

CONSERVATION AND VALORISATION OF BEE SPECIES *APIS MELLIFERA CARPATICA* IN CONTEXT CLIMATE CHANGE

Valentina CEBOTARI, Ion BUZU

Institute of Zoology of Academy of Sciences of Moldova (ASM),
1 Academiei Street, MD 2028, Chişinău, Republic of Moldova

Corresponding author email: valentinaceb@yahoo.com

Abstract

The aim of this scientific paper was to identify and highlight the innovative technology of conservation and valorisation of the bee species *Apis mellifera* Carpatica in the conditions of climate change. Scientific research has been conducted on bee populations growth in the experimental apiary of the Institute of Zoology. Research results have shown that the sustainable selection of purebred bee families, with the application of innovative methods of genetic amelioration in climate change, contributes to the conservation of the population with an appropriate level of development of morpho-productive traits. The conservation of bee populations requires their protection from pesticide residues, which are more and more common in the flowers of some entomophilous agricultural plants. Knowledge of the most dangerous and widespread pesticide residues, identification of ecological biotopes, are necessary actions of technology for the conservation of bee populations. The maintenance of bee families in comfortable and ecological hives is one of the technological methods that ensure the conservation of the species and race of bees *Apis mellifera* Carpatica. The hives of vertical models have comfort advantages for bees, compared to the horizontal ones, and ensure an increase of the prolificacy of the queens - by 3.5% ($td = 2.07$; $P < 0.05$), of the family power - by 6.0% ($td = 2.41$; $P < 0.05$) and honey production by 19.1% ($td = 5.33$; $P < 0.001$) and an economic efficiency of at least 23.8 euros per family of bees. Feeding bee families during poor harvesting periods in the wild with nutritious supplements, enriched with biologically active substances of different organic origin, contributes to strengthening the vital activity of bee families, ensuring the increase of queen prolificacy and the number of brood capacity by 7.7-45.9%; family power by 9.3-16.9%; flight intensity of bees by 6.8-7.7%; disease resistance by 5.0-8.4%; winter hardiness by 10.5%; the amount of wax raised in the nest by 36.7-39.3%; the amount of pasture with 23.3-27.6% and the amount of honey accumulated in the nest with 19.6-38.9%. Rational use of *Apis mellifera* Carpatica species and bee race can be achieved by exploiting bee families not only for obtaining bee products, but also for their use in the directed pollination of entomophilous agricultural crops that contribute to the increase of fruit harvest in orchards by 15-30% and sunflower seeds with 21.3-36.3%.

Key words: *Apis mellifera* Carpatica, genetic conservation, organic food, pesticides, protection, selection.

INTRODUCTION

Conservation of the bee species *Apis mellifera* in the 21st century has become a particularly current issue due to global climate change, intensifying the impact of anthropogenic activity, especially in modern agriculture (Cebotari et al., 2019b; Cebotari et al., 2021b). A European source (Consequences of climate change, 2018) is mentioned that climate change on Earth is occurring so rapidly that the survival of many plant and animal species is threatened. Many species of terrestrial, freshwater and marine animals have already migrated. Some plant and animal species are at risk of extinction if global average temperatures continue to rise uncontrollably, contrary to the Paris Agreement - the United Nations Framework Convention on Climate Change (2016) and Council Decision (EU) 2016/1841 (2016).

The Bee Decline - Greenpeace Research Laboratories Report (2013) is mentioned that "climate change, such as rising temperatures, changing rainfall patterns, and extreme or more irregular weather events have an impact on pollinator populations. Some of these changes may affect them individually, eventually affecting their communities, which is reflected in the increasing rate of extinction of pollinator species".

For example, it is documented that honey bees in Poland react to climate change by performing the cleaning flight (the time of "waking up" after winter) earlier than usual, in line with the phenomenon generally known as "changing seasons". The bee clearing flight took place a month earlier than the average 25-year observation, which was attributed to increases in atmospheric temperature (Sparks et al., 2010).

