#### АНОТАЦИИ НА МАТЕРИАЛИТЕ

## по чл. 76. от Правилника за РАС на ПУ"Паисий Хилендарски", включително САМООЦЕНКА НА ПРИНОСИТЕ

на доц. д-р Теодора Атанасова Стайкова,

предоставени за рецензиране по настоящия конкурс за заемане на академичната длъжност "професор" по област на висше образование 4.Природни науки, математика и информатика, професионално направление 4.3.Биологически науки (Генетика – Обща и популационна генетика)

#### I. Публикации в реферирани списания с IF

1. **Staykova T.** 2008. Genetically-determined polymorphism of nonspecific esterases and phosphoglucomutase in eight introduced breeds of the silkworm, *Bombyx mori*, raised in Bulgaria. 8pp. *Journal of Insect Science* 8:18, available online: insectscience.org/8.18. (IF 0.963) (цитирана в 17 публикации, 7 от които в списания с IF/SJR)

Isoenzymes are very suitable markers for the study of the inter-breed diversity of the silkworm *Bombyx mori* L. (Lepidoptera: Bombicidae). More than 250 breeds have been raised in Bulgaria, which are not very well studied with regard to the isoenzymic polymorphism. Polymorphism of nonspecific esterases from pupal haemolymph was analyzed, as well as of phosphoglucomutase from different organs of larvae, pupae and imago, from eight introduced breeds. Electrophoresis in polyacrylamide gels was used. A polylocus control of nonspecific esterases and possible monolocus control of phosphoglucomutase was ascertained. Biallele and triallele polymorphism of phosphoglucomutase locus and in three of the esterase loci was determined. The allelic frequencies of the polymorphic loci in each breed were analyzed. Inter-breed differences were found in different allelic frequencies, different heterozygosity and polymorphism.

2. **Staykova T**., Ivanova E., Grekov, D., Avramova K. 2012. Genetic variability in silkworm (*Bombyx mori* L.) strains with different origin. *Acta Zoologica Bulgarica*, Suppl. 4, 87-92. (IF 0.309) (цитирана в 5 публикации, 1 от които в списание с SJR)

Genetic diversity within and among ten silkworm *Bombyx mori* strains was investigated using isoenzymes of four metabolic enzymes viz., phosphoglucomutase (PGM), malate dehydrogenase (MDH), acid phosphatase (ACP) and aspartataminotransferase (AST) by polyacrilamide gel electrophoresis (PAGE). Three of the studied enzyme systems - PGM, MDH and ACP manifested intra- and inter-strain polymorphism with three or five alleles. AST was monomorphic in all strains tested. The observed heterozygosity was found to range from 0.000 to 0.266. Allele frequencies of all loci were used to estimate NEI's (1972) genetic distance, which was found to range between 0.004 and 0.457 among the strains studied. A perusal of genetic diversity within and among strains indicated that 34.72% of the observed variation occurred among strains and the rest of the variation (65.28%) within strains. Genetic relatedness of the ten strains revealed by the UPGMA dendrogram, showed genetic grouping of strains in two clusters. Populations of silkworm strains Kinshu and E 29, and Asahi and

Kinshu showed the highest percent of polymorphism and more number of alleles, respectively. Their rich genetic diversity needs to be exploited in conservation and breeding programme.

3. Ivanova E., Bienkowska M., Panasiuk B., Wilde J., **Staykova T**., Stoyanov I. 2012. Allozyme variability in populations of *A. Mellifera Mellifera* (Linnaeus 1758.), *A. M. Carnica* (Pollman, 1879) and *A. M. Caucasica* (Gorbachev, 1916) from Poland. *Acta Zoologica Bulgarica*, Suppl. 4, 79-86. (IF 0.309) (цитирана 2 публикации, 1 от които в списание с IF)

The genetic variability in honey bee populations of three subspecies reared in Poland (*A. m. mellifera*, *A. m. carnica* and *A. m. caucasica*) has been studied by usage of allozymic analysis of six enzymic systems (MDH-1, ME, EST-3, ALP, PGM and HK) corresponding to 6 loci. All loci were found to be polymorphic in the populations studied. Two alleles were detected at ME locus, three alleles – at MHD-1, ALP, PGM and HK, and five alleles – at EST-3 loci. The most frequent and the private alleles were discussed as suitable genetic markers for the subspecies characterization. The observed and expected heterozygosities (H<sub>o</sub> and H<sub>e</sub>) ranged from 0.233 (*A. m. carnica*) to 0.311 (*A. m. mellifera*) and from 0.268 (*A. m. carnica*) to 0.327 (*A. m. caucasica*), respectively. Allele frequencies of all loci were used to estimate nei's (1972) genetic distance, which was found to range from 0.025 (between *A. m. carnica* populations) to 0.518 (between *A. m. caucasica* and *A. m. mellifera* populations). The estimated mean F<sub>ST</sub> value from allozyme data was 0.28. UPGMA and Neighbour-Joining phylogenetic dendrograms were obtained by genetic distance matrix methods. The studied *A. m. mellifera*, *A. m. carnica* and *A. m. caucasica* populations were grouped in different clusters.

4. Ivanova E., Bouga M., **Staykova T**., Mladenovic M., Rasic S., Charistos L., Hatjina F., Petrov P. 2012. The genetic variability of honey bees from the Southern Balkan Peninsula, based on alloenzymic data. *Journal of Apicultural Research*, 51 (4), 329-335. DOI 10.3896/IBRA.1.51.4.06. (IF 1.926)

The genetic variability of honey bee populations, representing *Apis mellifera macedonica*, *Apis mellifera cecropia* and *Apis mellifera carnica* subspecies from the Balkan Peninsula countries of Bulgaria, Greece, Serbia and Montenegro, was studied using alloenzymic analysis of six enzyme systems (MDH-1, ME, EST-3, ALP, PGM and HK) corresponding to 6 loci. All loci were found to be polymorphic in most of the populations studied. The observed heterozygosity was found to range from 0.161 to 0.276. Allele frequencies of all loci were used to estimate Nei's (1972) genetic distance, which was found to range between 0.001 and 0.101 among the populations studied. UPGMA and neighbour-joining phylogenetic trees obtained by genetic distance matrix methods show that the honey bee populations from Bulgaria and Greece were clustered together, as were those from Serbia and Montenegro.

5. **Staykova T**., Ivanova E., Tzenov P., Vasileva Y., Arkova-Pantaleeva D., Grekov, D., Avramova K. 2015. Genetic analysis of isoenzymes polymorphism in silkworm (*Bombyx mori* L.) strains and phylogenetic relationships. *Acta Zoologica Bulgarica*, 67 (1), 117-125. (IF 0.31).

