGY CULTIVATION OF WINTER BARLEY

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Were studied the features of action of preparations of steroid glycosides Moldstim (MS) and Ecostim (EC) on the indicators of growth, photosynthetic activity and productivity to winter barley varieties intensive (Buran) and plastic (Osnova) type, placed in two predecessors crops (peas and soya) in the field crop rotation, legumes saturated.

It is established that the treatment plants of winter barley of *tillering – beginning of booting* stage of stem elongation preparation solutions MS and ES at a dose of 25 mg/l increase the growth parameters - the length and diameter of the stem and the leaf area at 1,1-1,7 times in the studied varieties as compared by the control. It increases the active work of the leaves during ontogeny and accumulation by plants of raw and the absolutely dry biomass in 1,1-1,5 times the variety Buran, in 1,2-2,1 times the variety Osnova.

Under the influence of the preparations MS and ES the activation occurs the photosynthetic activity that leads to the accumulation of plastid pigments - chlorophylls *a*, *b* and *carotenoids* in assimilating plant organs.

The influence of growth regulators to the content of plastid pigments – is one of the most regulated effects of their actions. It was revealed the variety-specific reaction of plants on their application. In the variety Buran appeared stimulating, prolonging their action in the accumulation in the leaves of plastid pigments - chlorophyll *a*, *b* and *carotenoids* during the vegetation period. Reaction of grade variety Osnova is inadequate action to the growth regulators, it depends on the phase of vegetation. The preparation solutions MS and ES stimulate the formation of plastid pigments only in the initial stages of their actions at this varieties (5th day after spraying) and cause depression in their accumulation in the next phase of growth and development of plants.

The calculations of the chlorophyll index, conducted by us, show that the total accumulation of chlorophyll in winter barley plants under the action of preparations MS and ES increases in 1,3-3,9 times independently of the varieties.

It was revealed a direct correlation between the accumulation of biomass, chlorophyll index and productivity of winter barley varieties by grown on various predecessors. Analysis tie showed its variability due, depending on the type of precursor. By soya varieties growing, the correlation coefficients calculated in the phase of earing are high ($r = 0,95-0,99$), on peas - average ($r = 0,43 - 0,63$), especially in the less favorable meteorological conditional years.

It was found, that the increasing growth of plants and photosynthetic functions under the influence of preparations MS and ES leads to increased productivity indices elements, increasing the mass of the spike, the grain mass per spike and productivity varieties in 1,1-1,5 (peas) and 1,3 - 1,7 times (soya), compared by the control.

Treatment of vegetative plants preparations steroid glycosides leads to the outflow of nutrients from vegetative to reproductive organs and their accumulation. Increases the content of the main groups of nutrients in the grain - protein 0,27-1,82%, starch 0,40-2,98%, fat 0,05-0,98% and reduces the amount of fiber and ash.

The experimental dates obtained by us, allow to make a conclusion about the prospects of the use of drugs of steroid glycosides in the crop production practice in order to increase the photosynthetic activity of plants of winter barley varieties, to increase crop yields, to improve product quality, as well as resistance to different environmental factors when growing them in crops by legumes predecessors.

ПОЛИПЕПТИДНЫЙ СПЕКТР СУММАРНОГО БЕЛКА СПИРУЛИНЫ, КУЛЬТИВИРУЕМОЙ В ПРИСУТСТВИИ СОЕДИНЕНИЙ GeO_2 И GeSe_2

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В последние десятилетия цианобактерия *Spirulina platensis* интенсивно используется в качестве биотехнологического объекта для получения биоактивных веществ, в том числе микроэлементов в биодоступной форме. Особый интерес представляет получение биомассы спирулины, содержащей такие микроэлементы как селен и германий.

Селен является необходимым микроэлементом для нормального развития живого организма, активирует процессы дыхания ткани, регулирует окислительно-восстановительные реакции, влияет на иммунологическую активность, на белковый метаболизм, в особенности на метаболизм аминокислот, содержащих серу. Его терапевтические возможности широко известны. При использовании спирулины, обогащенной селеном, в профилактических и лечебных целях в составе БАД отмечают положительные сдвиги у больных онкологическими заболеваниями (в особенности рака простаты). Биологическая роль селена зависит, прежде всего, оттого, что он является непосредственной составляющей глутатионпероксидазы, участвует в защите животных и растительных клеток от окисления, является катализатором образования тиреоидного гормона.

Германий – является сильным антиоксидантом. Также он обладает важными биологическими свойствами: способностью обеспечивать перенос кислорода в тканях организма; повышать иммунный статус организма; проявлять противоопухолевую активность и др.

Поскольку неорганические соединения этих микроэлементов, полученные химическим, путем являются токсичными, поэтому целесообразным является поиск альтернативных источников селена и германия биологического происхождения.

Целью данной работы было культивирование спирулины в присутствии соединений GeO_2

и GeSe_2 и изучение изменения состава полипептидного спектра суммарного белка SDS электрофорезом.

Для культивирования спирулины была использована модифицированная питательная среда Заррук. Культивирование осуществлялось в колбах Эрленмейера с объемом 100 мл культуры, в течение 144 часов при температуре $30^{\circ}\pm 1^{\circ}\text{C}$, освещение 2000-4000люкс, pH среды 9,5-10,0. Соединения добавлялись в следующих концентрациях: GeO_2 - 30мг/л и GeSe_2 - 20мг/мл.

Белки экстрагировали при комнатной температуре в буфере: 0,628 мМ Трис-HCl, pH 8,0 (1:5), 0,03% аскорбиновой кислоты, 1 мМ ЭДТА и 1% меркаптоэтанол в течение 30 мин с последующим центрифугированием 15 мин при 6000 об/мин. Белок осадков проводили с сульфатом аммония при концентрации 90%, затем осадок промывали 5% ТХУ. Полученный осадок промывали этанолом, ацетоном, эфиром, сушили и использовали в анализе.

Результаты электрофоретического анализа спектра полипептидов суммарного белка спирулины, культивируемой в присутствии соединений GeO_2 и GeSe_2 , показали присутствие двух интенсивных полос с кажущимися молекулярными массами 20,5 кДа и 19 кДа соответствующих α и β субъединицам фикоцианина, которые в количественном отношении намного выше по сравнению с эталонным образцом, полученным из спирулины, выращенной на среде Заррук. В случае образца, полученного из спирулины культивируемой в присутствии GeSe_2 наблюдается еще одна интенсивная зона с кажущейся молекулярной массой 16 кДа, которую можно отнести к β - субъединице аллофикоцианина.

Таким образом, при культивировании спирулины в присутствии GeO_2 наблюдается стимулирующий эффект на синтез фикоцианина, а в присутствии соединения GeSe_2 индуцируется не только синтез фикоцианина, но и аллофикоцианина. Эти 2 соединения могут быть использованы для получения биомассы спирулины, обогащенной как фикобилипротеинами, так и германием и селеном. Полученная биомасса может быть использована в фармацевтике, медицине, косметологии и в пищевой промышленности.

NOI PROCEDEE DE OBTINERE A BIOMASEI DE SPIRULINĂ CUPRU-, FIERO- ȘI CROMOCOMPONENTE

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Biomasa de spirulină este considerată una dintre cele mai importante surse nutriționale nonconvenționale utilizate în prezent, care oferă un spectru larg de substanțe bioactive. O direcție prioritară în cercetările biotehnologice ale spirulinei este obținerea biomasei fortificate cu microelemente importante. Ascensiunea continuă a maladiilor legate de afecțiuni metabolice ca diabetul zaharat și anemia, însoțite totodată de deprecierea nivelului funcției imunitare a organismului invocă soluționarea urgentă a aspectelor legate de elaborarea unor produse cu acțiune polivalentă și capacități terapeutice funcționale înalte. Drept soluție poate fi considerată oportună explorarea bioaditivelor cu efect nutraceutic imunostimulent, antianemic, antidiabetic, antiinflamator și anticancerigen obținute în baza biomasei de spirulină cu conținut prognozat de principii bioactive, cupru, fier și crom metabolizate – bioelemente cu implicare majoră în manifestarea anemiei și diabetului.

În contextul celor expuse mai sus ne-am propus ca scop elaborarea unor procedee noi de obținere a biomasei de spirulină cu un conținut majorat de cupru, fier și crom, ce ar putea servi drept sursă de obținere a unor preparate imunostimulatoare, antianemice și antidiabetice noi.

În calitate de obiect de cercetare a servit cianobacteria *Spirulina platensis* depozitată în Colecția Națională de Microorganisme Nematogene a R. Moldova de pe lângă Institutul de Microbiologie și Biotehnologie al AȘM. Pentru cultivarea spirulinei a fost utilizat mediul nutritiv mineral *SP-1* cu o compoziție echilibrată a macro- și micronutrienților. În calitate de reglatori ai creșterii au fost utilizați următorii compuși coordinați ai Cu(II), Fe(III) și Cr(III): bromo-[3-[(2-hidroxi-5-nitro-benziliden)-amino]-propan-1,2-diolo](1-cupru) $[(\text{Cu}(\text{L-H})\text{Br})]$, $[\text{Fe}_2\text{MnO}(\text{afur})_6(\text{THF})_3]$ și $[\text{Cr}(\text{HSSA})_2\text{Cl}\cdot\text{H}_2\text{O}]$, respectiv.

Rezultatele obținute au demonstrat că conținutul maxim al fierului cu oscilări foarte neînsemnate este realizat în biomasă pentru concentrația de 50mg/l a $[\text{Fe}_2\text{MnO}(\text{afur})_6(\text{THF})_3]$, constituind valoarea maximă 1080 mg%. În cazul cultivării spirulinei în prezența $[\text{Cr}(\text{HSSA})_2\text{Cl}\cdot\text{H}_2\text{O}]$ conținutul maxim al cromului constituie 29,68 mg% din BAU. Cercetările efectuate asupra acumulării cuprului în biomasa de spirulină la cultivare în prezența compusului coordinați $[\text{Cu}(\text{L-H})\text{Br}]$ au demonstrat că, conținutul de cupru acumulat constituie 11,14mg% din BAU.

Teoretic, după cum demonstrează datele obținute în urma aplicării planului „Mișcarea pe gradient”, conținuturile cuprului, fierului și cromului poate fi majorat și în continuare în biomasă prin majorarea concentrației compusului coordinați. Dar în procesele tehnologice de producere a biomasei de spirulină este important de a ține cont și de valorile productivității. Majorarea concentrației compusilor testați în continuare duce la scăderea productivității spirulinei, valorile cantitative fiind cu 10 -52 % mai mici decât martorul.

Avantajele procedeeilor elaborate constituie în majorarea conținutului de cupru de 3,46 ore, fier de 3,5 ori și cea de crom de 2,6 ori în comparație cu biomasa cultivată fără adaosul compusilor coordinați.

Procedeele de obținere a biomasei de *Spirulina platensis* îmbogățite cu cupru, fier și crom legat cu compuși organici componenți ai biomasei de spirulină, ar putea fi aplicate la elaborarea noilor tehnologii de obținere a preparatelor cu un conținut sporit și prognozat de bioelemente cu acțiune antimicrobiană, antiinflamatoare, anticancerigenă, antianemică, antidiabetică ș.a.

BIORATIONAL PESTICIDES IN THE INTEGRATED PROTECTION SYSTEM OF STONE FRUIT CROPS

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Our researches were directed to elaboration of an integrated protection system of stone fruit crops (plum and peach) against pests and diseases with the use of biorational pesticides and growth and immunity promoting factors, allowed by the Regulation for Ecological Agriculture (Regulament (CE) NR. 889/2008). As fungicides the preparation Funecol was used (s.a. – compound complex 2 - (NH) 4OH + + helator adjuvant) with low content of copper (8%) in dose of 4,0 l/ha, as well as biological fungicides – Trihodermin (with content of spores and/or mycelium of fungus *Trichoderma lignorum*) and Rizoplan (with content of liquid culture *Pseudomonas fluorescens*) in dose of 5,0 l/ha and 2,0 l/ha respectively. As insecticide of the natural origin Pelecol was applied (s.a. fatty acids esters + adjuvant of surface-active substances) in dose of 8,0 l/ha. Additionally, as immunity inducers and growth promoting factors there were used in gardens the preparations of the natural origin, total three versions (elaborated by the collaborators of IGPPP ASM): Reglalg (preparation was elaborated under the direction on the basis of the natural extracts from algae) in dose of 8,0 l/ha, Recol (on the basis of the vegetable extract from *Reynoutria sachalinensis* (F. Schmidt) Nakai) in dose of 5,0 l/ha and Albit (with content of purified active substances from soil bacteria *Bacillus megaterium* and *Pseudomonas aureofaciens*) in dose of 0,5 l/ha.