Climate change can lead to a change in flowering patterns, a shift in the flowering period of honey plants, which is a major source of food for bees, or a change in the season, in which case the flowering period no longer corresponds to the time when the bees "wake up" spring (Kremen et al., 2007). Due to changing flowering times and patterns of plants, climate change affects the action between pollinators and their food source. Thus, the research of some authors (Memmott et al., 2007) shows that 17-50% of pollinator species suffer from lack of food in the case of realistic climate change scenarios that cause changes in plant flowering patterns. The authors anticipate that the effect of these effects may lead to the potential extinction of both some pollinators and some plants, resulting in disruption of the essential interaction between them.

The invasion of the mite *Varroa destructor* is one of the most dangerous parasitic diseases, which attacks exacerbated the most valuable, from a productive point of view, useful species of insects, such as the bee *Apis mellifera* L., having a fairly accelerated character, with a destructive impact extremely harmful, endangering the existence of bee families (Cebotari et al., 2013). In this context, the protection of honey bees from this invasion by applying targeted selection after natural antivarous resistance and the index of low fertility mites, called SMR - "Suppression of mite reproduction", due to the preferential elimination by bees of breeding mites, renamed and "Varroa sensitive hygiene - VSH", as an inherited trait from parental families, is another important issue in *Apis mellifera* conservation technology (Cebotari et al., 2019c; Mondet et al., 2020).

In this context, research on the identification of bee populations adapted to local climate change conditions, their conservation through methods of selection and genetic improvement of bee families according to the main morpho-productive characters in order to increase their productivity and disease resistance, become scientific approaches particularly important and current (Cebotari, 2006; Cebotari et al., 2015a; Cebotari et al., 2021a).

Of particular concern, both in Europe and around the world, has recently been the main concern for systemic pesticides, which are used

in agriculture to treat seeds and spray crops to control pests and weeds.

According to "Beyond Pesticides" (2014), neonicotinoid pesticides have neurotoxic, reproductive and mutagenic harmful effects on insects, birds, fish, freshwater snails, earthworms, dragonflies, mosquitoes, and vertebrates, noting that "neonicotinoids could represent the new contemporary ecological disaster, being a threat to nature."

According to other sources (Gill et al., 2012), some pesticides, such as the organochlorine insecticide Fipronil, are one of the main chemical factors that are causing the collapse of bee colonies.

Several researchers (Alaux et al., 2010; Henry et al., 2012; Oliveira et al., 2013) have shown that there is a synergism of additive action when pesticides are applied in combination. For example, the neonicotinoid Tiacloprid becomes about twice as toxic to honey bees when used in combination with the fungicide Propiconazole, and three times as toxic - in combination with Triflumizole (Henry et al., 2012). Other research has shown that there is a significant synergy between fungicides, neonicotinoid insecticides and pyrethroids, as well as the acaricides Flumetrin, Cumafos and Fluvalinat (Oliveira et al., 2013).

Along with the interactions of different pesticides, insecticides also show synergies with other stressors, such as parasite infestations. For example, honey bee mortality was higher in those infested with the *Nosema* parasite and a synergistic interaction of factors was found, which reduce the enzymatic activity related to the sterilization of colony food (Alaux et al., 2010).

A particularly important role in the conservation technology of *Apis mellifera* belongs to the conditions of maintenance of bee families. The hive housing bees creates their comfort or discomfort that directly affects the vitality of the colony and, as a result, its productivity (Cebotari et al., 2012). Therefore, the creation of optimal conditions of comfort in ecological hives of vertical type, is an indispensable condition of the technology of conservation of bee populations.

A particularly specific problem is the feeding of bees during poor harvesting periods in the wild. To compensate for the lack of nutrients in the

diet of bees during poor harvesting periods in nature, most beekeepers feed bee families with sugar syrup, which, in addition to carbohydrates, lacks a significant number of biologically active substances. Under these conditions, the consolidation of their power and vigor through balanced feeding methods with nutritional supplements enriched with biologically active organic substances is of major importance (Toderas et al., 2012a, 2012b, 2012c).

The technology of conservation and valorisation of the species *Apis mellifera* becomes complex only if it (the species) is rationally used to pollinate entomophilous crops, contributing substantially to increasing the harvest and economic efficiency of the branches of phytotechnics and beekeeping (Cebotari et al., 2015b, 2015c, 2017). Thus, the development of innovative methods and techniques for bee-directed pollination of entomophilous agricultural crops is a current scientific issue of species valorisation.