This study carried out to evaluates the polymorphism in the silkworm of thirty strains with different origin using the isoenzymes electrophoresis to detect biochemical markers and to

investigate the genetics of populations for those strains. The isozymes of nonspecific esterases (EST), malate dehydrogenase (MDH) and acid phosphatase (ACP) from hemolymph and phosphoglucomutase (PGM), hexokinase (HK) and aspartat aminotranspherase (AST) from silk glands they were submitted to the electrophoresis in polyacrilamide gels to 7.5%. The total of 9 loci were detected, and 8 (89%) of them were polymorphic. Intra- and inter-strain polymorphis was obtained. The mean number of alleles per polymorphic locus ranged from 1.0 to 2.2. The value of  $F_{st}$  (0.4556) it shows that the strains are differentiated. The dendrogram obtained with the values of genetic distance separated two main clusters and many subclusters. From obtained results it is showed that hemolymph EST, ACP and MDH, as well as PGM and HK from silk glands are appropriate markers for examination the genetic diversity and differentiating the silkworm strains.

6. Stoyanov I., **Staykova T.,** Stojanova A., Vasileva P., Ivanova E. 2015. Isoenzymic genetic variability in populations of *Messor structor*, (Hymenoptera, Formicidae) from Bulgaria, *Acta Zoologica Bulgarica*, 67 (3), 337-344 (IF 0.31)

Genetic variability in 36 different *Messor structor* populations from Bulgaria has been studied using analysis of two enzymic systems corresponding to four loci (Sod-1, Sod-2, Sod-3 and Me-1). Three of the studied loci were found to be polymorphic. One of them was found as monomorphic (Sod-3). One to three alleles were detected for the different isozymic systems: one allele at Sod-3 locus (Sod-3<sup>100</sup>), two alleles at Sod-1 (Sod-1<sup>100</sup> and Sod-1<sup>95</sup>) and Sod-2 (Sod-2<sup>100</sup> and Sod-2<sup>97</sup>) loci and three alleles – at Me-1 locus (Me-1<sup>100</sup>, Me-1<sup>98</sup> and Me-1<sup>96</sup>). The observed and expected heterozygosities (H<sub>o</sub> and H<sub>e</sub>) ranged from 0.0 to 0.111 and from 0.168 to 0.372, respectively. Allele frequencies of all loci were used to estimate Nei's (1972) genetic distance, which was found to range from 0.001 (between Boliarino and Lubenova mahala) to 0.462 (between Chirpan and Merichleri). The estimated mean F<sub>IS</sub> and F<sub>ST</sub> values from allozyme data were 0.8738 and 0.1432, respectively. The Neighbor-Joining method and Unweighted Pair Group Method with Arithmetic Mean phylogenetic trees were obtained by genetic distance matrix methods. Two of the studied populations were grouped separately in dendrograms as compared to all other populations which formed a large cluster consisting of three sub-clusters.

7. **Staykova T.,** Grekov D., Evangelou V., Emmanouil N., Bouga M. 2016. Genetic varyability of the silkworm, *Bombyx mori* (Lepidoptera: Bombicidae) from different geographical origin, based on mtDNA gene segment sequencing analysis. *Acta Zoologica Bulgarica*, 68 (4), 589-595 (IF 0.413)

Genetic variability and phylogenetic relationships in eight strains of the economically important silkworm *Bombyx mori* of different geographical origin and belonging to the Bulgarian germplasm were examined for the first time using 12S rDNA mtDNA gene segment sequencing analysis. We found 58 variable sites. Intra-strain variability was detected based on neighbour-joining, minimum evolution, maximum parsimony, UPGMA and PCA analyses. Eight haplotypes were revealed corresponding to the eight strains studied. PCA analysis distinguished Plovdiv 14, Plovdiv 18 and Maiak 5 strains from the rest. Maximum parsimony and UPGMA trees demonstrated that the strain Plovdiv 14 showed a large sequence divergence as compared to the other studied strains. Our results could be applied in

future conservation strategies and breeding programs for *B. mori* in Bulgaria. 12S rDNA mtDNA gene segment sequencing analysis could be used with isoenzyme analysis for discriminating of strains of different geographical origin in relation to their involvement in breeding programs and preserving their original characteristics.

8. Stoyanov I., **Staykova T.,** Vasileva P., Ivanova E. 2017. Genetic variability in populations of *Messor barbarus* (Hymenoptera, Formicidae) from Bulgaria based on isoenzyme analysis. *Acta Zoologica Bulgarica*, Supl (8), 31-35 (IF 0.413)

Genetic heterogeneity in 13 populations of *Messor barbarus* (Linnaeus, 1767) (Formicidae) from Bulgaria has been studied using analysis of seven enzyme and protein systems, which have been found to be appropriate genetic markers for characterization of genetic variability within and between populations. Totally, 49 allelic variants were found for the studied loci. A comparative analysis of gene pool and genotypic structure of the tested populations have been carried out. Moderate to high percentage of polymorphism (59.1–86.4) and low levels of heterozygosity have been calculated (0.030–0.066). Deviations from Hardy-Weinberg equilibrium in almost all analyzed loci, in favor of homozygotes, have been recorded. The mean value of inbreeding coefficient (F<sub>IS</sub>) has been found to be high (0.8212), demonstrating a high level of inbreeding in the studied populations. The obtained data of the genetic variability provide new information concerning polymorphism and phylogenetic relations between the studied populations.

9. Georgieva V., Petrov P., **Staykova T.,** Lazarov S., Stoyanov I., Ivanova E. 2017 Genetic comparison between local *Apis mellifera macedonica*, selectively reared for production of bee queens and swarms in Bulgaria and honey bee colonies with indicative hygienic behaviour. *Acta Zoologica Bulgarica*, Supl (8), 25-29 (IF 0.413)

The genetic polymorphism of malate dehydrogenase (MDH-1) and esterase (EST-3) loci in selectively reared for production of bee queens and bee swarms in Bulgaria local honey bee *Apis mellifera macedonica* was studied in comparison with a group of honey bee colonies with indicative hygienic behaviour. Totally 851 worker bees collected from the selection bee rearing bases and 414 worker bee individuals from colo-nies with manifested hygienic behaviour were used for this comparative study. Both of the studied loci were found to be polymorphic all of the studied honey bee groups. Polymorphism with two and three alleles was found for MDH-1 locus and with four and five alleles – for EST-3 locus for compared hygienic colonies and those under selective control. Levels of polymorphism between 50% and 100% were calculated for the studied honey bee groups. The observed and expected heterozygosities ( $H_o$  and  $H_e$ ), mean  $F_{ST}$  and Nm values were calculated and compared. The observed similarities and slight differences among honey bee groups under selective control and this with indicative hygienic behaviour were discussed.