The experiences were carried out in plum tree (4,0 hectares, “Stenlei” and “Cabardinca raniaia” varieties) and peach (6,0 hectares, “Colins”, “Reithoven” varieties) gardens Ltd. „Agrobrio”, Bacioi village, Ialoveni, Central part of Moldova during the vegetable season 2013 according to generally accepted methods (*Îndrumări metodice*, 2002). The formation of trees crowns is typical, both gardens are situated in good agro-technical conditions, the soil between the rows and in rows – chernozem. The control and mass capture of the main pests (*Grapholita funebrana* (Treitschke, 1835), *Grapholita molesta* (Busck, 1916), (Lepidoptera: Tortricidae) and *Anarsia lineatella* (Zeller, 1839), (Lepidoptera: Gelechiidae) were carried out with the help of standard pheromone traps (“Delta” type), produced by IGPPP ASM. The determination of biological effectiveness of the mass capture method was carried out in the period of mass hatching out of larvae and appearance of pupae, against each generation of the pests according to generally accepted methods (Doljenko, 2004). To control the aphids population there was used the method of direct calculations (Dunayev, 1997). For specific determination of gathered material there were used the corresponding determinants of insects and magnifying arrangements (Nartshuk, 2003; Shaposhnikov, 1964).

After carrying out of the protective measures, as a result of the controls, there was established that the biological effectiveness of the mass capture method of males of *G. molesta* and of *A. lineatella* on peach before the harvest made up 93,4% on fruits and 41,9% on shoots in comparison with the control. The biological effectiveness of the mass capture method of males on plum tree *G. funebrana* before the harvest made up 81,3% (on fruits injuriousness) in comparison with the control. The application of the natural insecticide Pelecol (8,0 l/ha) has reduced the density of aphid colonies (*Hyalopterus pruni*) both on plum and peach trees less than threshold values. At that the population of the natural predators has been preserved at the level higher than the chemical standard, the correlation predator victim made up on average 1:10-15.

We have determined that the replacement of the chemical preparation to the natural fungicide Funecol (4,0 l/ha) on plum tree has promoted the decrease of monilia evolution till 1-2%. In the chemical standard the affection of the fruits with monilia has reached 4%. As well, it was established that the percentage of distribution and development of red spotting (agent - *Polystigmia rubra* Sacc.) and bacterial spotting (agent – bacterium *Xanthomonas pruni* (Smith.) Dowson) on plum tree was at the level of the chemical standard and has not exceeded the threshold value. The biological effectiveness of preparation Funecol against monilia on peach tree as well was at the level of the chemical standard. There was detected that the percentage of distribution and development of leaf curl of peach (agent - exoascale fungus *Taphrina deformans* Tul.), powdery mildew (agent *Sphaerotheca pannosa* Lev. var. *persicae* Woronich.) and cytosporosis (agents – *C. leucostoma* (Pers.) Sacc., *C. schulzeri* Sacc. et Syd.) was at the level of the chemical standard and has not exceeded the threshold value.

The obtained data attest that the replacement of the synthetic pesticides to biorational in the integrated protection systems of plum and peach trees do not decrease the effectiveness of protective measures. The reduction of chemical treatments, as well location along the gardens of forest belts, not undergone to treatment, favors to accumulation of biota and increases the ensuring of the crops with the predators, in the first place by Coccinellidae, Chrysopidae and Syrphidae.

IMPACT OF FERTILIZERS WITH DIFFERENT FORM OF NITROGEN ON SOYBEAN PLANT PRODUCTIVITY AT SOIL WATER DEFICIENCY

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Soybean plant tolerance to soil water deficiency may depend on reserves of N-containing compounds. Success can be provided by the effectiveness of their storing during the initial period of the growing season (before the active symbiotic nitrogen fixation) and the ability of plants to use these reserves during the period of arising water stress. Previous investigation has shown the essential role of exogenous sources of nitrogen in the maintenance of a coordinated relationship between soil agrochemical parameters and the biochemical parameters of processes taking place in the rhizosphere soil and roots of soybean plants.

The aim of presented research was the identification of exogenous sources of N which may contribute to the productivity of different soybean cultivars at soil water deficiency. The investigation has been performed in the northern agro-pedo-climatic zone of Moldova in frames of Experimental station of Research Institute of Field Crops (RIFC) „Selectia” (Bălți). Three soybean (*Glycine max* L. Merr) cultivars (Indra, Aura and Magie) have been grown in a field experiment on a nonirrigated typical chernozem with the preplant application of one of three kinds of nitrogen mineral fertilizers (ammonium nitrate, or urea, or ammophos) at the starter rates N 20 kg/ha.

It was shown, the preplant use of nitrogen fertilizers revealed the benefic effect on soybean productivity against of control plots without exogen N amendment. The realizing the productivity of soybean cultivars was associated with a preference for certain forms of nitrogen in the fertilizer (nitrate, ammonium, amide). Three soybean cultivars grown on typical chernozem demonstrated a clear advantage of urea and ammophos in the seed production compared to ammonium nitrate. Consequently, the reduced form of nitrogen in the fertilizer as ammonium and amide provided a much more favorable effect on the formation of seed yield by soybean plants in comparison to oxidized N form. In the climatic conditions of the year 2014 cultivar Indra showed the highest seed yield practically equal with the use of urea (26,9 q / ha) and ammophos (26,7 q / ha). Similarly for cultivar Aura the maximum seed yields were recorded at ammophos (26,0 q / ha) and urea (24,1 q / ha) administration. Cultivar Magic indicated the same trend in response to exogenous introduction of different forms of nitrogen. The highest yield was obtained by using urea fertilizer (18,6 q / ha).

The relative increase of seed production as a result of the use of ammonium nitrate were in the range of 18% ... 28%, depending on cultivar, while in the case of urea was stable 35% ... 36%, and for ammophos introduction - 30 % ... 45%. Cultivar Aura was the most receptive to additional nitrogen fertilizer management compared to the control. The soybean seed yield increase owing to the preplant ammophos application reached 45%.

Since the 2014 growing season was characterized by insufficient atmospheric precipitations taking place at Balti steppe, the distinct advantage of the reduced form of nitrogen fertilizers (ammonium, amide) for the three soybean varieties confirmed the working hypothesis on the universal mechanism of urease activation in soybean roots, reflecting an increase in the consumption of substances containing N as ammonium and amide at soil drought conditions.

Protein and oil content in soybean seeds of the studied 3 cultivars did not differ significantly among different nitrogen fertilizer treatments, but the calculation of efficiency of these products in terms of seed yield per hectare demonstrated a significant material importance.

Thus, a clear advantage of the reduced form of nitrogen (amide inside of urea or ammonium inside of ammophos) on soybean productivity was established for the example of three soybean cultivars grown at low soil water content. In order to achieve the productivity potential of soybean plants at soil water deficiency the fertilizers urea and ammophos may be recommended.

TEHNOLOGIE INOVATIVĂ DE OBTINERE A MANOPROTEINELOR DIN LEVURILE GENULUI *SACCHAROMYCES* CU APLICARE PRACTICĂ ÎN DIVERSE DOMENII

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Actualmente, polizaharidele parietale obținute din levuri își găsesc aplicarea practică în diverse domenii - medicină, industria farmaceutică, industria alimentară, vinificație, cosmetologie, etc.

Datorită proprietăților sale specifice, manoproteinele pot fi utilizate în calitate de stabilizatori și agenți de îngroșare, stabilizare și dispersie. Studiile recente au demonstrat, că manoproteinele se pot folosi în medicină ca antigeni imunoreactivi în diagnostică unor maladii, ca substituenți ai plasmei sanguine, ca agenți naturali de protecție cu efect imunomodulator, antineoplastic și antioxidant. O altă direcție, este aplicarea acestora în calitate de gelifianti în producerea siropurilor și gemurilor. Cercetările realizate în domeniul oenologiei, demonstrează eficacitatea utilizării manoproteinelor în scopul stabilizării tartrice și proteice a vinului, contribuind astfel la îmbunătățirea caracteristicilor senzoriale și organoleptice a produsului finit.

Levurile reprezintă un obiect biotehnologic de înaltă valoare și caracter aplicativ în procesele de producție. Interes sporit prezintă levurile ca sursă de polizaharide, în particular manoproteine. Acestea dețin avantaje semnificative față de alte materii prime costisitoare (de origine vegetală) cum sunt viteza înaltă de creștere, ciclul scurt de producere, sinecostul relativ mic a componentelor mediilor nutritive.

În acest context, scopul lucrării a constat în studierea particularităților de cultivare dirijată a tulpinilor de levuri înalt producătoare de polizaharide și elaborarea tehnologiei moderne de obținere a manoproteinelor cu utilizări polivalente.

Rezultatele cercetărilor destinate selectării tulpinii de levuri *Saccharomyces cerevisiae* CNMN-Y-18 - producătoare de manoproteine, concretizării concentrațiilor optime a surselor de carbon și azot, optimizării mediului nutritiv, selectării parametrilor optimali de cultivare dirijată a culturii, precum și stabilirii regimului optim de aplicare a undelor electromagnetice cu frecvență extra înaltă, au servit drept reper pentru elaborarea procedeele de sinteză orientată a polizaharidelor parietale, care sunt incluse în tehnologia nouă de obținere a produselor manoproteice din biomasa levuriană

Astfel, tehnologia de obținere a produselor manoproteice prevede cultivarea tulpinii de levuri *Saccharomyces cerevisiae* CNMN-Y-18, cu potențial sporit de sinteză a manoproteinelor, pe mediul nutritiv optimizat YP(GA), utilizarea undelor electromagnetice (cu frecvență 53.33 GHz, durata de tratare 10 minute), a condițiilor optime de cultivare a producătorului (temperatura 25...27°C; pH=5,5; oxigen molecular 57,8-81,0 mg/l; durata de cultivare 120 ore) și a procedului eficient de extragere a manoproteinelor. Avantajul aplicării acestei tehnologii constă în obținerea unei cantități de 1,9 ori mai mare de manoproteine.

PRODUCTION OF THE GRAIN MOTH EGGS (*SITOTROGA CEREALELLA* OL.) ON DIFFERENT GRAIN SUBSTRATES FOR *TRICHOGRAMMA* ENTOMOPHAGUE REARING.

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During the 2009 to 2010 years, for the production of grain moth (*Sitotroga cerealella* Ol.) on different substrates, there were carried out a series of experiments where the chosen grains were barley, wheat, maize, and triticale for the entomophague *Trichogramma* rearing. The substrates of barley, wheat, corn and triticale, each taken in the amount of 5 kg in 5 repetitions, were sterilized in the autoclave at 95-97°C in order to destroy all the grain's pests, such as *Habrcyctus cerealella* Ashm, *Pedicularides ventricosus* etc. After sterilization, the substrates from each variant were dried out and infested with the grain moth (*S. cerealella*) with the rate of 1 gram of moth grain eggs to 1 kg of grains; then followed all the steps from the technology of obtaining grain moth eggs, which was described by such authors as Абашкин, Воротынцева, Гринберг (1979).

The continuous reproduction of entomophague *Trichogramma* on grain moth eggs tends to reduce prolificacy and search ability of females, and significantly diminishes its size. In order to enhance the vitality of laboratory populations it is recommended that the native culture be annually renewed by collecting butterflies' clutches parasitized by *Trichogramma* from the nature. Instead, the host *S. cerealella* is widely used in the production of entomophague *Trichogramma*, because it can be easily reproduced on various grain substrates and is less expensive than other hosts.

The objective of the research was determination of the grain substrate which would be optimal for the production of the laboratory host *S. cerealella* used at entomophague *Trichogramma* rearing.

To achieve the objective, the percentage of infestation on different substrates was determined along with the biological indices of *Trichogramma pintoi* Voeg. and *Trichogramma evanescens* Westw., reared on grain moth eggs from different cereal substrates. In order to estimate the percentage of infestation of the grain substrates and biological indices of *T. evanescens* and *T. pintoi*, there was determined the percentage of the infected seeds and the percentage of *S. cerealella* eggs parasitized by *Trichogramma* obtained from each substrate. 5000 grains from each substrate were analyzed to establish the percentage of infestation. In order to determine the percentage of parasitizing, 5 grams of moth eggs were used. Each substrate was examined in 5 repetitions.