Therefore, the targeted selection of bee families in order to increase their adaptability to local environmental conditions, increase their productivity and disease resistance (especially in *Varroa*), protect bee families from pesticide residues, maintain bee families in comfortable vertical beehives, organic feeding of bees during poor harvesting periods in nature, as well as the use of bee families in the directed pollination of entomophilous agricultural crops present, as a whole, the innovative technology of conservation and valorisation of *Apis mellifera* L.

MATERIALS AND METHODS

Scientific research on the selection of bee families has been carried out on the *Apis mellifera* *Carpatica* bee population grown in the experimental apiary of the Institute of Zoology. The selection conservation of the local bee population was carried out by the progressive targeted method according to the independent limits of some important morpho-productive characters of the bee families. The selection was made according to the level of development of the following characters: queen prolificacy, family strength, winter hardiness, viability of brood, disease resistance, honey production accumulated in the nest, rating class. The evaluation of the level of development of

morpho-productive characters was performed, according to the methods developed by us (Cebotari et al., 2010) for the Zootechnical Norm regarding the rating of bee families, breeding and certification of beekeeping breeding material, approved by Government Decision of the Republic of Moldova no. 306 of 28.04.2011 (2011).

Taking into account the biological peculiarities of the bee population (Cebotari et al., 2011), after the results of the assessment of all bee families in the apiary, the best performing families were selected annually to complete the breeding lot, which was later used to produce breeding used for reproduction.

The protection of bee families from pesticide residues was achieved by placing bee families stationary and harvesting in areas (sites) safe and harmless to bees.

Previously, the honey flora in the area intended for location was tested for the residual content of the most dangerous 46 pesticides, of which 22 pesticides monitored by the EU (such as: α -HCH, β -HCH, Lindan, DDT-total, PSV, Aldrin, Dieldrin, Endrin, Hexachlorobenzol, Phenol, Ethylenedibromide, Coumafos, Fluvalinate, Flumetrin, Chlordimeform, Carbandazine, Naphthalen, Amitraz, Cymiazole, Tetramethrin, Glyphosate, Fipronil) and 24 more common pesticides in our country, such as: organochlorine insecticide (Spirodiclofen), organophosphorus insecticides (Azoxystrobin, Carbendazim, Dimethoate, Chlorpyrifos), neonicotinoid insecticides (Imidacloprid, Tiacloprid, Tiametoxam, Clotianidin), pyrethroid insecticides (Tau-fluvalinate, Deltamethrin, Cypermethrin, Pyrethrin), triazole fungicides (Bitertanol, Fenhexamide, Diphenconazole, Mepanipyrim, Cyproconazole), dicarbosimidic fungicides (Cyprinodil), herbicides (Sulfosulfurol, Amidosulfuron, Amitrol, Petoxamide, Pendimethalin). For this, at the beginning of the flowering period of the honey plants, mixed flower samples were taken from the main honey plants, located in the site where the apiary was expected to be placed at harvest. From the flight area of the bees, 5-7 flower samples were taken from different places, at different distances from the place planned for the location of the apiary. Each sample weighed at least 100 g of flowers. The samples were packed in plastic bags and

transported the same day for analysis to the accredited laboratory of the U.S. "Center for Applied Metrology and Certification", in accordance with the Sanitary-Veterinary Norms on the methodology of sampling, processing, packaging and transport of samples for laboratory examinations (2010). The results of the laboratory analyzes, regarding the pesticide residues in flowers, were examined by comparing the data obtained with the maximum admissible limits, according to the Sanitary Regulation on the maximum permitted limits of pesticide residues, approved by Government Decision no. 1191 of 23.12.2010 (2010). If the concentrations of pesticide residues did not exceed the maximum permissible limits, then the environment was considered unpolluted and suitable for organic beekeeping.

In order to determine the comfort level of bees in hives of different types, experimental tests were performed for the maintenance of bee families in hives of horizontal and vertical type (Cebotari et al., 2012).

For the organic feeding of bees during the deficient periods of harvesting in nature by us, a series of nutritional supplements and new procedures for feeding bee families were tested and developed (Toderas et al., 2012a; 2012b; 2012c; Toderas et al., 2014; Toderas et al., 2016a; 2016b; 2016c; 2016d), which ensures the increase of the morpho-productive performances of bee families.