10. Antov M., Stoyanov I., Stojanova A., **Staykova T.** 2017. Genetic variability of *Eupelmus* species (Hymenoptera: Eupelmidae) based on allozyme markers. *Acta Zoologica Bulgarica*, Supl (8), 17-23 (IF 0.413).

Four enzyme systems (MDH, ME, PGM and HK) of three parasitoid species of the genus *Eupelmus*, i.e. *E. vesicularis* (Retzius), *E. urozonus* Dalman and *E. microzonus* Förster were studied. A total of five populations were examined, three of them belonging to *E. urozonus*. Polymorphism of four alleles of MDH-1, PGM, HK-1 and HK-2 loci and three alleles of ME

locus was observed. Taxonomic markers for the tested *Eupelmus* spp. were found. The degree of polymorphism and heterozygosity of each species were calculated. UPGMA cluster analysis confirmed that *E. vesicularis*, which belongs to the subgenus *Macroneura*, is discriminated from *E. urozonus* and *E. microzonus*, which belong to the subgenus *Eupelmus*. One of the populations of *E. urozonus* from Osogovo Mt. was more similar to a population from Vitosha Mt. than to another population from Osogovo Mt.

11. Antov M., Stoyanov I., Stojanova A., **Staykova T.** 2017. Allozyme variability in three *Eupelmus* species (Hymenoptera: Eupelmidae) from Bulgaria. *North-Western Journal of Zoology*, 13 (2), 220-226 (IF 0.733).

Four enzyme systems i.e. malate dehydrogenase (MDH), malic enzyme (ME), acid phosphatase (ACP) and aspartate aminotransferase (AST) were studied in three parasitoid species of the genus Eupelmus (E. aseculatus (Kalina, 1981)), E. urozonus Dalman, 1820 and E. microzonus Förster, 1860) using PAGE. Only MDH and ME produced clear bands. No bands were obtained for ACP and AST. MDH enzyme was found to be useful to distinguish between Eupelmus species. Polymorphism with four alleles of the Mdh-1 locus and monomorphism of the Mdh-2 and Me loci were observed. Intraspecific and interspecific variability of Mdh-1 alleles in the studied species from Bulgaria were detected. Mdh-159 allele was unique to E. aseculatus. The degree of polymorphism and heterozygosity of each species were calculated. Cluster analysis, based on Nei's genetic distance confirmed that E. aseculatus, which belongs to subgenus Macroneura is divergent from E. urozonus and E. microzonus, which belong to subgenus Eupelmus.

#### II. Публикации, приравнени към такива с импакт фактор

12. Ivanova, E., **Staykova, T**., Velcheva, I. Cytotoxicity and genotoxicity of heavy metal- and cyanide-contaminated waters in some regions for production and processing of ore in Bulgaria. Bulgarian Journal of Agricultural Science, 2008, 14 (2), 262-268. *(цитирана в 14 публикации, 3 от които в списания с IF/SJR)* 

This study generalizes the data of investigations on cytotoxic and genotoxic effect of heavy metal- and cyanide-contaminated waters in 2001, 2003 and 2005. The water samples were collected from different water sources in the region of "Assarel-Medet" Copper Refinery Works. The contents of copper, arsenic, cadmium, lead, and cyanides (mg.dm-3) were determined using the method of automatic photometry. The *Allium cepa* and *Pisum sativum* plant systems were used for testing of the cytotoxicity and genotoxicity of heavy metals and cyanides. A lower mitotic index and a higher frequency of chromosome aberrations were established in all test samples than in the control ones. Chromosome fragments, anaphase and telophase bridges, micronuclei, lagging chromosomes and C-mitotic effect in cells were observed. It was concluded that the pollution found in the regions for production and processing of ore has cytotoxic and genotoxic effect on cells and it could be a potential threat to water ecosystems and human health.

13. Ivanova E., **T. Staykova**, I. Stoyanov, P. Petrov. 2012. Allozyme genetic polymorphism in Bulgarian honey bee (*Apis mellifera* L.) populations from the south-eastern part of the Rhodopes. *Journal of BioScience and Biotechnology*, 1 (1), 45–49.

Allozyme genetic polymorphism in Bulgarian honey bee populations from four different locations in the south-eastern part of the Rhodopes Mountain was studied on six enzymic systems (MDH, ME, EST, ALP, PGM and HK) corresponding to six genetic loci. Allozyme analysis revealed that all studied loci were polymorphic in almost all investigated populations. The observed heterozygosity was found to range from 0.110 to 0.208 and Nei's genetic distance – between 0.016 and 0.061 among the studied populations. These honey bee populations were clustered in two groups in the UPGMA dendrogram. The Tihomir population was in a separate clade while other three populations (Kardzhali, Krumovgrad and Dolni Yurutci) were grouped together. (μμπυραμα ε 3 ηγόλιμκαμμα, 1 οπ κουμπο c IF)

#### III. Публикации в реферирани списания без импакт фактор

14. Ivanova, E., Staykova, T. Stage specificity in the expression of proteins of honey bee fat body (*Apis mellifera* L.) in the course of ontogenesis. // Journal of Cell and Molecular Biology, 2007, 6, 2, 129-135.

The aim of this study was to investigate and analyze some peculiarities in the age specificity of protein expression in the fat body of *A. mellifera* during the ontogenesis of the individuals. By using 7.5% ployacrylamide gel electrophoresis, 189 individual samples of fat body extracts were investigated in the course of their development in the following stages and ages: larvae-third, fourth and fifth larval age (L3, L4, L5); prepupae (PP) and pupae-white-eyed and dark-eyed pupae (WP, DP). A total of 25 protein fractions were expressed in the fat body of male and female individuals during their development. Some sex-differences in studied fat body proteins were noticed. The age and stage specific dynamics in the expression of established proteins of the fat body spectrum were analyzed and commented.

15. Vasileva Y., Tzenov P., **Staykova T**. 2009. Study on productivity and genotype structure by several enzyme loci of silkworm (*B. mori* L.) parthenoclones obtained by thermal and combined (low-high temperature) method. *International Journal of Industrial Entomology*, 18 (2), 131-134.