Analyzing the obtained results, it could be stated that in the variant with the thermal treatment, the amount of infested grains in barley, wheat, corn and triticale substrates varied from 81% - 85,20%. The amount of parasitized eggs observed on the variants by *T. evanescens* varied from 81% - 83%, and 82,5% - 86,4% by *T. pintoi*. In comparison (control) where there was no thermal treatment, these indices ranged respectively: infested grains from 72% to 74,5%; the amount of the *T. evanescens* parasitized eggs ranged from 70% to 74,0%, and by *T. pintoi*, ranged from 71,8% to 75,2%.

Comparing the infestation and parasitizing values of the moth eggs by *T. evanescens* and *T. pintoi* through variants on substrates among thermally treated and non-treated, the difference is not significant ($t_d = 1,87 - 2,35 > 0,05 = 1,96$) in all cases.

It can be stated that the percentage of infestation and the percentage of parasitizing in the variants with thermal treatment is higher than in the control (non-treated thermally). In the variants which have undergone thermal treatment, the quantity of eggs produced on substrates in a 1 kg of barley, wheat, corn and triticale was higher, with a collected amount of 7-8 grams. In control, where substrates have not been treated, this index ranged from 6,0-6,5 gr respectively. This means that from 100 kg of grains, it was obtained by variants 700-800 grams and 600-650 grams respectively in control eggs. The values of the infestation percentage on barley and the parasitizing percentage by *T. evanescens* and *T. pintoi*, on the eggs obtained from barley substrate is slightly higher than in the wheat, corn and triticale. Biological indices of *T. evanescens* moth eggs grown on the barley (preferred) substrate are: for *T. evanescens* –prolificacy – 21,60 eggs / female, hatching individuals (females and males) – 80,5% hatching females – 53,5%, static quality criterion – 9,30. Biological indices of *T. pintoi* – reared on moth eggs on the substrate of barley are: prolificacy – 22,4 eggs / female, number of individuals (females and males hatching) – 80,5%, hatching females – 54,0%, the criterion static quality – 9,8.

Analyzing the results obtained, it can be stated that for the production of the laboratory host *S. cerealella* for the entomophague *Trichogramma* rearing all tested substrates of barley, wheat, corn and triticale can be used with no high difference.

APPLICATION OF BIOTECHNOLOGICAL TOOLS IN FORESTRY

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The problem of biological diversity is one of the main concerns of humankind, and its conservation is a prerequisite for the sustainable development of socio-economic systems. The extinction of numerous species of plants and animals was based on human actions more or less rational. Nowadays, the problem of biodiversity conservation goes beyond the limits of scientific directions and is a stringent objective, a priority of states and international institutions.

In order to achieve performances in this field, it is necessary to continue the research processes in the field, the monitoring, to intensify the activities on environmental education and training, to increase public awareness, conservation and rational use of natural resources, to increase energy efficiency and to reduce the impact of climate change. The natural reservations, public and private parks, green areas of Republic of Moldova, are both providing a source of dendrological biodiversity an essential attribute of developments and with superior decorative traits. These have a rather complex function – ecological role, equilibrium factor for the environment, social and health providing element for the society

Species of ornamental woody plants (*Thuja*, *Juniperus*, *Taxus*, *Chamaecyparis*, *Buxus*, *Eonymus*, *Callicarpa*, *Calicanthus*, *Kerria*, *Buddleja*, *Clematis*, *Vinca* and others) production is marked by a great diversity of species and cultivation practices. The large biodiversity of decorative plants species and their great movement across the globe have allowed many usages, besides food, such as phytotherapy, cosmetics, landscaping, etc. The ornamental value of decorative woody plants results from the morphological characteristics of the plants themselves: size, general habitus of plants, shape, appearance, size and colour of the leaves. The importance of cultivating decorative woody plants with ornamental value originates from the constant need of man for finding beauty. Decorative woody plants with ornamental value, along with other decorative plants help fight air pollution, establishing equilibrium in living spaces. Cultivated by man in green spaces, these plants extend their utilitarian-social importance. The vegetation in parks, gardens, squares and roads visibly influences the microclimate of population centres. They clear the air of contemporary life, which, in the unprecedented development of industry, harms the environment.

Biotechnology has become an integrated part of plant breeding and in recent years new methods have been developed for the breeding and propagation of important plants in the agricultural, ornamental and forestry sector.

Among the various biotechnologies, in vitro techniques and tissue culture are most readily integrated into the improvement of forestry and ornamental woody species. Tissue culture methods of propagation are already a commercial success for a variety of woody crops. The methods for elite trees include using of buds (terminal, axillary and fascicle) and plantlet production from stem callus cultures (organogenesis) as well as artificial embryo production from such callus cultures (somatic embryogenesis).

Although progress in micropropagation of woody plants through organogenesis from organ explants has been achieved, rooting of the micropropagated shoots (microcuttings) and the acclimatization of plantlets are still a problem. According to many workers, further research on the influence of factors such as donor age, genotype, type of explant, microcutting quality, auxin treatment, root system and environmental conditions, on rooting and acclimatization is required.

The team of scientists of Embryology and Biotechnology Laboratory in frame of the Botanical Garden (Institute), have initiated investigations of new plants species for Moldova, which are of interest to the country's economy, whose regeneration is difficult on traditional way. The application of these modern biotechnologies provides planting material of high biological value that meets important criteria such as genetic uniformity, quality of the planting material and cost.

ORIGANUM VULGARE - QUANTITATIVE ASSESSMENT OF ACTIVE PRINCIPLES AND ANTIOXIDANT ACTIVITY IN RELATION WITH SOME ENVIRONMENTAL FACTORS

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Within the same species, the active principles content, the antioxidant capacity and many other specific parameters of medicinal and aromatic plants (MAP's) depend on a variety of factors: subspecies and variety, the substrate in which plants are growing, climatic conditions, etc.

One of the species with a huge potential in using as MAP's is *Origanum vulgare*, species which the subject of the present study. Specimens were collected during 2013 and 2014 from the spontaneous flora of Romania (Tarnita, Brasov, Maramures area, Calinesti and Zugreni) and Moldova (Vasieni and Orheiul Vechi). The extracts used in analyzes were methanol extracts in 2.5% dry:extract-ratio.

Parameters determined followed two major directions: on the one hand plants biosynthetic capacity and on the other hand their antioxidant potential.

According to the literature, polyphenols and flavonoids reduce the oxidative stress in cells and thus, they are used in many human diseases/disorders as anti-proliferative, anti-tumor, antimicrobial, estrogenic, anti-inflammatory activities, but also used in cancer, cardiovascular disease, neurodegenerative disorders, etc.

Biosynthetic capacity was quantified by determining the total amount of polyphenols and flavonoides using spectrophotometric methods. According to the pharmacopoeias, polyphenols were determined using Folin-Ciocalteu reagent; the total content was expressed as equivalent galic acid. Flavonoides were determined based on their capacity to form intensely colored (yellow) complexes in presence of Al^{3+} ; the results were expressed as rutoside equivalent.

Antioxidant capacity was determined by quantifying DPPH and ABTS: a) the free radical scavenging capacity of samples was tested by its ability to bleach the stable 1,1-diphenyl-2-picrylhydrazyl radical (DPPH) [1,2]; the antioxidant activity was expressed as $\mu\text{mol Trolox g}^{-1} \text{ DW}$; b) the 2, 2'-azinobis (3-ethylbenzothiazoline-6-sulfonic acid) radical cation (ABTS) scavenging activity was measured according to the method described by Erel, with some modifications [3]; the antioxidant activity was expressed as $\text{mmol Trolox g}^{-1} \text{ EEP}$.

Overall, significant variations were found both intra- and interpopulational. At intrapopulational scale, environmental factors contributing to the active principles contents and antioxidant capacity plays a minor role, the numerical values determined having a low amplitude. Much larger variations were found between populations, in this case pedoclimatic conditions being primarily responsible.

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EFFECT OF VIRAL INFECTION AND GAMMA RAY ON REGENERATION CAPACITY OF BARLEY EMBRYO-DERIVED CALLUS

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Somaclonal variation in plants regenerated by morphogenesis from callus culture caused by plant response to stress conditions. Numerous reports have demonstrated that tissue culture, gamma rays and viral infection are potential stress factors, which can induce genetic and epigenetic changes. Stresogenic conditions have been found to have a positive effect on regeneration in some crop plants, like flax and oat. The aim of the present study was to investigate the influence of barley stripe mosaic virus (BSMV) and gamma ray on regeneration ability of callus derived from barley immature-embryos.

Three two-rowed spring barley cultivars (*Hordeum vulgare* L., $2n = 14$): Sonor, Galactic, Unirea were used in the experiments. The donor plants were obtained from irradiated seeds with gamma rays (at doses of 100, 150 and 250 Gy), using ^{60}Co as source. The dose delivery rate was 0,16 Gy/sec. Barley plants from both irradiated and untreated seeds were mechanically inoculated with a BSMV extract. All plants were tested for the presence of virus particles by negative contrast electron microscopy.

Spikes were collected at 12 – 14 days after pollination and were stored in the refrigerator at +4°C for 5-7 days.

The callus cultures were obtained on optimized MS medium (Murashige-Skoog, 1962). The statistical analysis of data was carried out using the software package Statgraphics Plus (version 2.1). The differences among the value means were compared using Student's t test. Regeneration capacity was estimated as number of plantlets per regenerating morphogenic callus.

The results of the study showed that regeneration capacity was influenced by the genotype and source of variation (viral infection or gamma rays).

The mean value of number of barley plantlets per regenerating callus in the control variants were respectively 5,7 (Sonor), 5,5 (Galactic) and 3,8 (Unirea).

Viral infection alone reduced significantly ($P \leq 0,01$; 0,001) the number of regenerants in cv. Sonor (47,37 %) and Unirea (55,26 %) comparing with control. In case of cv. Galactic, BSMV conducted to the increasing of this value by 81.81% over the control ($P \leq 0,001$).

Gamma rays influenced the regeneration ability of evaluated cultivars in dependence of genotype and radiation doses. Application of 100 Gy, 150 Gy and 250 Gy of gamma rays caused significantly ($P \leq 0,01$; 0,001) decreased mean value (10,52 - 50%) of the number of regenerants per morphogenic callus in barley Sonor and Unirea as compared with mean value of control, while 100 Gy of gamma ray increased it (16,36 %) in cv. Galactic.

BSMV in association with gamma irradiation caused a significantly reduction of number of regenerants (26 % - 72,73 %) in cv. Unirea (100, 150 and 250 Gy) and in cv. Galactic (100, 150 Gy).

In the cv. Sonor viral infection in combination with gamma rays at doses of 100 and 150 Gy increased regeneration capacity by 7,02 – 26,32 % over the control. Virus infection influenced similarly to gamma irradiation at dose 100 Gy and 150 Gy in barley Sonor and Unirea, except the cv. Galactic, where BSMV enhanced (56,25 %) the number of plantlets over the gamma rays (100 Gy)

Analysis of results showed the importance of genotype in the regeneration capacity of barley calli. The effect of different doses of gamma ray on barley regeneration ability showed that, low doses could be enhanced plant regeneration, but high dose caused significant reduction as compared to the control. Decline in mean values of number of plantlets per regenerative callus in irradiated and infected explants could be attributed probably to the heterogenic state created by these stress factors and activation or deactivation of some genes involved in the regulation of plant morphogenesis.

Obtained data clearly attest, that viral infection alone or in combination with gamma rays can be used for increase somaclonal variation in barley plants.

BIOHYBRID TECHNOLOGY: SYMBIOSIS OF ORGANISMS AND ROBOTS

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Technological and biological systems have their own application fields and challenges. These systems interact with each other only in a few disciplines such as bionics, where biological principles are transferred to engineering design, or in production machinery in agriculture, livestock farming or biotechnology.