In order to identify the methods of rational use of *Apis mellifera* in the pollination of entomophilous agricultural crops, a series of scientific researches have been undertaken and several techniques (methods) of bee-directed pollination of plum and apple fruit crops have been developed (Cebotari et al., 2015b), as well as sunflower (Cebotari et al., 2015c; Cebotari et al., 2017).

The data obtained as a result of the research were statistically processed using the computer software "STATISTICS - 12" and their certainty was assessed, according to the biometric variational statistics, according to the methods of Плохинский Н.А. (1989).

RESULTS AND DISCUSSIONS

As a result of the multi-annual scientific research carried out in the Apiculture

Laboratory of the Institute of Zoology, a series of innovative procedures (techniques) for genetic amelioration, organic feeding, maintenance and ecological care of bee families, as well as rational use of to the pollination of entomophilous crops, which is, as a whole, the innovative technology of conservation and valorisation of the species *Apis mellifera*.

Conservation of the species *Apis mellifera*.

Among the conservation technologies of the species *Apis mellifera* are part of it: selection and breeding of bees from the local *Apis mellifera Carpatica* purebred population, protection of bee families from pesticide residues, maintenance of bee families in comfortable ecological hives, innovative organic nutrition of bee families in periods of inadequate harvesting, prophylaxis and treatment by ecological methods of diseases and pests of bees.

Conservation by selection of the local population *Apis mellifera Carpatica*.

The technology of conservation of the local genetic fund of bees is realized, according to the National Program for conservation and genetic amelioration of the local bee population in the Republic of Moldova, by selecting and raising them in purebred (Cebotari, 2006).

The selection process provides for a systematic and sustainable annual assessment, over several years, of the level of development of a series of morpho-productive characters. Following the results of the evaluation, the best performing bee families were revealed and the selected batches of breeding were created, intended for the reproduction of beekeeping breeding material.

For reproduction, selected queens of the solocyte type were bred by innovative methods and tested according to the qualities of the descendants (Cebotari et al., 2015d; Cebotari et al., 2018). For the directed mating of queens, the most efficient innovative method was used, such as the instrumental insemination with semen of drones selected from the best performing paternal families.

Research results (Cebotari et al., 2021a) showed that instrumentally inseminated queens have a higher prolificacy compared to their contemporaries naturally mated in the nuptial flight, with 164 eggs / 24 hours, or 10.0% (td = 6.1; P <0.001). Working bees in the families of instrumentally inseminated queens had a higher

development of external morphometric indices compared to their contemporaries in the families of queens naturally mated in the nuptial flight, as shown in: tube length - by 0.19 mm, or 2.9% (td = 6.71; P <0.001), ulnar index of the anterior right wing - 4.3 absolute units, or 10.2% (td = 5.00; P <0.001) and the share of bees with positive discoidal dislocation - by 11.0 absolute units, or by 15.4% (td = 3.10; P <0.01). As a result, the bee families of instrumentally inseminated queens had a significantly higher level of development of morpho-productive characters, compared to their contemporaries of queens naturally mated in nuptial flight, as

follows: colony power - 0.17 kg, or by 6.5% (td = 6.07; P <0.001), disease resistance - by 3.4 absolute units, or by 4.1% (td = 3.15; P <0.01), viability brood - by 4.0 absolute units, or by 4.7% (td = 3.70; P <0.001) and honey production - by 8.39 kg, or by 19.9% (td = 5.31; P <0.001).

An eloquent example of the sustainable selection of bee families can serve the result obtained in the genetic amelioration and purebred conservation of the *Apis mellifera Carpatica* bee population raised in the experimental apiary of the Institute of Zoology during the years 2015-2021 (Table 1).