The purpose of this study was to establish whether there are differences in the productivity of the same silkworm ( $B.\ mori\ L.$ ) parthenoclones, obtained by two different methods – thermal and combined, as well as to study their genotype structure by several enzyme loci. It was established that all individuals of parthenoclones Joana, Joana ( $\downarrow\uparrow$ ), Pohi and Pohi ( $\downarrow\uparrow$ ), are homozygous by the studied esterase and phosphoglucomutase loci, which substantiated the clones' genetic stability. By comparative analysis of some biological and technological properties, it was found that parthenoclone Pohi ( $\downarrow\uparrow$ ) obtained by low-high temperature activation is characterized by higher values of these properties as compared to parthenoclone Pohi obtained by thermal parthenogenesis. Comparing the two methods of inducing ameiotic parthenogenetic development, we would recommend that parthenoclone Joana is sustained by thermal parthenogenesis, and parthenoclone Pohi – by the combined method (low-high temperature).

16. **Staykova, T.,** Ivanova, E., Tzenov, P., Vasileva, Y., Arkova-Pantaleeva, D. Differentiation of silkworm (*Bombyx mori* L.) strains by isoenzime markers. Genetics and Breeding, 2009, 38 (1), 47-55.

This study included 12 strains of various origin, grown at the Sericulture Experiment Station of Vratza, Bulgaria. The nonspecific esterases from the haemolymph, phosphoglucomutase and aspaftate aminotransferase from the silk glands of V<sup>th</sup> instar larvae were analyzed by 7.5% PAGE. Polymorphism was found in the nonspecific esterases and the phosphoglucomutase. The allele frequencies, the average number of alleles per locus, the percentage of polymorphic loci, the observed and expected heterozygosity, genetic distance Nei, 1972) and the fixation index (Fst) were calculated through the BIOSYS-1 program. In the constructed UPGMA dendogram the studied strains were grouped in two main clusters. It was found that nonspecific esterases are very suitable markers for analyzing inter-strain polymorphism and strain differentiation, while phosphoglucomutase is more suitable for studying intra-strain polymorphism and determining the degree of intra-strain genetic variability.

17. Ivanova E., Ivgin-Tunka R., **Staykova T.** 2009. Genetic characterization of honey bee (*Apis mellifera* L.) populations from Bulgaria using allozymes. *Genetics and Breeding*, 38 (1), 67-74.

The genetic variability of honey bee populations from Bulgaria was screened with three enzyme system. Allozyme analysis of MDH-1, ME and PGM loci revealed that all of them were polymorphic in all of population studied. The mean number of alleles per locus was 2.0. Observed heterozygosity was between 0.165 and 0.249. Nei's genetic distance ranges between 0.002 and 0.064. UPGMA trees cluster the Bulgarian honey bees of population studied in two groups. The estimated percentage of polymorphic loci was between 33.3% and 100% in populations studied. The estimated mean  $F_{ST}$  value of 0.0575 for all allozyme data in this research indicated a low level of genetic differentiation among populations – 5.75%.

18. Ivanova, E., **Staykova**, **T**., Dhzambazov, B. Cytotoxicity and genotoxicity of the cyanoprokaryote *Nostocmicroscopicum*. *Genetics and Breeding*, 2009, 38 (1), 13-19.

Allium root meristem was used as an in vivo test system to evaluate the toxicity of cyanoprokaryote Nostoc microscopicum. Extracts in water-Dimethyl sulfoxide (9:1, v:v), water-methanol (1:9, v:v) andwater-acetic acid (9.5:0.5, v:v) solutions were used for testing. To studying the cyto- and genotoxicity of Nostoc microscrspicum temporary squash preparations were prepared from Allium cepa root meristem tissue. Seeds germinated in dechlorinated tap water were analyzed as a control sample. Seeds germinated in the extracts mentioned above but diluted 50 and 100 times were used as test samples. Two thousand cells per sample were examined. Comparing the test samples with the control, a lower mitotic index after treatment with water-methanol extracts and higher mitotic index after treatment with water-dimethyl sulfoxide extracts was established. In most of the samples tested, the total frequency of chromosome aberrations was higher than in the control. It was found that different extracts of cyanoprokaryote Nostoc microscopicuminduce cause damages such as C-mitosis, fragments, laggards, anaphasic and telophasic bridges, disturbed anaphase and micronuclei.

19. Ivanova E., **Staykova T.,** Petrov P. 2010. Allozyme variability in populations of local Bulgarian honey bee. *Biotechnology & Biotechnological Equipment*. 24 (2), 379 – 384. (цитирана в 7 публикации, 4 от които в списания с IF)

Genetic variation of honey bee populations from six different locations corresponding to tree geographic regions: North-western, North-central and North-eastern of Bulgaria was studied on 6 enzymic systems (MDH, ME, EST, ALP, PGM and HK) corresponding to 6 genetic loci. Allozyme analysis revealed that all loci studied were polymorphic in almost all populations studied. The mean number of alleles per locus varied from 1.8 to 2.5. The estimated percentage of polymorphic loci was between 50% and 100%. The observed and expected heterozygosities (Ho and He) ranged from 0.17 to 0.221 and 0.250 to 0.315, respectively. There are not significant deviations of genotype frequencies from Hardy-Weinberg expectations at most of the loci in most populations (0.99 > P > 0.1). The estimated mean FST value from allozyme data was 0.0443 which shows that 4.43% of the overall genetic diversity observed was among populations, as opposed to 95.57% within populations. The values of genetic distance range from 0.002 to 0.036. UPGMA dendrograms were constructed.

20. **Staykova, T.**, Ivanova, E., Panayotova, G., Cvetkova, I., Dzhoglov, S., Dzhambazov, B. General toxicity and genotoxicity of *Nodularia moravica (Cyanoprokaryota, Nostocales)*. *Biotechnology & Biotechnological Equipment*, 2010, 24 (2), 397 – 400.

General toxicity and genotoxicity of cyanoprokaryote *Nodularia moravica* was investigated on the base of *Allium* root meristem in vivo test system. Seeds sprouted in dechlorinated tap water were analyzed as a control sample. Seeds sprouted in the water-Dimethyl sulfoxide (9:1, v:v) extract, diluted 50 and 100 times (D1-797 and D2-797, respectively) were used as test samples. It was found that different concentrations of dilutions of the initial extract of cyanoprokaryote *Nodularia moravica* induced damages such as micronuclei (MNi), fragments, anaphasic and telophasic bridges and laggards with higher total frequency than in the control. Germination percentage and root length were found to be in positive correlation with percentage of chromosome aberrations and depend on the extract concentration. It was concluded that *Nodularia moravica* extract in water-DMSO solution induces general and genotoxycity in *Allium cepa* root meristem cells. There were not found data about cytotoxicity.