Biohybrid system represents a combination of closely interacting biological and technological elements. It can be considered to some extent as a symbiosis between living organisms and programmable autonomous robots. There are several goals targeted by biohybrids. One of them is to provide adaptability, plasticity and self-healing properties for such systems. They often utilize evolutionary processes for self-development of artificial objects, which even involve new principles of the so-called "programmable" or "targeted" evolution. In addition, integrating plants and animals into existing engineering or IT infrastructure allows balancing a coexistence of fast-growing human and natural ecosystems. Thereby a sustainability of natural ecosystems is emphasized. An interesting consequence of this technology is, for example, an information interaction between different species, which is not possible in natural environment. Controlling the robot from biological part is also an important topic, appearing in medical autonomous prostheses or human-robot interfaces. Many kinds of microorganisms and plants are superior in sensing environmental, pathogenic or unconventional impact factors. Such biohybrids, denoted as smart biosensors or phytosensors, are used in traditional technological devices and systems.

Integration of robots and living organisms looks different on micro- and macroscales. For example, the microsystems of size 50-150µm integrate the CMOS functional substrate with simple chemical systems - so-called chemo-hybrids. Technological part controls the liquid environment, hydrophobic/hydrophilic properties of surfaces, electrostatic interactions; bio-/chemo- part relates these elements into a multicellular hybrid organism. The macroscopic integration of robotic elements in plant and animal populations is also different. Plants are usually integrated directly with robots, whereas behavioral interactions occur between animals and robots.

The European Commission has initiated a series of projects related to biohybrid systems. Leading European institutions, combined in consortia within FP7 and H2020 programs, investigate different aspects of biohybrid. For example, the projects ANGELS and CoCoRo explored underwater weak bioelectric field, develop and integrate generators/sensors in underwater robots and studied the interaction of these robots and electric fish. Among others, a subsymbolic communication between animals and a possible robot-animal interaction in underwater ecosystems were the focus of research. The projects EvoBody and Symbion coordinated European activities in the field of artificial evolution of multicellular systems, in particular, on the level between chemo- and bio- hybrids. The project ASSIS|bf explores robot-animal interactions in populations of collective animals such as bees and fish, and control a communication with/between these populations. The project Flora Robotica applies this principle to plant systems, where the symbiosis between plant organisms and technological systems should be reached. For instance, the robots control the light, liquid, EM and other conditions for the plants. Plants perform carrier, sensing functionalities and provides energy to robots. These plant biohybrids are intended to be utilized in a new generation of architectural systems and other artificial objects embedded in human environment.

In line with the project Flora Robotica we collect experimental material for possible use of biohybrid system "vegetable object – robot" in phytosensing/phytoactuating purposes especially for telecommunication and remote control. The idea of this control is based on the discovered effect of non-electromagnetic interactions between plant organisms. The experiments are performed with such biological objects as seeds, seedlings and pollen, and sensitive fluidic measurement systems.

THE DETERMINATION OF THE EFFECTIVENESS OF THE IMPACT OF BAS ON FRUIT CROPS THROUGHOUT THE ACCOUNTING METHOD OF CHANGING CHLOROPHYLL FLUORESCENCE

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One of the most important tasks for fruit crops is finding the best practical way to use the photosynthetic function of the plant as the basis of their productivity. One of the biotechnological element of growing fruit trees is the use of bio regulators of development that increase the productivity and the resistance. Examples are steroidal glycosides selected in the IGPPP from wild-growing and cultural plants: trigonellozid, capsicozid, melongozid, linarozid. The influence of these biological active substances (BAS) is studied more on herbaceous plants.

One of our tasks was to investigate the influence of BAS on the viability, growth, photosynthetic productivity and yield of stone fruit crops and also trying to find the most effective method of evaluating this action. We also studied the aftereffect of different concentrated stimulants, terms of introducing BAS, their influence on different types of crops taking into account the particularities of each varieties and the fruit load of each crop. Different types of crops were cultivated in the lyzimeters of the Institute. The research was done on the young seedlings and fructifying trees of apricots and peaches. The study implied the research from the period of vegetation of bloom until the abscission of leaf of the photosynthetic and respiration gas exchange. The transpiration of leaves was made with the RTM-48A, supplied by the company „Bioinstruments” LTD, analyzing the morphological parameters of the leaves and the shoots, the conditions of the pigmented fund and monitoring of the production process.

These methods consume a lot of energy and demand a lot of resources, both financial and time. The search of the express methods of diagnostics of the effects of BAS brought us to the research of the delayed fluorescence of chlorophyll. As we all know, the chemical factors and the climate are able to change the kinetics and the spectral features of the fluorescence, because the molecule of the chlorophyll is the sensor of the external signals that influence the cell. The research of the kinetics of the fluorescence gave us some important information about the nature of the influence on the intensity of photosynthesis. This is why, besides practicing the traditional methods of research, we chose the accounting method of induced fluorescence chlorophyll, for the determination of the effectiveness of the impact of the studied bio regulators of growth on apricots and peach trees. The measurements were made with the help of the Portable Chlorophyll Fluorometer PAM-2100.

We obtained meaningful data obtained from the analysis of the kinetically parameters like background fluorescence (F_0), maximum fluorescence (F_m), variable fluorescence (F_v), ETR- the relative movement speed of the electrons on the chain of transport, indicators of photochemical and non-photochemical extinguishing of fluorescence. The ratio F_v/F_m or the Yield ratio is used as the indicator of the functionality of the photosynthetic system of the plants. The Y indicator can be easily measured and this is the reason why the ratio is widely used in the most various light reactions of photosynthesis. We established the high degree of result matching, obtained both throughout the traditional methods and the method of induced fluorescence about the BAS influence on fruit crops. Thus, the fluorescent analysis of the photosystem II in the leaves apricots of the variety Kostuijensky has shown that under the influence of capsicozid, the values of Y and ETR were above the controlling point with 17-25%. This is correlated with the results of the photosynthetic productivity: the potential was $229,5 \text{ th.m}^{-2}\text{day ha}^{-1}$ whereas the processed version had a potential of $238,7 \text{ th.m}^{-2}\text{day ha}^{-1}$. The intensity of the photosynthesis in the leaves of the controlled version was $2,30 \mu\text{mol CO}_2\text{m}^{-2}\text{s}^{-1}$ and in the leaves of the version with the use of linarozid – $5,79 \mu\text{mol CO}_2\text{m}^{-2}\text{s}^{-1}$. The surface of the leaves on one peach trees of the variety Collins in the controlled version was $6,4 \text{ m}^2$, whereas in the surface of the leaves processed with capsicozid– $8,5 \text{ m}^2$. The harvest was 4,8 kg and 6,1 kg from one tree respectively. The differences in the effectiveness of the photosystem II (Y) and in the speed of movement of electrons in the ETR of these versions were the same as mentioned above.

The processing of leaves with biological active preparations causes changes in the intensity of the chlorophyll fluorescence of leaves. The changes in the optical characteristics of the chlorophyll, as a reaction on the external impact make possible the express diagnosis of the functionality of the pigment system with the help of the fluorometer. The accounting method of variable fluorescence allows us to evaluate quite objectively and easily the effectiveness of the BAS influence on the photosynthesis of different varieties of apricot and peach trees.

USING PLANTS SATURATED BY NANOPARTICLES AS A PHOTOELECTROCHEMICAL CELL

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Photoelectrochemical cells (PEC) convert solar radiation into electricity. Three types of PEC are known [1], the most popular type represents the combination of photovoltaic cell with a conventional electrochemical cell. Drawbacks of available PECs are not only their complex structure, but also the manufacturing, which requires significant financial and human resources. Moreover, the composing materials become corroded and during recycling they pollute the environment.

In this paper we propose a PEC consisting of photoelectrode, electrolyte, and electrolyte bridge. Photoelectrodes are created from plants with a high intensity of growth. Chlorophyll is a part of light-harvesting complexes. It acts as an antenna transmitting solar energy to the reaction centers I and II. The whole plant represents a photoelectrode (anode, cathode) and is saturated by nanoparticles.

The saturation can occur: a) through seeds (soaking); b) by inserting the cuttings of plants into a dispersion of nanoparticles; c) spraying the vegetating plants with an aqueous dispersion of nanoparticles, and by other methods of plant growing. It is possible to inject an aqueous dispersion of nanoparticles, i.e. a rapid saturation of already grown plants by nanoparticles. In this way, inorganic nanoparticles are distributed throughout the plant, moving a) through their roots and stems into leaves or b) through the leaves into the stem and roots. They complement a power of light-harvesting complexes of plants, interact with these systems and make a contact (electric double layer) with the electrolyte.

For a plant impregnation it is desirable to use metal nanoparticles of Au, Ag, Cu, platinum metals or oxides, salts, nonmetals with semiconducting properties, or their mixtures with a particle size of from 0.2 to 100 nm. Smaller nanoparticles (less than 0.2 nm) can be rapidly dissolved (oxidized) in the plant, and are therefore undesirable. Larger nanoparticles poorly penetrate into the plant. The concentration of the nanoparticles must not be toxic to plants and their usage is restricted by keeping the photosynthetic functionality. Different substances are used as the electrolyte: water solutions of various substances, pastes, emulsions, non-toxic-to-plants porous materials.

To create a PEC one a photoelectrode (Au) is dipped in a 0,001 M KCl solution of anode space. Another photoelectrode (Cu) is immersed in the same solution of cathode space. The anode and cathode spaces are connected by an electrolyte (saturated KCl solution) bridge. EMF of a PEC was measured by the compensation method: EMF of such a cell is about 0,24 V in the shade (cloud) and 0,45V in sunlight (no clouds) in the summer at 32 °C.

When soaking seeds of cereal crops and vegetables in an aqueous dispersion of metal nanoparticles, these crops increase yield from 20% to 30% [2]. Apparently, the metal nanoparticles at early stages of development and growth help to better utilize a solar energy by photoelectrochemical structures of the plant. This feature allows the plants to be more resistant to adverse environmental factors of the habitat.

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2.Maslobrod S. N., Mirgorod Yu.A. et.al. *J. Nano- Electr. Phys.*, **2013**. V. 5. No.4. P. 04018(3pp)

THE CUTTINGS ROOTING OF DIFFERENT CULTIVARS OF BLACKBERRY X RASPBERRY HYBRIDS

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The exchange of plant material between institutions from one country as well as from institutions from foreign countries is a very important thing for the conservation and for the overspread of genofond and for scientific purposes.

In december 2014 Botanical Garden (I) obtained from such an exchange of plant material 18 unrooted cuttings of 6 cultivars of blackberry x raspberry hybrids from the National Clonal Germplasm Repository (Corvallis, Oregon, USA). The received cultivars are: Olallie, Kotata, Chehalem, Phenomenal, Santiam, Lincoln Logan.

The propagation by seeds have a lack, progeny plants are not identical as productivity with the parent plant. The propagation by cuttings is a vegetative propagation, as a result the characteristics of the parent plant are kept identical. That's why some plants is better to propagate by cuttings or grafting.

The hybrids of *Rubus loganobaccus* L.H. Bailey HYBRIDBERRY are some valuable crops both in terms of food (having tasty fruits that can be eaten fresh or used for making jams, pies or wine) and from medical point of view as well, because the high content of vitamins and antioxidants make them be very appreciated as a dietetic product.

The goal of this experiment was to determine the optimal conditions for rooting cuttings listed above.

In the study was taken the commercialized phytohormon for the induction and stimulation of rooting – „Cornevin” a powdered hormonal mixture whose active ingredient is IBA (indole-3-butyric acid). As rooting substrate was used perlite /river sand blend in a ratio of 1 : 1. The rooting was achieved in two variants: in the first the lower tip of the cutting was dusted with hormonal powder (with the concentration of the active ingredient (IBA) of 5gr/kg), after what they were incorporated into the wet substrate.

In the second variant the cuttings were held in „Cornevin” solution (with the concentration of IBA of 5mg/l) for 24 hours after what they were planted in the wet perlite/sand blend and covered with a transparent cover to maintain humidity.

After 1,5 months in the first variant, out of 12 cuttings only 2 specimens of Lincoln Logan and one of Olallie formed small roots of about 3cm and one of Chehalem formed small roots of about 0,5cm from one bud in the upper tip of the cutting. These small roots were incorporated in a small container with peat for future development.

In the variant with the cuttings exposed in the „Cornevin” rooting medium, listing 6 units, Kotata cutting formed 3,0cm roots, Chehalem cultivar formed several 0,5cm roots. Olallie formed roots at the bottom of two buds in the aerial part of the cutting that were incorporated in the peat as well.