Table 1. Dynamics of the development level of the morpho-productive characters of the bee families from the experimental apiary in the period 2015-2021

Name of morpho-productive characters	Level of development of morpho-productive characters, M ± m				Standard of the race	Mediate 2015 - 2021	2021, % compared to the standard
	2015 N=50	2017 N=50	2019 N=50	2021 N=50			
Prolificity of queens, eggs / 24 hours	1795±17	1678±16	1716±20	1644±17	1600	1708	102,8
Family power, kg	3.04±0.03	2.36±0.03	2.38±0.03	2.34±0.02	2.33	2.53	100.4
Winter hardiness, %	88.6±0.4	86.8±0.4	71.3±2.1	83.5±0.4	75	82.6	111.3
Brood viability, %	95.8±0.4	95.5±0.2	88.1±0.5	94.3±0.3	80	93.5	117.9
Disease resistance, %	86.3±0.7	92.6±0.6	91.6±0.7	93.1±0.4	60	90.9	155.2
Honey production, kg	44.2±0.6	34.2±0.4	49.9±0.6	57.5±0.6	45	46.4	127.8
Rating class, %:							
El. record + Elite	8.0±3.9	-	22.0±5.9	-		7.5	
Class I	36.0±6.8	-	10.0±4.3	58.0±7.1	100	26.0	58.0
Class II	38.0±6.9	2.0±2.0	12.0±4.6	38.0±6.9		22.5	
Class III	18.0±5.5	42.0±7.1	16.0±5.2	4.0±2.8		20.0	
Extraclass	-	56.0±7.2	40.0±7.0	-		24.0	

The progressive targeted selection, carried out during seven consecutive years, contributed to the consolidation of the performances of the morpho-productive characters selected at the average level of development above the breed standard.

Thus, the prolificacy of queens in the period 2015 - 2021 was on average 1708 eggs / 24 hours, varying depending on climate change of the year, from a minimum of 1644 ± 17 eggs / 24 hours in 2021 to a maximum of 1795 ± 17 eggs / 24 hours in 2015. It should be noted that, after average prolificacy, queen bee families in the experimental apiary exceed the standard breed level by 108 eggs / 24 hours or 6.8% (td = 4.8; P <0.001).

The strength of bee families has been strengthened to an average of 2.53 kg, varying depending on climate change of the year, from a minimum of 2.34 ± 0.02 kg in 2021 to a

maximum of 3.04 ± 0.03 kg in 2015. It should be noted that, according to the average number of bees in the nest (power), the bee families in the experimental apiary exceeded the standard level of *Apis mellifera Carpatica* by 0.2 kg or 10.1% (td = 5, 6; P <0.001).

The winter hardiness of bee families in the selected population has been consolidated at an average level of 82.6%, with variations depending on the climate changes of the year, from a minimum of 71.3 ± 2.1% in 2019, to maximum 88.6 ± 0.4% in 2015. We would like to mention that, after winter hardiness, the bee families in the experimental apiary exceeded the standard level of the *Apis mellifera Carpatica* race by 7.6 absolute units or 10.1% (td = 3.6; P <0.001).

The viability of broods in the bee families of the selected apiary has been strengthened to an average of 93.5%, with variations depending on

the climate change of the year, from a minimum of $88.1 \pm 0.5\%$ in 2019 to a maximum of $95, 8 \pm 0.4\%$ in 2015. It should be noted that, according to the average viability of the brood, the bee families in the experimental apiary exceeded the standard level of the *Apis mellifera Carpatica* race by 13.5 absolute units or 16.9% ($td = 25.0$; $P < 0.001$).

Disease resistance of bee colonies, subject to progressive targeted selection during this period, was maintained at an average level of 90.9%, showing a consecutive increase from $86.3 \pm 0.7\%$ in 2015 to $93.1 \pm 0.4\%$ in 2021. We note that, according to disease resistance, bee families in the experimental apiary exceeded the standard level of the race *Apis mellifera Carpatica* by 30.9 absolute units or 51.5% ($td = 38.1$; $P < 0.001$).

The strengthening of the morpho-productive capacities of the bee families in the selected population at a level above the race standard resulted in the increase, during this period, of the

honey production accumulated in the nest from 44.2 ± 0.6 kg in 2015, until 57.5 ± 0.6 kg in 2021.

We mention that climate change has caused, in some years (2017), considerable decreases in honey production below the standard of the race. Despite these challenges, after the average honey production accumulated in the nest during this period, the bee families in the experimental apiary had an obvious tendency to exceed the standard level of the *Apis mellifera Carpatica* race by 1.4 kg or 3.1% ($td = 1.94$; $P < 0.1$).