21. **Staykova, T.**, Ivanova, E., Tzenov, P., Vasileva, Y., Arkova-Pantaleeva, D., Petkov, Z. Acid phosphatase as a marker for differentiation of silkworm (*Bombyx mori*) strains. *Biotechnology & Biotechnological Equipment*, 2010, 24 (2), 379 – 384.

This study was carried out on twelve silkworm strains maintained by the Sericulture and Agriculture Experiment Station germplasm bank, located in Vratza, Bulgaria. The polymorphism of acid phosphatase from larval haemolymph was investigated by method of electrophoresis in polyacrylamide gel. Five fundamental types of this enzyme were found in the studied strains of various origins. The acid phosphatase isozymes were considered to be controlled by five codominant alleles. It was found out that the acid phosphatase is very suitable marker for analyzing the inter- and intra-strain diversity and the strain differentiation.

22. **Staykova, T.,** Ivanova, E., Grekov, D., Petrov, P., Tzenov, P., Vasileva, Y., Petkov, Z., Arkova-Pantaleeva, D. Development of a specialized center for scientific, training and diagnostic work for the needs of Apidology and Sericology in Bulgaria. *Advances in Bulgarian Science – Annual*, 2010, 1, 63-67.

The aim of the project is the establishment and development of a specialized center for scientific, training and diagnostic work for the needs of apidology and sericology in Bulgaria, uniting the scientific-research units of "Pasisii Hilendarski" University of Plovdiv, the Agricultural University - Plovdiv, Sericulture Experimental Stationat the Agricultural Academy and the National Breeding Association of Apiculture. The following activities will be carried out in the center: interdisciplinary researches for a complex evaluation of populations A. mellifera and B. mori through phenotype and genotype characterization at different levels, training of students, master degree students, postgraduate and PhD students, qualification of farmer-beginners and active farmers in the field of apiculture and sericulture and selective-genetic diagnostics through reliable isoenzymic and DNA-markers. The project realization will lead to improving and updating the research facilities of the consortium, as well as the conditions for research and training activity. New opportunities for work at the European university and scientific space will be created through establishing an optimal environment for international cooperation. The complex scientific-research activity of the operating center will be also oriented to creating highly productive breeds, lines and hybrids of silk worm, as well as highly productive queen bees and initial drone families. All this, together with the training of farmer-beginners and active farmers and the promotion of the center activity, will provide opportunities for more intensive development of apiculture and sericulture in regions with suitable climatic conditions and resource. The specialized center will have the necessary intellectual and material-technical potential to continue its scientificresearch, training and diagnostic activity for an indefinitely long period of time.

23. **Staykova, T.**, Ivanova, E. Concerning genetic variability and usable isozyme markers for characterization of *A. mellifera* L. populations and *B. mori* L. breeds in Bulgaria. *Advances in Bulgarian Science*, 2011, 20-28.

Genetic variability in different populations of Bulgarian honey bee *Apis mellifera* from the all six regions in the country have been studied using analysis of six enzymic systems corresponding to six loci (MDH 1, ME, EST 3, ALP, PGM and HK). All of the studied loci were found to be polymorphic. Three to six alleles were detected for the different allozymic systems: three alleles – at MDH-1, ALP and HK loci; four alleles – at ME and PGM loci and six – at EST-3 locus. Genetic markers, usable for discrimination of Bulgarian honey bees were described in this study. A genetic analysis was conducted on 10 breeds of *B. mori*. Suitable isoenzyme markers were specified for evaluation of interbreed and intrabreed polymorphism and breed differentiation for this species. The phylogenetical relationships were studied. It was established that breeds E 27, Japanese 106 and Jena may be used in future breeding programs as donor breeds as they are genetically distant from all others.

24. **Стайкова, Т**., Иванова, Е. Вътрепороден и междупороден генетично детерминиран полиморфизъм по малатдехидрогеназа и хексокиназа и филогенетична диференциация при черничевата копринена пеперуда *Вотвух тогі* L. *Пловдивски университет "Паисий Хилендарски" Юбилеен сборник, Биологически науки за подобро бъдеще*, 2012, 253 – 263.

Изследвани са десет породи с различен произход, отглеждани в България. Чрез 7.5% PAGE са анализирани малатдехидрогеназата (MDH - EC 1.1.1.37) от хемолимфата и хексокиназата (HK - EC 2.7.1.1) от коприноотделителните жлези на ларви V възраст. Констатиран е вътрепороден и междупороден полиморфизъм по малатдехидрогеназата

и междупороден полиморфизъм по хексокиназата. Чрез BIOSYS-1 програмата са изчислени алелните честоти, средният брой алели за локус, процента на полиморфии локуси, установената и очаквана хетерозиготност, генетичната дистанция по Nei и индексът на фиксация на Wright ( $F_{ST}$ ). Построена е дендрограма, в която изследваните породи са групирани в два основни кластера. Установено е, че MDH е подходяща за изследване на вътрепородния полиморфизъм, а HK е подходяща за анализиране на междупородния полиморфизъм и породната диференциация.

25. **Staykova, T.**, Popov, P., Ivanova, E., Breed specific expression of gut silkworm (*Bombyxmori*L.) nonspecific esterases. *Journal of BioScience and Biotechnology*, 2012, 1 (1), 27–31.

Nonspecific esterases of silkworm (*Bombyx mori* L.) gut were investigated by means of polyacrilamide gel electrophoresis (PAGE). Stage-specific expression of eleven esterase bands was detected during larval development of breeds and inter-breed hybrids kept in Bulgaria. In two esterase zones, intra- and inter-breed polymorphism was found. The polymorphism in fraction GES I1 specific for the gut may be used for testing of the breeds raised in our country with reference to determining the degree of genetic heterogeneity. The specific expression in zone GES L1 observed at present study confirmed gene determinate polymorphism in zone BES E1, observed earlier.

26. **Staykova T.** 2013. Inter- and intra-population genetic variability of introduced silkworm (*Bombyx mori* L.) strains raised in Bulgaria. *Journal of Bioscience and Biotechnology*, 2 (1), 73-77.

The genetic variability of four populations belonging to two introduced silkworm strains (*Bombyx mori* L.) of various origins has been studied using isoenzymic analysis of six enzyme systems. Nonspecific esterases, phosphoglucomutase, malate dehydrogenase, acid phosphatase, alkaline phosphatase and hexokinase from different tissue of larvae 5th instar have been analysed using PAGE. Polymorphism in six from a total of nine loci has been found. Inter- and intra-population differences have been ascertained expressed in different allele composition of the gene pool and different frequencies of alleles. A higher degree of inter-population variability has been reported on the acid phosphatase and a lower one – on the phosphoglucomutase.