The cuttings which root size reached 3,0 cm were planted in pots with substrate soil / peat, 1:1.

The remaining cuttings were aroused with „Cornevin” rooting medium (in the concentration of 5 mg/l of IBA).

And after another 1,5 months 100% of the cuttings of the second variant gave roots. In the first variant only the Chehalem cutting roots had reached the size that permitted to plant it in the pot.

So, the rooting rate in the I variant is 33% and in the II variant is 100%.

TRADIȚII ȘI SARCINI PRIVIND DIVERSIFICAREA SORTIMENTULUI DE CAIS ÎN REPUBLICA MOLDOVA

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Sortimentul de cais se schimbă periodic după cum se schimbă în general și cerințele consumatorului. Primul sortiment recomandat pentru Basarabia (a.1911) s-a bazat pe soiurile Royal (sin. Korolevskii) și Krasnoșciokii (sin Krasnobokii). De notat că calitatea fructelor produse nu se afla la un nivel înalt. Pînă în a. 1918 suprafețele livezilor, precum și sortimentul caisului s-a dezvoltat destul de bine în baza activităților de implementare a pepinieriștilor locali, precum și a prezenței piețelor de desfacere a fructelor din S.Petersburg, Moscova, Kiev, Odessa, Riga, Harkov, Varșava, Lodz. Totuși pînă în anul 1940 sortimentul caisului a variat foarte mult, purtînd un caracter de dezvoltare mai mult amatoresc și domestic. În continuare pentru asigurarea cu materie primă a industriei de procesare formate, pentru o perioadă mai îndelungată au fost pe larg promovate afit soiuri timpurii (Ambrozia, Luizet, biotipuri locale de zarzăr, Esperena), cît și tardive (biotipuri locale de zarzăr, Persicovii, Vengherskii pozdnii, Tabarza). Diversificarea de mai departe a sortimentului de cais (a.1952) a fost datorată omologării soiurilor: timpurii- Alexandr rannii, medii-Vengherskii krupnii, Șalah, Nikitskii, tardive-Krasnoșciokii pozdnii, Crasnii partizan, Spitak, Tiraspoliskii pozdnii, - soiuri preferate afit pentru consum în stare proaspătă, cît și conservate. În a. 1958 în sortiment au intrat un număr relativ redus de soiuri, dar era specificată posibilitatea cultivării în proporții reduse a altor soiuri (de exemplu: biotipuri locale, Vengherskii krupnii, Șalah și Persicovii). Pentru perioada de maturare timpurie a fructelor s-a omologat soiul Luizet, medie – Crasnoșciokii, tardivă – Crasnoșciokii pozdnii și Tabarza. Prin investigațiile efectuate de Institutului de Cercetări pentru Pomicultură s-a evidențiat că soiurile introduse Spitak și Esperena se caracterizau prin rezistență foarte slabă la boli, iar ca urmare, - fructificare instabilă, ceea ce a servit drept bază pentru excluderea lor din lista de bază a sortimentului omologat.

Începînd cu anul 1980 în republică au fost treptat omologate o serie de soiuri, create în Institutul Pomicultură (ICP), astfel, că în următoarele decenii sortimentul omologat era constituit aproape în exclusivitate din soiuri moldovenesti (printre ele: Bucuria, Kișinevskii rannii, Moldavskii olimpieț, Badărk, Nadejda, Detskii, Kostiuenskii, Raduga). Ele cuprind perioada de maturare a fructelor de la jumătatea a doua a lunii iunie pînă în prima decadă a lunii august. Promovarea lor se datorează faptului că ele posedă o plasticitate ecologică largă și o adaptabilitate bună la condițiile microclimatice și pedo-edafice variabile ale reliefului fragmentat al Moldovei. În ultimele 3 decenii în rezultatul studierii a peste 400 soiuri introduse și a peste 3700 de hibrizi, obținuți de la hibridările intravarietale au fost selectate peste 27 selecții, înregistrate 3 soiuri noi cu caracteristici biologice și agronomice performante. Crearea și implementarea în practică a soiurilor noi de cais, corespunzătoare cerințelor moderne de fructe este o sarcină importantă, mai ales avînd în vedere schimbările de climă și apariția piețelor noi. În prezent sunt sofisticate soiuri cu caracteristici valoroase ale fructelor, dar și pretabile la densități mari de plantare și pentru mecanizarea majorității lucrărilor; sistem radicular al portaltoiului adaptabil la diferite tipuri de soluri, în special din cele "grele", specifice pentru multe teritorii ce revin caisului în cazul republicii Moldova; rezistență genetică la ger, fluctuații de temperaturi ștersante din timpul iernii și primăverii; potențial fiziologic de echilibrare a creșterii și fructificării. Însuși fructele trebuie să fie mari (peste 70-90gr.), foarte atractiv colorate, cu pulpa fermă dar succulentă și textura fină, gust echilibrat și aromă specifică de cais, cu sîmbure cît mai mic, detașabil de la pulpă. Pentru fructele destinate procesărilor industriale este indispensabil ca ele să posedă formă și mărime constantă, detașarea bună a pulpei, păstrarea culorii după fierbere, cu conținut ridicat de substanță uscată, zahăr, pectine, macro- și microelemente, etc. Pe lîngă adaptabilitatea și plasticitatea ecologică înaltă, soiurile noi trebuie să fie rezistente la bolile și dăunătorii principali ai speciei, în special la Sharka și cancerul bacterian. Sortimentului de cais actualmente înregistrat pentru înmulțire în țară constă din 17 soiuri, dintre care 9 sunt create la ISPHTA, iar 8 introduse din străinătate. În calitate de soiuri de perspectivă se mai testează temporar în producție încă 10 soiuri moderne, introduse din diferite arealuri de cultivare. Practic toate soiurile sunt pretale atât pentru consum proaspăt, cît și pentru diferite tipuri de procesare separată sau împreună cu alte specii de fructe. S-a lărgit coveierul de caise proaspete: prima decadă a lunii iunie – a doua decadă a lunii august. Tendențele permanente promovarea caisului țin de lărgirea sortimentului cu soiuri de maturare cît mai timpurie și cît mai tardivă cu conservarea calităților performante a fructelor, în special capacitatea de transportare la distanțe lungi.

DEZVOLTAREA EMBRIONILOR IMATURI DE NUC *IN VITRO*.

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În calitate de structuri experimentale au servit embrionii imaturi, segmente de cotiledoane izolate, cotiledoane întregi de nuc (*Juglans regia* L.), precum și axul embrionar împreună cu o parte din cotiledon) obținute la polenizarea liberă a soiului de nuc Chișinău. Mediul bazal de cultură a fost Murashige – Skoog, suplimentat cu zaharoză – 30g/l, agar – 6,5g/l, inozit – 100mg/l. Concentrațiile biostimulatorilor experimentați Moldstim (M) și Ecostim (E) au fost următoarele: 5, 1, 25, 50 și 100 mg/l. Flacoanele experimentale au fost menținute în camera de creștere la fotoperioada de 16 ore, temperatura 24°C, luminozitatea – 2000lk. Rezultatele experimentelor efectuate au demonstrat, că după prima săptămână de cultură „*in vitro*” are loc numai hipertrofia structurilor integrale embrionare, cotiledoanele excizate integral mărindu-se de 1,5 ori față de mărimea inițială la toate concentrațiile experimentale de M.La 25 mg/l M s-a produs și înverzirea cotiledoanelor. Cele mai semnificative schimbări morfogenetice au loc peste 2 săptămâni de cultură „*in vitro*”. S-a depistat că hipertrofia moderată a cotiledoanelor izolate ce se produce la 10 mg/l M are loc concomitent cu începerea dezvoltării sistemului radicular. 25 mg/l M s-a dovedit a fi concentrația, care a favorizat atât hipertrofia puternică, înverzirea cotiledoanelor, cât și dezvoltarea sistemului radicular, constituit din rădăcina principală și a 3-5 rădăcini laterale la cotiledonul cu ax embrionar, sau numai 3-6 rădăcini scurte pentru cotiledoane aparte. S-a evidențiat, că la 50mg/l M rădăcina principală este foarte subțire, iar hipertrofia cotiledonară – slab manifestată. De notat că efectele morfogenetice sunt cu mult mai accentuate în cazul excizării cotiledoanelor împreună cu axul embrionar. La concentrația maximală experimentată – 100 mg/l M a stimulat o hipertrofie diferită la structurile studiate și o înverzire excesivă a lor. S-au depistat zone albe (mozaicitate tisulară) în cadrul embrionului integru. Pe parcursul săptămânii a treia a continuat hipertrofia cotiledonară, precum și formarea rădăcinilor la 5 și stoparea dezvoltării lor la 10mg/l M. La 25 mg/l M continuă dezvoltarea rădăcinii deja hipertrofiate. Concomitent are loc regenerarea intensivă a țesutului calusar. În același timp la 50 mg/l M dezvoltarea rădăcinii principale este moderată, iar fenomenul înverzirii cotiledonare se depistează numai la cotiledoanele excizate împreună cu axul embrionar. La toate concentrațiile experimentate bioreglatorul E a provocat în prima săptămână hipertrofia cotiledonară la ambele structuri studiate. În săptămâna a doua a urmat continuarea hipertrofiei țesuturilor cotiledonare concomitent cu înverzirea epidermei cotiledonare, începând cu 10 mg/l E. Odată cu creșterea concentrației E are loc intensificarea ne întreruptă a înverzirii. Peste 4 săptămâni la 5 mg/l începe dezvoltarea rădăcinii principale, iar la locul “rănirii” țesutului cotiledonar începe eliminarea unui lichid necrotizant. La această etapă ce începe și dezvoltarea plantulei din gemulă în cazul prezenței ei integrale. Ca și la experimentarea M, la concentrația de 25 mg/l E se dezvoltă rădăcini adventive, care la ambele cazuri își i-au începutul din zona structurilor vasculare ale cotiledonului excizat integral. De notat, că cea mai pronunțată hipertrofie cotiledonară s-a observat la 50 mg/l E. De asemenea are loc și hipertrofia excesivă a rădăcinii centrale și a celor laterale. Calusul verzui s-a dovedit a fi și promotorul dezvoltării primilor embrioizi. În cazul M calusul și rădăcinile adventive se dezvoltă în principal la 25 mg/l, în cazul E fenomenele analogice au loc în limitele 25 –100 mg/l. Apariția embriozilor are loc la 25 și 50 mg/l E, și numai la 25 mg/l M. S-a stabilit, de asemenea, că hipertrofia cotiledonară este mai bine exprimată la utilizarea E, comparativ cu M. Astfel se poate concluziona, că ambii bioreglatori sunt efectivi în cultura țesuturilor embrionare zgotice a speciei *Juglans regia* L. Pentru investigațiile ulterioare se pot recomanda concentrațiile 25, 50 mg/l, atât în cazul M, cât și al E. Embriozii obținuți se pot cultiva separat pe medii nutritive optimizate în vederea obținerii de plante noi, care pot reprezenta variații somaclonale ale genotipurilor utilizate în cultura “*in vitro*”.

Cercetările comparative efectuate de noi asupra embriogenezei *in vivo*, ne demonstrează vădit, că de la zigot și până la starea de maturare, embrionul nukului trece prin câteva stadii morfofiziologice bine distincte. Stadiul „embriion globular” se deosebește prin cel mai intensiv metabolism înainte de apariția primordiiilor cotiledoanelor. În același timp maximumul depozitării substanțelor de rezervă coincide cu perioada diferențierii totale, după trecerea endospermului din starea nucleară în cea celulară. Embrionii *Juglans regia* L., este raportat la leucoembrioite în clasificția respectivă. În această ordine de idei e de mirare prezența stomatelor bine dezvoltate în epiderma integumentului (în continuare coaja seminței). După părerea noastră aceste structuri contribuie în mod special la dezvoltarea seminței ca atare, iar în viitor – la germinația embrionului. Considerăm, că pentru cultura *in vitro* a embrionilor imaturi a speciei *Juglans regia* L dezvoltarea stomatelor poate avea o semnificație aparte.