Annually, from the population of bee families, evaluated according to the complex of characters, were selected batches of descendants with the most valuable families, which according to the morphological features of the outside corresponded to the requirements of the race standard, and morpho-productive characters significantly exceeded these requirements (Table 2).

Table 2. Dynamics of the development level of the morpho-productive characters of the bee families from the breeding batches in the period 2015-2021

Name of morpho-productive characters	Level of development of morpho-productive characters, $M \pm m$				Standard of the race	Mediate 2015-2021	2021, % compared to the standard
	2015 N=13	2017 N=12	2019 N=8	2021 N=11			
Prolificity of queens, eggs / 24 hours	1846±20	1757±20	1721±44	1727±23	1600	1763	107.9
Family power, kg	3.20±0.04	2.56±0.02	2.36±0.08	2.48±0.02	2.33	2.65	106.4
Winter hardiness, %	89.9±0.7	88.5±0.7	83.0±0.9	84.8±1.0	75	86.6	113.1
Brood viability, %	96.4±0.6	96.1±0.4	87.8±1.1	94.4±0.6	80	93.7	118.0
Disease resistance, %	85.9±1.0	92.0±1.1	92.9±0.8	93.4±1.0	60	91.1	155.7
Honey production, kg	49.6±0.5	38.2±0.3	54.3±1.5	62.3±0.6	45	51.1	138.4
Rating class, %:							
Elite record	-	-	37.5±18.3	-		9.4	-
Elite	30.8±13.3	-	62.5±18.3	-		23.3	-
Class I	69.2±13.3	-	-	100±0.0	100	42.3	100
Class II	-	8.3±8.3	-	-		2.1	
Class III	-	91.7±8.3	-	-		22.9	

According to the average level of development of the morpho-productive characters, the bee families in the breeding batches in the experimental apiary substantially exceeded the races standard: the queen's prolificacy - by 10.2%, the family strength - by 13.7%, the winter hardiness - by 15.5%, at the viability of the brood - by 17.1%, after the resistance to diseases - by 51.8%, at the production of honey - by 13.5%. From the data presented, it can be seen that the genetic value of the bee families in the breeding batches has permanently increased

during this period in terms of disease resistance and honey production.

Thus, the disease resistance of bee families increased from $85.9 \pm 1.0\%$ in 2015 to $93.4 \pm 1.0\%$ in 2021, the increase being 7.5 absolute units or 8.7 % ($td = 5.3$; $P < 0.001$). The honey production of bee families in the breeding flocks increased from 49.6 ± 0.5 kg in 2015 to 62.3 ± 0.6 kg in 2021, the increase being 12.7 kg or 25, 6% ($td = 16.3$; $P < 0.001$). With the exception of 2017 (which had unfavorable climatic conditions), the genetic value of beekeepers of

bee families in these batches, assessed by the complex of morpho-productive characters, is expressed in the highest classes of bonitas (Elite record, Elite and Class I). The average share of bee families of the upper class of rating - Elite-record is 9.4%, Elite - 23.3% and Class I - 42.3%.

It should be noted that the previous results of guided selection of bee families after natural antivirus resistance (Cebotari et al., 2019c) showed that bee families in the brood group significantly outperformed their contemporaries in the experimental batch after queen prolificacy - by 18.4% ($P < 0.05$), the amount of captive brood - by 18.6% ($P < 0.05$) and honey production by 6.8% ($P < 0.01$). At the same time, they tended to have a lower degree of bee infestation with *Varroa* mites, a higher level of colony power and an increased winter hardiness, ranging from 82.3 to 89.0%.

Therefore, the application of progressive targeted selection of bee families with the reproductive use of high value beekeeping material has contributed to the conservation of the *Apis mellifera Carpatica* bee population with increased morpho-productive capacities, resistant to wintering and disease.

Protecting bee families from pesticide residues. This objective is achieved by placing bee families stationary and gathering in areas (sites) that are safe and harmless to bees. The content of harmful residues in honey flowers largely reflects the compliance of the environment for the practice of organic beekeeping and the protection of bee families. Our scientific research (Cebotari et al., 2019a; Cebotari et al., 2021c) has shown that in the samples of acacia and linden flowers collected from the forest site, in the flowers and bee products collected from the industrial lavender fields and sage, as well as those of sunflower and apple, collected from the gardens of households in the rural locality, no detectable values were detected, or pollutants, of residues of some of the 69 pesticides investigated.