27. Stoyanov I., Ivanova E., Vasileva P., **Staykova T.** 2015. Soluble proteins in *Messor structor* (Latreille, 1798) (Hymenoptera: Formicidae) populations from Bulgaria – genetic variability and possible usage as population-genetic markers. *ZooNotes*, 72: 1-9.

The genetic variability of ant species determined as *Messor structor* (Latreille, 1798) or close to it ("*M*. cf. *structor*") from Bulgaria has been studied using polyacrylamide gel electrophoresis analysis of five soluble protein systems (Sp-1, Sp-2, Sp-3, Sp-4 and Sp-5) corresponding to 5 loci. Four of the studied loci were found to be polymorphic. Two alleles were detected at Sp-1 and Sp-2 loci and three – at Sp-3 and Sp-5. The observed and expected heterozygosities (H<sub>o</sub> and H<sub>e</sub>) ranged from 0.0 (Yambol) to 0.140 (Topolovo) and from 0.170 (Nova Zagora) to 0.401 (Tvarditsa), respectively. The calculated mean value of inbreeding coefficient (F<sub>IS</sub>) was 0.8263 and demonstrated high level of inbreeding within populations, which correlated with a low level of observed heterozygosity compared to the expected one.

The estimated mean fixation index  $(F_{ST})$  value was 0.2746. Allele frequencies of soluble protein loci were used to estimate Nei's (1972) genetic distance and to obtain the Unweighted Pair Group Method with Arithmetic Mean (UPGMA) and Neighbor-Joining (NJ) dendrograms, where Topolovo and Nova Zagora populations were grouped separately than other populations.

#### IV. Публикации в сборници от международни конгреси и конференции

28. Tunka, R., Staykova, T., Ivanova, E., Kence, M., Grekov, D. Differentiation of silkworm, *Bombyx mori* strains measured by RAPD analyses. *Proceedings of the International conference "Sericulture challenges in the 21<sup>st</sup> Century & the 3<sup>rd</sup> BACSA meeting, 18-21 september, 2007, Vratza, Bulgaria, 247 – 251.* 

The patterns of random amplified polymorphic DNA (RAPD) variation among six silkworm ( $Bombyx\ mori$ ) strains with different origins are examined. A total of ten RAPD primers were used and all of the loci are polymorphic. The genotypic diversity (H') inferred from Shannon's index (0.4459 for all populations) is higher than gene diversity (H<sub>t</sub>) (0.2896 for all populations), which suggests that diversity is distributed among lineages. Estimates of gene diversity in populations are higher in total (H<sub>t</sub>) as compared to within population diversity (H<sub>s</sub>). Highest genetic differentiation is obtained between China 23 and the other five strains. The highest genetic distance is determined between China 23 and Shova (0.1713). The highest gene flow (6.8048) is obtained between Asahi and Gindga 8 strains, whereas the lowest between China 23 and Shova. The highest  $G_{st}$  value corresponds well with the lowest Nm value whichdesignates that there is a small amount of gene flow. The dendogram is constructed by using the UPGMA method based on Nei's (1978) genetic distance and separated the China 23 strain from all the others that formed together a large cluster. The genetic diversity in the studied strains is moderately low.

29. Ivanova, E., Staykova, T., Petrov, P. ALP as population-genetic markers for Apis mellifera. *International conference "Science, education and time as our concern"*, Smolian, 2007, III, 23-26.

Seven honey bee populations from different regions in Bulgaria have been examined by using PAGE in order to study the genetic polymorphism of the alkaline phosphatase. Monoloci polymorphism of this enzyme system has been investigated. There were found two alleles for Alp locus (Alp-80 and Alp-100). Genotype and allele frequencies and enzyme heterozygosities were calculated. The test goodness of fit of gene frequencies to Hardy-Weinberg expectations was carried out using  $x^2$  analysis. This polymorphism in honeybee populations from Bulgarian locations studied has been reported at first time.

30. **Staykova T.**, Grekov D. 2008. Some biological, technological and biochemical-genetic characteristics of mulberry silkworm (*Bombyx mori* L,) lines established through insertional mutagenesis. *Proceedings of XXI*<sup>st</sup> Congress of the International Sericultural Commission, 3<sup>d</sup> - 6<sup>th</sup> November 2008, Athens-Greece, 96-99.

Increasing productivity of silkworm is the major and final aim of breeding science. Modern knowledge in the area of genetics, cytology and biochemistry opens wide possibilities for development and introduction of new methods. One of those possibilities is the method of artificial obtaining of mutations. Ethylmethanesulfonate (EMS), 1.20g relative weight, was used as a mutagenic factor. Nine lines were selected. The major biological, technological and biochemical-genetic characteristics were studied. Biochemical markers (non-specific esterases) were used for investigating the genetic variability in the studied variants. Some biological and technological differences of the major characteristics were established. Bi- and triallele polymorphism was detected by PAGE in three of the esterase loci. Different allelic frequencies were found in the separate variants.

31. **Staykova T. A.**, P. I. Tzenov, Y. B. Vasileva, D. B. Arkova-Pantaleeva, D. Grekov and K. Avramova. 2011. Phylogenetic differentiation of silkworm (*Bombyx mori* L.) strains with different origin, rased in Bulgaria. *Proceedings of 5<sub>th</sub> BACSA International Conference* "SERICULTURE FOR MULTI PRODUCTS – NEW PROSPECTS FOR DEVELOPMENT" *SERIPRODEV*, *April* 11<sup>th</sup> – 15<sup>th</sup> 2011, *Bucharest*, *Romania*, 102-108.

Ten diverse strains of the silkworm  $Bombyx\ mori\ L$ . with various origin were analysed using hemolymph nonspecific esterase polymorphism. The allele frequencies, the average number of alleles per locus, the percentage of polymorphic loci, the observed and expected heterozygosity, genetic distance by Nei and the fixation index by Wright ( $F_{ST}$ ) were calculated through the BIOSYS-1 program. The average number of alleles per locus varied from 1.3 to 2.5. The degree of polymorphism ranging from 100% to 25% with heterozygosity value of 0.392 - 0.083. The mean value of  $F_{ST}$  calculated on the base of the esterase isozymes polymorphism, showed that 31.16% of the genetic variability was observed between the different strains, which corresponds to the level of the inter-strain genetic differentiation. The phylogenetic tree constructed by the UPGMA method consisted of two major subgroups from E 29 strain. These data for the inter- and intra-strain diversity and strain differentiation could be used for breeding purposes of the mulberry silkworm  $Bombyx\ mori\ L$ . and marker-assisted selection.