ELECTROPHORETIC ANALYSIS OF PHYCOCYANIN PREPARATIONS PURIFIED BY METHODS OF CHROMATOGRAPHY

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An increased interest in the exploration of algae and cyanobacteria as the sources of bioactive substances of natural origin, replacing drug substances produced by chemical synthesis, allowed the scientific community to consider algae as a resource abundant of substances with beneficial effects on the human body. Among the bioactive substances of medicinal interest produced by *Spirulina* sp. there are the protein-pigments - phycobiliproteins. The phycobiliproteins are used as natural dyes in food industry and as fluorescent agents in immunodiagnosis. A number of investigations have shown their healing properties and pharmaceutical applications. Among phycobiliproteins, phycocyanin plays key role due to its many biological and pharmacological properties [3, 5, 8]. Studies in recent decades have revealed that phycocyanin from spirulina has obvious effect of stimulating hematopoiesis and immunity, as well as anticancer and antitumor activity [7, 9].

C-phycocyanin (C-PC) is the major component of the phycobiliprotein in cyanobacteria *Spirulina platensis*. The fundamental unit of hycocyanins were α and β subunits. The subunits are associated in an (α β) protomers, which in turn can be associated in trimmers (α β)₃ and hexamers (α β)₆ [2, 4]. Phycocyanin absorption maximum is in the range of 615-620 nm (blue intense color). Phycocyanin purity is measured by the ratio A620 / A280. Depending on the obtained values of this ratio, preparations of phycocyanin may be considered of food grade (0,7), as chemical reagent (3,9) and analytical grade (>4,0).

Over the past two decades, various methods have been developed for the purification of phycocyanin: density gradient centrifugation, ammonium sulfate precipitation, chromatographic techniques, isoelectric precipitation, aqueous 2-phase extraction etc. [1, 6, 10, 11]. However there are some unsolved issues relating to the duration of the phycocyanin purification process and purity evaluation of obtained phycocyanin preparations.

The aim of this work was the selection of chromatographic methods of purification and electrophoretic analysis of obtained phycocyanin preparations.

Purification of phycocyanin. Phycocyanin was extracted by bidistilled water from biomass of *Spirulina platensis*, previously subjected to destruction of cell walls by repeated freezing-thawing. Bidistilled water (4 volumes) was added to 1 volume of spirulina suspension (20 mg/ml) and after stirring at 4 °C within 30 min, the sample was subjected to centrifugation at 6000 rpm.

Phycocyanin extract was fractionated with (NH₄)₂SO₄ in 2 stages: first stage, up to 25% saturation, after which the precipitate was removed, and, second stage, the supernatant was subjected to saturation of (NH₄)₂SO₄ to 50%. The precipitated phycocyanin was separated by centrifugation at 6000 rpm and dissolved in 3M NaCl, pH 8,0, Tris-HCl 0,05M.

The following phycocyanin purification was performed on phenyl-sepharose column (\varnothing = 1,7 cm, h = 5 cm) equilibrated with 3M NaCl, Tris-HCl 0,05M pH 8,0, elution rate 50ml/h. The successive stepwise elution was carried out with 1, 0,5, 0,25, 0,1 M NaCl, respectively, and finally, with H₂O. The first bit received on elution with 1 M NaCl (Fig.1 lane 2) was diluted with H₂O and supposed to chromatography on DEAE-Sepharose, equilibrated with 0,05 M sodium phosphate buffer, pH 7,5. Phycocyanin purified fraction was obtained by elution with 0,25 M NaCl, sodium phosphate buffer pH 7,5. Detection and recording of protein fractions eluted from the column was performed at 280nm (Uvicord UV-1, Pharmacia).

ANTIOXIDANT ACTIVITY OF ETHANOL EXTRACTS FROM NOSTOC LINCKIA BIOMASS

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The multicomponent antioxidant mixture obtained by extraction with ethanol (96%) from *Nostoc linckia* biomass was tested on storage conditions under +4 °C for 90 days. The stability of the extract was periodically checked applying the ABTS (2,2 Azinobis 3-sulfonic acid ethylbenzothiazoline-6) assay.

The monitoring of the antioxidant activity of 96% ethanol extract showed duration of preservation limited to 30 days (Figure 1). The antioxidant extract lost in the first 24 hours 13% of the ability to reduce the ABTS radical that was recovered through the next 24 hours. Overall, the antioxidant activity of the tested extract increases by 30% during the first 30 days. In the next 60 days the antioxidant test results decrease by 28%.

The importance of long-term preservation of antioxidant activity of multicomponent microalgal extracts is widely recognized. The supplementation of antioxidant preparations with synthetic antioxidants such as tocopherol, Trolox and ascorbic acid is proposed in order to strengthen the antioxidant effect and to minimize oxidation processes. Therefore 96% ethanolic extract was supplemented with tocopherol. In order to avoid the duplication of antioxidant effect, the concentrations of 1, 10 and 100 µg/ml of antioxidant supplements were used. The preparations have been preserved under similar conditions for 3 months: +4°C in smoked glass vessels.

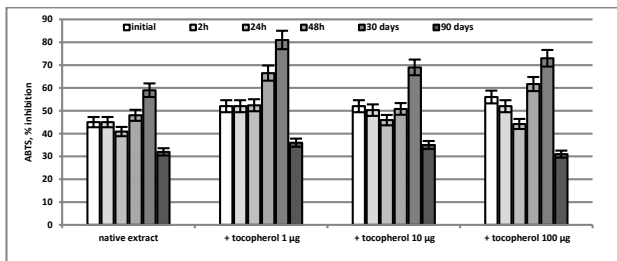


Figure 1. Changes of antioxidant activity of 96% ethanol extract of *Nostoc linckia*, supplemented with tocopherol, during the storage at +4°C

The antioxidant activity increased by 15% in extracts supplemented with 1 µg/ml and 10 µg/ml tocopherol and by 24% in the case of 100 µg/ml tocopherol supplementation. During the first 24 hours of storage, the antioxidant activity of the ethanol extract containing 1 µg/ml tocopherol remained unchanged. In the case of other extracts was recorded a reduced antioxidant activity. The next 24 hours some changes of antioxidant activity were noted: the antioxidant activity returned to the initial values in the extract supplemented with 10 µg/ml tocopherol, increased by 28% and 10% in the extracts supplemented with 1 µg/ml and 100 µg/ml tocopherol respectively. By the end of 30-day period the antioxidant activity of the extracts increased, with the completion of the stabilization process. The antioxidant activity values, determined by ABTS assay, increased by 30-33% in the native extract and the extracts supplemented with 10 µg/ml and 100 µg/ml tocopherol; and by 56% in the extract supplemented with 1 µg/ml tocopherol. After three months of storage, the tested extracts of *Nostoc* biomass have lost the ability to reduce ABTS radical.

The results of this study reveal the existence of a stabilization period of both the native ethanol extract and the extracts supplemented with tocopherol.

APPLICATION OF THE BIOLOGICAL PREPARATION GLIOCLADIN-SC ON SUNFLOWER

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From variety of biological products for plant protection multifunctional products with a broad spectrum of fungitoxic and fungistatic actions, growth stimulating effects and immunostimulation activities in plant have the particular value. The biological products on the base of the fungi genus *Trichoderma* Pers. ex Fr. are some of them. Their biological efficacy is based on the living cells of microorganisms and their metabolic products.

There are various methods of application of biological products on the base of fungi *Trichoderma*, such as addition into soil in dry form, treatment of seed by liquid solutions, soil drench, drip irrigation, use in greenhouses. The result is: reducing the phytopathogens, plant-growth promotion and development of the root-system architecture, resilience to diseases and finally increase of productivity. Application of biological products in the plant protection system is especially important for Republic of Moldova, where agricultural production is a priority.

The aim of our research was to determine the influence of the liquid form of the biological preparation Gliocladin-SC on some growth parameters and inhibition the growth of the root and basal rots of sunflower, caused by a pathogen *Sclerotinia sclerotiorum* and fungi of the genus *Fusarium*.

Bioproduct Gliocladin-SC – microbial fungicide, created in our institute on the base of the fungus *Trichoderma virens* Miller, Giddens and Foster, strain 3X.

Gliocladin-SC was tested under field conditions during 2014. The trial was conducted on sunflower variety Don- Macrocarpa. Gliocladin-SC was applied as 5% - aqueous suspension for pre-treatment of seed before sowing. Seed soaking was done by placing seeds into warm water for 24 hours. Control – untreated seeds. As a standard chemical fungicide *Royal-flo42SL* was used.

At the final part of the study it was found that the germination of seeds while using a biological product Gliocladin-SC was 11,5% higher, and the number of productive plants was 20,6% higher than one in the control.

The significant difference between treatment and control groups in the middle baskets diameter was established during these trials. Under treatment conditions the middle baskets diameter was 15,2% greater than one in the control. While the number of small baskets (diameter -11-15 cm) was 18,0% less, and the one of the large baskets (diameter - more than 21 cm) was 20,2% greater in comparison with the control. Mass of the seeds from one basket increased by 49,2% and the mass of 1000 seeds increased by 9,0% in comparison with control (Table).

Table. Indicators of the development of sunflower by the using the biological product Gliocladin-SC

Variant	Number of productive plants /ha	baskets diameter, cm	Mass of seeds from 1 basket, kg	Mass of 1000 seeds, gr
Control	29 101	17,1	0,063	70,19
Etalon <i>Royal-flo 42SL</i>	25 597	18,6	0,075	71,92
Gliocladin-SC	35 107	19,7	0,094	76,54
LSD _{0,05}	2 717	1,5	0,011	1,85

Biological efficiency of a biological product Gliocladin-SC in reducing the development of the sunflower's basket forms of white rot was 82,4%, in reducing the development of root rot – 64,7%.

The testing of effectiveness of biological product Gliocladin-SC showed that the preparation reduces the development of basal and root rot, enhances germination and number of productive plants, has a stimulating effect on the formation of the size of the baskets and seed weight.

**PARTICULARITIES OF LONG-TERM STORAGE OF RARE SPECIES
UNDER THE *IN VITRO* CULTURE CONDITIONS**

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To meet the challenges of preserving and restoring the gene pool of rare and endangered plant species, the *in vitro* culture method is widely applied nowadays. *In vitro* collections can be stored in a state of active or slow growth for a long time. However, constantly transplanting *in vitro* plants is a rather laborious task. Therefore, the method of long-term non-transplanting storage of test-tube plants is of great importance in creating a collection of clonal *in vitro* plants. Deposition reduces the time and labor spent, and allows lowering the reagent costs. The techniques that provide for slow growth are based upon temperature and light reduction, and the medium modification, especially by adding osmotics or retardants. For different cultures, special modifications to these procedures are applied; hence, finding the ways to create favorable conditions for long-term non-transplanting storage of test-tube plants is an urgent problem for each species to be cultivated. Our research is focused upon the species listed in the Red Book, such as *Fritillaria montana*, *Bellevia sarmatica*, and *Lilium martagon*, a highly decorative rare species. We have previously found the best conditions possible for introducing these species in the *in vitro* conditions, and optimized the microclonal reproduction system depending on the mineral and hormonal factors of the growth-supporting nutrient medium. After obtaining a sufficient quantity of regenerates, the next stage of our research in developing a system for conservation of rare species has been focused upon finding the optimal conditions for making long-term non-transplanting storage of plants efficient. When developing a storage system, we have used the available opportunities as a starting point and held a series of experiments to demonstrate the efficiency of using sucrose, which is an available and inexpensive reagent, for deterring plant growth. For each of the species, optimal conditions that allow storing the cultures for 12 months without replanting them, have been determined. A high percentage of test-tube plant viability (84 – 93%) has been achieved by cultivating microbulbs in the basic environment according to MS (Murashige and Skoog, 1962), supplemented with different concentrations of sucrose, for the following:

- *Lilium martagon* – 100 g / l;
- *Fritillaria montana* - 90 g / l;
- *Bellevia sarmatica* - 90 g / l.

The plants have been stored in the culture room (at 20 °C) in the dark.

THE INFLUENCE OF THE COMPOSITION OF NUTRIENT MEDIUM ON THE NUMBER OF BACTERIAL CELLS IN THE PROCESS OF CULTIVATION UNDER LABORATORY CONDITIONS

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The revealing of antibiotic properties of the microorganisms is affected by many factors. Microorganisms isolated from natural habitats and then cultivated in the laboratory, as a rule, are not inherent condition of existence. In the laboratories, the majorities of microorganisms are maintained and studied as a pure culture. In this state, the microbes have never met in nature. The culturing of the organisms in pure cultures excludes possibility the influence on them other microorganisms, the favorable or, on the contrary, the adverse effects of metabolites of other organisms are not showing.