Based on the results obtained, it was concluded that forest sites, industrial lavender and sage fields, as well as rural home gardens, do not contain pesticide residues that could affect the health of pollinating insects and have safe areas for the families of pollinators. bees both stationery and harvesting, for the practice of

organic beekeeping with the production of organic bee products.

At the same time, in the same and other similar research of ours (Cebotari et al., 2019a; Cebotari et al., 2020), it was found that in the flowers of honey plants in the industrial orchards of apple and plum, as well as the industrial fields of flower- of the sun, rapeseed, peas and maize, contain residues in detectable concentrations from 8.1 to 37.7% of the 62 pesticides investigated, of which, for some pesticides (Azoxytrobilin, Carbendazim-L, Dimethoate, Glyphosate, Tiametoxam) were recorded residues in concentrations from slightly polluting, exceeding the LMA level by 7-20%, to strongly polluting, exceeding the LMA level by 50-78%. These results show that some sites with orchards and industrial crop fields contain pesticide residues in concentrations that are quite dangerous for the health of bees and the safety of the bee products obtained.

In this context, in order to protect the health of bee families, which are to be located in the harvesting and pollination of apple and plum orchards, as well as industrial crops of sunflower, rapeseed, peas and corn, it is recommended to pre-test the flowers in the respective plantations to the content of the residues of my above-mentioned pesticides. It will also ensure the safety of bee products obtained by harvesting and pollinating these crops.

Keeping bee families in comfortable hives.

Our research (Cebotari et al., 2012) has shown that the development of bee families in different times of the year depends significantly on the comfort of maintenance offered in hives of different types. The test results showed that vertical hives have comfort advantages for bees compared to horizontal ones. The maintenance of bee families in vertical hives ensures an increase in the prolificacy of queens - by 3.5% ($td = 2.07$; $P < 0.05$), of family power - by 6.0% ($td = 2.41$; $P < 0.05$) and honey production by 19.1% ($td = 5.33$; $P < 0.001$). The exploitation of bee families in vertical hives ensures an economic efficiency of at least 23.8 euros per bee family. Comfortable hives must also be environmentally friendly. They need to be made of natural wood (preferred fir), according to current standards. For weather protection, the hives can be painted with linseed oil, or with

special ecological paints. The frames of the hive must also be made of natural wood (fir, lime), and the wires for fixing the honeycombs must be made of stainless steel.

Organic feeding of bees during poor harvesting periods in nature. It is known that at the end of the winter period (February) and the beginning of spring (March-April), bee families face annually the problem of depletion of natural food reserves in the nest. In the body of bees there is a deficiency of bioactive nutrients, especially carbohydrates, proteins, trace elements, vitamins, which play a decisive role in the physiological processes of vital activity of the bee's body, determining the reproductive capacity and further development of the bee family. as a whole (Toderas et al., 2016a; 2016b; 2016c; 2016d).

To compensate for the lack of nutrients in the diet of bees during poor harvesting periods in nature, most beekeepers feed bee families with sugar syrup, which lacks, with the exception of carbohydrates, a significant number of biologically active substances. In order to strengthen the vital activity capacity of bee families during these periods, we have developed a series of procedures for feeding bee families with carbohydrate nutritional supplements, enriched with biologically active substances of different organic origin.

The essence of these processes consists in feeding the bee families during the deficient periods of harvesting in nature with a nutritious carbohydrate supplement, either 50% sugar syrup, or sugar powder cakes mixed with honey in a ratio of 7: 3, enriched with a solution of 1-2% of the biomass extract of the cyanobacterium *Spirulina platensis* - patents: MD 475 Z 2012.09.30 (Toderas et al., 2012a), MD 476 Z 2012.09.30.17 (Toderas et al., 2012b) and MD 477 Z 2012.09.30 (Toderasi et al., 2012c); of the biomass of aquatic microalgae - MD 1061 Y 2016.08.31 (Toderas et al., 2016a), MD 1062 Y 2016.08.31 (Toderas et al., 2016b) and MD 1079 Y 2016.10.31 (Toderas et al., 2016c); or some coordinating organic compounds - MD 850 Z 2015.08.31 (Toderas et al., 2014) and MD 4438 B1 2016.10.31 (Toderas et al., 2016d).