32. Стоянов И., П. Василева, Т. Попова, **Т. Стайкова**, Е. Иванова. 2012. Токсично и мутагенно действие на фунгицида Верита ВГ върху *Pisum sativum* растителна тестсистема *in vivo*. *Юбилейна национална научна конференция с международно участие "Традиции, посоки, предизвикателства", Смолян, 19-21 октомври, 2012. Том ІІ, част І, Природни науки, математика и информатика (Природни и аграрни науки. Медицина). Издателство ПУ "Паисий Хилендарски", филиал – Смолян. 37-45.* 

The Pisum sativum plant test sistem in vivo was used for testing of cytotoxic and genotoxic effects of widely used in recent years fungicide Verita WG. The effect of different solutions concentrations was studied. It was establish higher toxic effect in the germinated seeds with different fungicide concentration compared to control probe. The results of cytogenetic analysis showed cytotoxic and mutagenic effects of the tested fungicide.

33. **Staykova T.,** P. Tzenov, Y. Vasileva, D. Arkova-Pantaleeva, D. Grekov, K. Avramova. 2013. Passport data of six Bulgarian strains of silkworm *Bombyx mori* L. on the base of population genetic parameters. 6<sup>th</sup> BACSA INTERNATIONAL CONFERENCE "Building"

Value Chains in Sericulture" "BISERICA" 2013, Padua, Italy, April 7<sup>th</sup> – 12<sup>th</sup> 2013, 246-253.

Six silkworm strains created in Bulgaria were tested with polyacrilamide gel electrophoresis (PAGE) to assess the genetic structure of the populations on some isoenzyme loci. Among ten investigated isozyme loci, six loci (Bes B, Bes D, Bes E, Pgm, Mdh A, Bph A) proved to be polymorphic and manifested intra- and inter-strain polymorphis. The other four loci (Bes A, Hk A, Adh A, Ast A) were monomorphic in all strains tested. The mean number of alleles per polymorphic locus ranged from 1.5 to 1.9. The expected heterozygosity was higher than the observed one. Passport data of each of the strain on the base of population genetic parameters were indicated. Phylogenetic relationships between strains were revealed by the UPGMA and Neighbor-joining dendrograms.

#### Г. Учебници и учебни пособия:

34. Иванова, Е., **Стайкова, Т**., Андреенко, Е. Генетика с биологични основи на поведението и психогенетика. Университетско издателство "Паисий Хилендарски". Пловдив, 2011, 380.

В настоящия учебник са разгледани основите на общата генетика в комбинация с биологични основи на поведението и с психогенетика. Това комбинирано четиво е подходящо за студенти, както от специалностите "Психология" и "Специална педагогика" (Дефектология) на Педагогическия факултет, изучаващи дисциплините Биологични основи на поведението и генетика и Основи на генетиката, така и за студентите от специалностите на Биологическия факултет, изучаващи дисциплините Основи на генетиката, Генетика, Обща и популационна генетика. Учебникът може да се използва успешно и от студентите на специалност Дефектология, изучаващи дисциплината Анатомия и физиология на висшата нервна дейност, както и от всички студенти по биология, които изучават морфологията и физиологията на нервната система като база за изява на човешкото поведение.

Темите, включени в съдържанието на учебника покриват в по-голямата си част програмите на посочените дисциплини в учебните планове на акредитирани специалности в двата факултета на Пловдивския университет "Паисий Хилендарски". Учебникът е разработен от специалисти с дъгогодишен преподавателски опит в областта на общата и популационна генетика, анатомията и физиологията на висшата нервна дейност и има амбицията да поднесе полезно и интересно учебно съдържание, провокиращо стремеж към познание и творчество. Посочено е участието на всеки от авторите при разработване на тематичното съдържание.

35. Иванова, Е., **Стайкова, Т.,** Андреенко, Е., Василева, П., Калайджиева, В., Стоянов, И., Джоглов, С., Дончева, В., Панайотова, Г., Цветкова, И. Генетични основи на поведението. Университетско издателство "Паисий Хилендарски". Пловдив, 2011, 237.

Книгата "Генетични основи на поведението" е разработена от голям колектив, включващ преподаватели от три катедри на Биологическия факултет на ПУ "П.

Хиледарски" – Биология на развитието, Анатомия и физиология на човека и Зоология, както и магистри и магистранти от магистърската програма по генетика.

Книгата е планирана като учебно пособие за студенти по психология и дефектология, както и за студенти-биолози, интересуващи се от (и изучаващи) генетика на поведението. Съдържанието е структурирано в четири части: Основи на генетиката; Биологични основи на поведението; Генетика на поведението (при животните) и Генетика на човешкото поведение. Към всяка част са посочени конкретните автори.

## Обобщение относно приносите

### в представените за рецензиране научни трудове

Научните интереси и разработки на доц. д-р Теодора Атанасова Стайкова са в областта на популационната генетика, онтогенетиката и екологичната генетика и включват:

- Анализиране на популационно-генетични параметри при:
  - два вида насекоми със стопанско значение черничевата копринена пеперуда (*Bombyx mori*) и медоносната пчела (*Apis mellifera*);
  - други видове насекоми мравки от род Messor и паразитоидни оси от семейство Eupelmidae;
- Определяне на подходящи генетични маркери за анализиране на вътрепопулационната и междупопулациона изменчивост при *Bombyx mori* и *Apis mellifera* с оглед на използването им в селекционната и развъдна дейност;
- Определяне на подходящи генетични маркери за анализиране на степента на генетична хетерогенност в популации на други видове насекоми и изследване на филогенетичните връзки между тях;
- Изследване на филогенетичните връзки между породи *Bombyx mori* и популации *Apis mellifera* с различен географски произход;
- Изучаване на тъканната и стадийна специфичност в изявата и генетичния контрол на общи протеини и някои изоензими при *Bombyx mori* и *Apis mellifera*;
- Проучване на цитотоксичния и генотоксичен ефект на замърсени с тежки метали и цианиди питейни и природни води;
- Изучаване на обща токсичност, цито- и генотоксичност на цианокариоти от видовете *Nostoc microscopicum* и *Nodularia moravica*;
- Проучване на възможности за осъществяване на експресен екологичен мониторинг на базата на цитогенетични методи с използване на растителни тестобекти