Among the most significant factors that influence the manifestation of the antibiotic properties of the microorganisms isolated from natural sources is the composition of the medium, active acidity, temperature, methods of joint cultivation of two or more species of microorganisms, etc

In the time of identification the potential possibility of microorganisms to produce antibiotics the selection of the media should be given serious attention.

In this regard, one of the primary tasks for cultivation of bacteria cultures in the laboratory with a sufficiently high titer is the selection of an optimal nutrient medium.

Materials and methods. In the experiment were four strain used bacteria of the genus *Bacillus* and four culture media (PD - potato dextrose medium - control, natural medium of uncertain composition; medium-1 and -3 - synthetic medium; medium-2 - mixed composition).

The temperature of the incubation was of 28-30 °C. The results of the experience of the medium PD and medium-2 showed the best result for all four bacterial cultures. On the medium-2 after 2 days growth, the bacterial culture showed that the titer of bacterial cells reaches the optimal number and 2 days is plenty of time for the cultivation of the present bioagents. For *B. subtilis*-S4 medium of all compositions were the least appropriate, although quantity of bacterial cells increased on the third day on medium-2 and -3. It can be assumed that further cultivation can increase of number cells, but 4 days and more of this process for the bacterial agents is no rational. It shows once more that for each microorganism cultivation conditions must be chosen individually. From the data obtained, it can be assumed that the accumulation of biomass under these conditions for some the studied bacterial cultures were suspended (table.1, 2). It is because the organisms used the main components of the medium, and the medium were enriching with a number of metabolites that can lead to changes in the inhibitory process of biomass accumulation.

Table 1. The number of bacteria after 2 days of culturing, cells/ml

Bacterial culture	Medium PD	Medium № 1	Medium № 2	Medium № 3
<i>B. subtilis</i> L	$1,6 \times 10^8$	$3,4 \times 10^7$	$1,3 \times 10^9$	$1,7 \times 10^7$
<i>B. subtilis</i> S2	$2,1 \times 10^9$	$1,1 \times 10^6$	$1,7 \times 10^9$	$1,1 \times 10^9$
<i>B. subtilis</i> S4	$1,0 \times 10^9$	$2,4 \times 10^6$	$7,0 \times 10^7$	$3,7 \times 10^7$
<i>B. subtilis</i> Fn	$7,2 \times 10^8$	$1,0 \times 10^7$	$6,6 \times 10^9$	$7,3 \times 10^6$

Table 2. The number of bacteria after 3 days of culturing, cells/ml

Bacterial culture	Medium PD	Medium № 1	Medium № 2	Medium № 3
<i>B. subtilis</i> L	$1,1 \times 10^9$	$2,6 \times 10^8$	$2,6 \times 10^9$	$1,3 \times 10^7$
<i>B. subtilis</i> S2	$1,8 \times 10^{10}$	$7,0 \times 10^6$	$7,1 \times 10^9$	$1,3 \times 10^9$
<i>B. subtilis</i> S4	$3,3 \times 10^9$	$5,2 \times 10^6$	$2,3 \times 10^8$	$3,2 \times 10^8$
<i>B. subtilis</i> Fn	$1,2 \times 10^9$	$1,6 \times 10^8$	$1,4 \times 10^{10}$	$1,7 \times 10^7$

Conclusions. Culture medium-2 of definite composition for the cultivation of *B. subtilis*-L, *B. subtilis*-S4 и *B. subtilis*-Fn allows obtain with a high yield of viable cells during the 48 hours of cultivation.

PROTEINELE DE REZERVĂ ȘI CALITATEA SEMINTELOR DE GRÎU COMUN DE TOAMNĂ ȘI TRITICALE

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Caracterele de bază ale culturilor cerealiere sunt productivitatea recoltei, rezistența plantelor la diferiți factori climatici de stres (temperaturi joase negative, secetă, maladii ș.a.) și proprietățile bio-chimico-tehnologice care formează calitatea nutritivă și de panificație ale boabelor.

Calitatea biologică, nutritivă depinde de cantitatea și componentele proteinelor de rezervă totale când prevalează proteinele ușor solubile (albuminele, globulinele) cu conținut mai ridicat de aminoacizi esențiali, prezența vitaminelor grupei B, ale microelementelor etc. Calitatea de panificație a făinii semintelor maturizate de cerealiere este influențată de conținutul cantitativ al glutenului și particularitățile lui fizico-chimice care reflectă calitatea glutenului determinată de coraportul componentelor proteinice gliadine:glutenine. Rolul de bază la formarea cantității și calității glutenului le aparține gluteninelor. Pe lângă proteine în componența glutenului sunt prezente pentozanele și lipidele. Toți acești compuși în aluat formează un complex prin legăturile covalente, disulfidice, hidrofobice și hidrogenice.

În lucrare sunt prezentate rezultatele studierii variabilității genetice al conținutului cantitativ a proteinei de rezervă, celor glutenice și al glutenului umed din făina semintelor de grâu comun de toamnă: *Moldova 5*, *Moldova 11*, *Moldova 55*, *Moldova 77*, *Moldova 79*, *Moldova 99* și soiul *Cuialnic* – standard, de triticale: *Ingen 33*, *Ingen 35*, *Ingen 40* și *Ingen 93* și secară: *Zimbreni 70* și *Tetram* - toate create în Institutul de Genetică, Fiziologie și Protecție a plantelor recolta anilor 2011 – 2014. În făina fin măcinată cantitatea proteinei totale a fost determinată cu metoda biuret, glutenul umed – cu metoda clasică: spălarea aluatului cu apă salină până la eliminarea completă al amidonului și tărfței. Conținutul proteinelor glutenice sumare s-a calculat prin uscarea glutenului umed până la masa constantă.

În făina boabelor împlinite și maturizate în condiții climatice relativ normale (aa. 2013, 2014) ale soiurilor de grâu comun de toamnă *Moldova 11* și *Moldova 79* proteina de rezervă totală constituie 12,2 – 12,7% și este la nivelul standardului *Cuialnic*, la celelalte soiuri ea prevalează față de standard cu 1,5 – 2,6%. La soiurile obținute în ultimii ani *Moldova 55* și *Moldova 99* acest indice constituie 14,4% și 14,0% respectiv. În boabele soiurilor de secară *Zimbreni 70* și *Tetram* proteina totală variază de la 10,0% la 13,3%, în făina celor de triticale *Ingen 35*, *Ingen 40*, *Ingen 93* și 2 forme noi *CCSN1* și *CCSN6* proteina de rezervă totală este la nivelul grâului comun de toamnă și constituie 11,9% - 13,5% cu excepția soiului *Ingen 33* recolta a. 2014 unde ea este numai de 9,8%.

Condițiile de stres hidric (seceta pronunțată) din anul 2012 în perioada de formare și maturizare a semintelor activează semnificativ acumularea proteinelor de rezervă în ele la toate soiurile de grâu, secară și triticale cu 1,3% - 3,5%. Un alt indice al calității de panificare este cantitatea și calitatea glutenului umed din boabe. Rezultatele studierii nivelului cantitativ al glutenului umed din făina boabelor recoltelor aa. 2011 – 2013 indică variabilitatea genetică și fiziologică al acestui indice. Glutenul umed în boabele soiurilor grâu comun de toamnă maturizate în condiții climatice normale - anii 2011, 2013 constituie de la 27,6% la *Moldova 5* până la 32,0% - *Moldova 77*, la triticale el variază între 21,5% la *Ingen 40* și 26,3% la *Ingen 35*. Cantitatea proteinelor glutenice sumare este de 10,2% - 11,6% la soiurile de grâu și diferă la triticale între 7,5% - *Ingen 40* și 9,1% - *Ingen 35*. Acestea constituie până la 85% din cantitatea proteinei de rezervă totale în semintele cerealiere.

Stresul hidric și temperaturile caniculare majorează acumularea proteinelor glutenice în boabele de grâu cu 1,0 – 3,0%, iar în cele de triticale cu 2,5%. Un caracter important al glutenului umed și de panificație a făinii este și capacitatea de hidratare al glutenului care variază de la 63,1% până la 65,7% în glutenul recoltei a. 2013 și 59,1 - 62,2% la glutenul boabelor a. 2012

Rezultatele obținute reflectă dependența conținutului cantitativ al proteinelor de rezervă totale, celor glutenice și glutenului umed de particularitățile genetice ale genotipului și condițiile de creștere ale plantelor.

Datele experimentale privind unii indici biochimici ai calității de panificare la soiurile de cerealiere spicoase studiate pot fi utilizate în lucrările de ameliorare.

PLANT RESOURCES FOR PRODUCING RENEWABLE ENERGY UNDER THE CONDITIONS OF THE REPUBLIC OF MOLDOVA

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The XXI century can be regarded as an era of climate change in which concerns about climate are accompanied by other drivers relating to globalization, food security, energy and sustainability issues. Energy is the dominant factor that determines the welfare of the country and people influences the level of development of all spheres of activity in society. The complex problems of the development of renewable energy have become a global political dimension.

The Republic of Moldova has few fossil energy resources, so being forced to import near 95%, depends entirely on the supplying countries. Therefore, the issue of renewable energy sources has been and remains actuality. Moldova has taken the first steps in this field in 2007 when it was approved the Renewable Energy Law, in 2013 – the Energy Strategy by 2030, implemented the project “Moldova: Energy and Biomass”, financed by the European Union and UNDP Moldova. Plant species are efficient users of solar energy for converting CO₂ into biomass. The structure expected by 2020 of the total production and consumption of energy obtained from renewable sources based on biomass will constitute approximately 70,0%.

Over more than half a century, as a result of the introduction and acclimatization researches done in the Botanical Garden (Institute) of the ASM, collections and exhibitions of plants with multiple use, necessary for the development of the national economy, were founded. The investigation of local as well as introduced tall perennial species for renewable energy production is an important object. Currently, about 100 species of plants from the Botanical Garden can be used to produce different types of biofuels.

Agro biological peculiarities and energy characteristics of biomass provide perspectives for implementation in the Republic of Moldova of the herbaceous species: *Macleaya cordata* (Willd.) R. Brown fam. *Papaveraceae*, hybrid *Miscanthus giganteus* (M. sinensis Andersson and M. sacchariflorus (Maxim.) Franch.) fam. *Poaceae*, *Polygonum sachalinense* Fr. Schmidt fam. *Polygonaceae* natives of eastern Asia; *Sida hermaphrodita* fam. *Malvaceae*, *Helianthus tuberosus* L., *Symphotrichum novi-belgii* (L.) G.L.Nesom and *Silphium perfoliatum* L. fam. *Asteraceae* natives of North America; *Sorghum alnum* Parodi natives of South America; *Galega orientalis* Lam. and *Astragalus galegiformis* L. fam. *Fabaceae* natives of Caucasus with 10-20 years longevity. Cultivation and harvesting of these species do not need sophisticated mechanisms and specific equipment as in forest exploitations. The areal biomass of these species can be harvested with technical means of fodder collection.

The bulk density of harvested dry biomass of these species varies from 118 to 288 kg/m³ and is used to produce briquettes and pellets. The gross calorific value of the dry matter reaches 18,3-20,0 MJ/kg. The ash content is 1,5 – 3,7 %. Specific density of the briquettes is 720-1162 kg/m³. The potential of energy production is 190-470 GJ/ ha/year, equivalent to 6,5-16,2 t of brown coal or 4,6-11,4 t conventional oil.

The fresh aerial biomass and silos of the species *Silphium perfoliatum*, *Sida hermaphrodita*, *Helianthus tuberosus*, *Polygonum sachalinense*, *Sorghum alnum*, *Galega orientalis* and *Astragalus galegiformis* are used as substrate for obtaining biogas (295- 534 m³/t dry matters), with 52-70 % methane content. The resulted solid digestate from the fermentation is one of the most valuable sources of soil fertility restoration.

By individual selection of the introduced population, were created the local varieties: *Speranta Galega orientalis*, *Gigant Polygonum sachalinense*, *Vital Silphium perfoliatum*, *Energio Sida hermaphrodita*, *Solar Helianthus tuberosus* and registered in the Catalogue of plant varieties of the Republic of Moldova, the yield reached 80 - 142 t/ ha/year of green mass.