#### СПРАВКА ЗА ПРИНОСИТЕ

# в научните трудове на доц. д-р Теодора Атанасова Стайкова за периода след придобиване на академичното звание "доцент"

### І. Приноси с научен характер

#### 1. Оригинални приноси

## 1.1. Оригинални приноси свързани с проучвания на черничевата копринена пеперуда *Bombyx mori*

- Оригинален научен принос е направената детайлна популационно-генетична характеристика на български и интродуцирани породи черничева копринена пеперуда от генетичния ресурс на България, и анализирането на подходящи изоензимни маркери за разграничаването им (публикации 1, 2, 5, 16, 21, 23, 24, 26, 31).
- Оригинален научен принос са проучените чрез изоензимен анализ филогенетични връзки между различни породи черничева копринена пеперуда отглеждани в България (публикации 2, 5, 16, 21, 23, 24, 26, 31).
- За първи път, чрез RAPD ДНК метод са анализирани популационно-генетични характеристики на отглеждани в България породи и хибриди *Bombyx mori* с различен географски произход, като са разгледани и филогенетичните зависимости между тях (публикация 28).
- За първи път при породи *Bombyx mori* от генетичния ресурс на България е проведен митохондриален ДНК анализ за изследване на генетичната изменчивост и установяване на филогенетични зависимости (публикация 7).
- За първи път в хода на ларвното развитие на отглеждани в България породи и хибриди *Вотвух тогі* е изследван спектъра на неспецифичните естерази в черво и е установена стадийноспецифична експресия, както и породноспецифичен полиморфизъм, въз основа на който е допуснато съответствие между спектрите на хемолимфа и черво по отношение на един от естеразните изоензими (публикация 25).

- Оригинален научен принос са публикуваните паспортни данни на български породи черничева копринена пеперуда, които включват популационно-генетични параметри, характеризиращи генофонда и генотипната им структура по подбрани изоензимни маркери (публикация 33).
- Оригинален научен принос е едновременното проучване на някои биологични, технологични и биохимико-генетични признаци при различни линии *Bombyx mori*, създадени в България чрез метода на индуциран мутагенез, във връзка с перспективата за използването му в селекцията на този вид у нас (публикация 30).
- За първи път по отношение на някои ензимни локуси е анализирана генотипната структура на създадени в България партеноклонове на черничевата копринена пеперуда (публикация 15).

### 1.2. Оригинални приноси свързани с проучвания на други видове

- Оригинален научен принос е проучената чрез алозимен анализ генетична изменчивост в популации на *Apis mellifera mellifera*, *Apis mellifera carnica* и *Apis mellifera caucasica* (от Полша), както и популации на *Apis mellifera macedonica*, *Apis mellifera cecropia* и *Apis mellifera carnica* (от България, Гърция, Сърбия и Черна гора) и характеризирането на филогенетичните зависимости между тях (публикации 3, 4).
- За първи път въз основа на изоензимен анализ е направена детайлна популационно-генетична характеристика на някои популации български медоносни пчели като са изчислени алелни честоти, нива на хетерозиготност и полиморфизъм, генетична дистанция и близост (публикации 13, 17, 19, 23).
- За първи път е установена висока степен на генетично сходство при сравнение между селекционно контролирани популации на *Apis mellifera macedonica* (тип *rodopica*) и подбрани пчелни семейства от България с подчертано силно изявено хигиенно поведение (публикация 9).
- Оригинален научен принос са характеризираните особености в изявата на разтворимите протеини от мастното тяло при медоносните пчели в хода на онтогенезата им (публикация 14).

- За първи път на базата на изоензимен анализ и изследване на общи протеини е проучен генетичния полиморфизъм и е характеризирана степента на генетична хетерогенност в популации на мравки от видовете *Messor structor* и *Messor barbarous* (публикации 6, 8, 27).
- Оригинален научен принос е характеризирането на степента на генетична диференциация между популациите на двата вида *Messor structor* и *Messor barbarous* и проучването на вътревидови и междувидови филогенетични зависимости (публикации 6, 8, 27).
- За първи път на базата на изоензимен и алозимен анализ е изследвана генетичната изменчивост в популациите на видове паразитоидни оси от род *Eupelmus* в България *Eupelmus urozonus* и *E. microzonus* (принадлежащи към подрод *Eupelmus*), и *E. vesicularis* и *E. aseculatus* (принадлежащи към подрод *Macroneura*) (публикации 10, 11).
- За първи път е проучена степента на генетична диференциация между популациите на видове паразитоидни оси от род *Eupelmus* в България и са характеризирани установените филогенетични зависимости (публикации 10, 11).

## 1.3. Оригинални приноси свързани с проучвания на цитотоксичност и генотоксичност

• Оригинален научен принос е проучването на общата токсичност, цитотоксичност и генотоксичност на цианопрокариоти от видовете *Nostoc microscopicum* и *Nodularia moravica* чрез *Allium сера* тест метода (публикации 18, 20).

#### 2. Приноси с потвърдителен характер

• Потвърден е установеният в други изследвания цитотоксичен и генотоксичен ефект на тежки метали и цианиди, съдържащи се в питейни и природни води (публикация 12).

- Потвърдено е мнението на други автори относно възможностите за осъществяване на експресен екологичен мониторинг на базата на цитогенетични методи с използване на растителни тест-обекти (публикации 12, 18, 20, 32).
- Потвърдено е установеното и от други автори ниско ниво на хетерозиготност в популациите на ципокрили насекоми (публикации 6, 8, 10, 11, 27).
- Чрез популационно-генетичен анализ е потвърдена видовата принадлежност на популациите на *E. urozonus*, *E. microzonus*, *E. vesicularis* и *E. aseculatus* от различни местонахождения на територията на България, определена на базата на морфологични признаци (публикации 10, 11).

#### II. Научно-приложни приноси

- За първи път на базата на различни генетични методи е направена популационно-генетична характеристика на породи *Bombyx mori* отглеждани в България, която може да бъде използвана при планирането на бъдещи селекционни дейности и осъществяване на междупородна хибридизация (публикации 1, 2, 5, 7, 16, 21, 23, 24, 25, 26, 28, 31, 33).
- За първи път на базата на изоензимен анализ е направена генетична характеристика на популации медоносни пчели в България и Европа като получените резултати са значима база за генетичен мониторинг на *Apis mellifera* в страната и могат да се използват при реализирането на национална селекционна програма (публикации 4, 13, 17, 19, 23, 29).
- Създаден е специализиран център за научна, учебна и диагностична дейност за нуждите на апидологията и серикологията в България (публикация 22).
- Установени са генетични маркери за разграничаване на видовете *Eupelmus urozonus*, *E. microzonus*, *E. vesicularis* и *E. aseculatus*, принадлежащи към двата различни подрода на род *Eupelmus Eupelmus* и *Macroneura* (публикации 10, 11).

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