The establishment of energy plantations, using also the forms and varieties of plants created in the Botanical Garden (Institute) of the ASM, may serve as a valuable energy source for Moldova that will help to strengthen the national security in terms of reducing the use of imported energy and ameliorating the ecological situation, would improve the balance of payments, would make possible to exploit degraded and unused agricultural land by growing energy crops, would help to create new jobs and increase the labour productivity in agriculture, would lead to the gradual transformation of agriculture into an efficient producer of renewable energy sources.

ACȚIUNEA UNDELOR MILIMETRICE ASUPRA PRODUCTIVITĂȚII ȘI CONȚINUTULUI DE CARBOHIDRAȚI LA LEVURILE *SACCHAROMYCES CEREVISIAE*

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La efectele biologice importante ale undelor milimetrice, cu referință la biotehnologie, poate fi menționată acțiunea asupra biosintezei principiilor bioactive în celulă. În special, în cazul acțiunii undelor milimetrice, un rol important îi revine membranei celulare. Este cunoscut că în membrana celulară a levurii se conțin mai multe tipuri de carbohidrați, de bază fiind glucanii și mananul legat cu proteinele învelișului celular. În cadrul cercetărilor efectuate anterior s-a constatat că undele milimetrice cu frecvență extra înaltă pot fi utilizate ca unul din factorii de reglare a multiplicării levurilor [1,2].

Până în prezent nu este elaborată o ipoteză unică a acțiunii undelor milimetrice asupra sistemelor biologice și nici a metodelor de evaluare a diferitor procedee de dirijare cu astfel de sisteme. Este evidentă importanța cercetărilor legate de aplicarea undelor milimetrice în scopuri practice.

Scopul investigațiilor a constituit determinarea efectelor undelor milimetrice cu frecvență extra înaltă asupra productivității și conținutului de carbohidrați la tulpinile de levuri brevetate ca producători performanți de β -glucani (*Saccharomyces cerevisiae* CNMN-Y-20) și manani (*Saccharomyces cerevisiae* CNMN-Y-18).

Ca generator de unde milimetrice a fost utilizat dispozitivul KB4-ДД, RS-232, cu lungimile de undă $\lambda = 4,9, 5,6, 7,1$ mm, ceea ce corespunde frecvențelor $f = 60,12$ GHz, 53,33 GHz și 42,19 GHz, (maxim 10 mW/cm^2), model 2007 (Fed. Rusă). Aparatul este certificat și permis spre utilizare în practica medicală. Cultivarea levurilor în profunzime s-a realizat pe mediul YPD.

Analiza influenței undelor milimetrice asupra producției de biomasă la tulpina *Saccharomyces cerevisiae* CNMN-Y-20 a arătat că iradierea culturii cu frecvențele menționate, emise timp de 10, 20, 30 minute, practic nu modifică conținutul de biomasă acumulată timp de 120 ore la cultivarea în profunzime. Unele mici devieri a rezultatelor obținute la tulpina iradiată cu frecvența $f = 60,12$ GHz (expoziție 10 minute) sau cu frecvența 42,19 GHz (expoziție 30 minute) nu sunt semnificative, acestea se află în limitele marjei de eroare.

În cazul investigațiilor influenței undelor milimetrice asupra dezvoltării tulpinii *Saccharomyces cerevisiae* CNMN-Y-18 s-a constatat că favorabilă este frecvența 53,33 GHz. La iradierea timp de 10 minute s-a înregistrat o sporire a conținutului de biomasă cu 17,8%, iar la iradierea cu durata de 20 minute producția de biomasă a crescut cu 22,4 %, comparativ cu proba martor. Frecvențele examinate, aplicate timp de 30 minute, modifică nesemnificativ conținutul de biomasă levuriană.

În cadrul investigațiilor de determinare a influenței undelor milimetrice asupra biosintezei carbohidraților s-au constatat deosebiri veridice între martor și variantele experimentale. Astfel, efect evident de stimulare a conținutului de carbohidrați (16,7% respectiv 43,5%) s-a stabilit la iradierea tulpinii *Saccharomyces cerevisiae* CNMN-Y-20 cu frecvențele $f = 60,12$ GHz și $f = 53,33$ GHz, expunerea 20 minute. O creștere semnificativă a conținutului de carbohidrați totali (cu 29,1...35,8%) la tulpina *Saccharomyces cerevisiae* CNMN-Y-18, se produce la acțiunea undelor milimetrice cu frecvența 53,33 GHz, aplicate timp de 20 - 30 minute.

Prin urmare, studiul a stabilit posibilitatea utilizării undelor milimetrice cu frecvență extra înaltă în biotehnologia cultivării levurilor *Saccharomyces cerevisiae* în vederea amplificării proceselor metabolice, implicit al producției de biomasă și conținutului de carbohidrați.

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INCREASING OF VINE RESISTANCE TO UNFAVORABLE GROWING CONDITIONS BY USING OF TRACE ELEMENTS AND MICROORGANISMS COMPOSITIONS

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The vineyards of Moldova constitute a major factor in its agricultural productivity. However, long-term plant cultivation imposed by a monoculture such as the vineyards leads to exhaustion of the soil, reduction of microbiological activity in rhizosphere, and accumulation of products from pesticides, such as copper and herbicides. The biomass of microorganisms in the soils under the vineyard is reduced 3- to 4- fold, because of chemical applications and nutrients available to the plants become exhausted. The result of these stresses is reduced productivity, less resilience of plants to pathogens, and lowered quality of the grapes and wine. Development of new efficient biofriendly techniques of agricultural management is paramount object for integrated sustainable agriculture.

The possibility to elaborate a new advanced technology to maintain vineyard health under sustainable conditions and to increase plantation longevity was undertaken in last 4 years. The novel approach is to capitalize on boosting plant performance using a mixture of beneficial microbes combined with a complex of nutritive elements. The goal is to reduce the chemical load on the ecosystem and to correct plant growth in the soils already suffering from heavy metals and pesticide residues. A trace elements complex Microcom-V, developed by the authors of the article, is shown to optimize grape cultivation. The beneficial effects of the nutrients are supplemented with suspensions of living beneficial microorganisms and their metabolites to create a new complex for plant productivity. Such approach have to possess a wide range of action, be ecological friendly, compatible with the standard technology of grape cultivation and offer cost-saving to be competitive on the world market.

The experimental results obtained during 2011-2013 confirmed the hypothesis that became the basis for the researches - the ability to use trace elements and some strains of microorganisms to improve growth of plant growing under unfavorable environment conditions.

It was highlighted for the first time the possibility of common use of trace elements and saprophytic microorganisms to enhance the growth and development of vine cuttings. Incorporation into the soil of microorganism's suspension and plants fertilization by micronutrients and microbial metabolites in the growing season has a positive effect on metabolic processes and plant growth. Using microorganisms is followed by quality of the material of planting increase and of trace elements in complex Microcom-V reduction. It was established for the first time beneficial effect of saprophytic microorganisms on vine seedlings and fruitful plants growth under unfavorable conditions (copper excess, low temperature, application of herbicides); ability to decrease by 50% the dose of herbicide Raundap adding consortium of 3 microorganisms with the same effect on weed control, maintenance of nitrification activity in soil and plant nutrient status to a higher level.

The optimization of the metabolic processes in the course of plant vegetation through application of microelements and metabolites of microorganisms has influenced shoot growth and maturation, which resulted in the enhancement of plant resistance to frost during the period of dormancy. The evident increase in the degree of annual wood tissues maturation of the treated plants is associated with the intensification of reserve substances accumulation, especially protective ones. It contributes to the formation and a fuller manifestation of genetically based potential of frost and winter resistance of vine, increase of plant productivity and longevity.

The obtained data demonstrate the possibility of developing an effective process for the control of growing conditions and plant nutrition on the soils contaminated with Cu and pesticides, for productivity and resistance to pollution and low temperatures increase, improve product quality, increase the longevity of the vine, reducing chemical pressing on the environment. A consequence of abovementioned is increase in quality and quantity of seedlings, longevity of vineyards and profitability of viticulture.

RECENT ACHIEVEMENTS IN MICROBIOLOGICAL PLANT PROTECTION

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Based on the severity of the phytosanitary issues caused by the action of pests on the background of worsening ecological situation resulting from the application of pesticides to combat them, now the need to develop alternative means of plant protection increases, among which a more important one being the biological products made of various useful microorganisms. Microbiological protection recorded remarkable results in controlling various pests (pathogenic agents of diseases, insect and mite pests, and weeds), permanently extending the range of the useful agents that are used, as well as the spectrum of the protected crops. There is a growing awareness that microbial pesticides are inherently different from chemical pesticides, with fundamentally different modes of action and that they should therefore be assessed on their own merits and problems and data requirements should be set accordingly. However, the basis for the proposed data requirements needs improved scientific justification.

The scientists of the Institute of Genetics, Physiology and Plant Protection of Academy of Sciences of Moldova, by isolating, identifying and determining biological particularities of various useful microorganisms (viruses, bacteria and fungus) have developed original technological procedures of production and application and submitted for approval some biological preparations effective in controlling pests with the most severe impact on crops. These were submitted for approval or extension of the scope of use of National Council for Approval of Products for Phytosanitary Usage and Fertilizers.

Baculoviral preparation Virin-HSP was elaborated to fight *Helicoverpa armigera* which in recent years recorded expanding the area of spreading, as well as the spectrum of attacked crops. The product is made on the basis of nuclear polyhedrosis virus with a high degree of specificity of the insect host and has titer of 6 billion polyhedra/g in the form of paste. The preparation has specific action on insects and causes noctuid epizootic phenomena acting on subsequent generations and protecting crops. In the Republic of Moldova it is recommended to protect vegetables (tomatoes, peppers), technical crops (sugar beet, sunflower, and tobacco), and cereals (corn), decorative and medicinal plants. Consumption norm – 0.2 kg/ha.

Paurin - contact bactericide obtained under bacterium *Pseudomonas fluorescens* BKM CP 330 D expected to combat the pathogen *Agrobacterium tumefaciens* Sm. fnd Town. in orchards and grapes, as well as root rots in vegetable crops, soybeans (*Fusarium gibbosum*, *Rhizoctonia solani*, *Pythium debaryanum*, *Alternaria sp.*, *Penicillium sp.*, *Aspergillus sp.*) and in potato (*Fusarium solani*, *Pectobacterium carotovorum*).

Trichodermin Th-7F SC - fungicide constituted under *Trichoderma harzianum* strain Th-7F (CNMN F-16) expected to combat pathogens in vegetable crops (*Rhizoctonia solani* Kuehn, *Botrytis cinerea* Pers, *Sclerotinia sclerotiorum* de Bary, *Myrothecium verrucaria*, *Ascochyta cucumis* (melonis) Fautr. Et Roum, *Colletotrichum lagenarium* E. et H., *Fusarium spp.*, *Streptomyces*, *Pythium debaryanum* Hesse); decorative (*Rhizoctonia solani* Kuehn, *Botrytis cinerea* Pers, *Sclerotinia sclerotiorum* de Bary; *Fusarium spp.*, *Verticillium dahliae* Kleb.); tobacco (*Fusarium spp.*, *Verticillium dahliae* Kleb., *Thielaviopsis basicola* Ferr, *Pythium debaryanum* Hesse, *Botrytis cinerea* Pers); grape-vine (*Botrytis cinerea*) Pers.

Trichodermin SC proposed as liquid fungicide constituted under *Trichoderma lignorum* strain M-10 expected to fight pathogens in sunflower (*Sclerotinia sclerotiorum*), soybean (*Fusarium spp.*), grape-vine (*Botrytis cinerea*), and ensuring high biological, economic and ecological effectiveness. The preparations enhance also the biological indicators of crop development.

Gliocladin SC - fungicide obtained on the basis of the active substance of *Trichoderma virens* strain 3X, expected to fight white rot in sunflower (*Sclerotinia sclerotiorum*), soybean (*Fusarium spp.*), grape-vine (*Botrytis cinerea*). The preparation ensuring high biological, economic and ecological effectiveness.

The pace of biopesticide research is increasing. From the beginning of 2006 through early April 2015, 4 biological preparations had been registered with National Council for Approval of Products for Phytosanitary Usage and Fertilizers. Many of these are agriculture related and registered for use as such. The value of microbiological means of protection developed by the scientists of the institute does not consist just in their considerable biological, environmental and economic effects, but also their possibility to be included in conventional and organic farming systems.

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