Scientific research has shown that the biologically active substances of the new nutritional supplements contribute to increasing

the prolificacy of queens and the amount of brood per capita by 7.7-45.9%; family power by 9.3-16.9%; flight intensity of bees by 6.8-7.7%; disease resistance by 5.0-8.4%; winter hardiness by 10.5%; the amount of wax raised in the nest by 36.7-39.3%; the amount of pasture with 23.3-27.6% and the amount of honey accumulated in the nest with 19.6-38.9%.

The result is determined by the presence in the nutritional supplements of biologically active substances, such as: amino acids, essential lipid acids, peptides, vitamins (especially B12 and B6), antioxidant pigments and trace elements in necessary quantities, being catalysts of important regeneration functions. queens' ovarian tissue cells, as well as lactating and ceriferous glands of worker bees, with stimulating, immuno-modulatory and antioxidant properties, being a component part of hormones and enzymes in the hemolymph, which contributes to improving the penetrability of organic tissue cells, participates in the process of regeneration of hemocytes and strengthening the body's immune system the first days, the development of the family and the increase of its productivity.

Valorisation of the species *Apis mellifera*. In order to increase the economic efficiency of the beekeeping branch, bee families must be exploited not only for obtaining traditional bee products, but also for the directed pollination of entomophilous agricultural crops.

Multiple researches in the field of entomophilic crop pollination (Cîrnu et al., 1973; Coman, 2012; Curennoi, 1973; Falaleev, 1973; Gerster, 2013; Frediani, 1973; Furgala, 1973; Magdici, 2005; Vaissieres, 2013) have shown that, the free (cross-pollination) pollination of fruit crops with the participation of insects, contributes 20-150% to the total production. In addition, the quality of fruits and seeds resulting from entomophilic pollinated flowers is at least 10-20% higher than those produced from pollinated flowers without insects. Trees with flowers that are poorly pollinated by insects produce fruit with an affected shape, less sweet and with few seeds.

Numerous researches in the field of sunflower (Gerster, 2013; Frediani, 1973; Furgala, 1973), carried out in different countries on different varieties and hybrids, have shown that the pollination of this agricultural crop with the help

of bees contributes to the increase of seed production. with 16-105%.

According to information (Magdici, 2007), in the US 33% of food consumed in this country comes from plants pollinated by insects, of which 75-90% bees, and the total value of crops and goods to which bees contribute by pollination is amounts to about \$ 19 billion.

In the Republic of Moldova, bee pollination of agricultural crops is not widely applied, and some farmers question the effect of this pollination.

In order to highlight the contribution of honey bees to the pollination of agricultural crops and increase their harvest, we have undertaken a series of scientific research in this field.

As a result of the research, some techniques (methods) for bee-directed pollination of plum and apple fruit crops have been developed and proposed for beekeepers and agricultural growers (Cebotari et al., 2015b), as well as sunflower (Cebotari et al., 2015c; Cebotari et al., 2017).

When pollinating plum and apple trees in industrial orchards (Cebotari et al., 2015b).

For saturated pollination of plum and apple orchards, the bee load must be at least 3 families/ha. Each family must have at least 7 ranges of bee frames. The hives with the bee families are placed inside the orchard between the rows of trees, in a row, at a distance of 100 m from each other and over every 7th row of trees. In all areas of the orchard, hives with bee families are placed at the beginning of the flowering period of the trees and kept at least 6 days after the day of placement. In order to speed up the process of accustoming bees to the scent of tree flowers and to increase the intensity of flight, all bee families placed at pollination are fed daily throughout the pollination period, with 50% sugar syrup, mixed with flower infusion, freshly collected from the respective trees, in the amount of 50 g of flowers per 1 liter of syrup. The mixture is administered 50 ml at each interval of bee frames.

It was found that bee-directed pollination of plum and apple orchards by the proposed technique (method) ensures a significant increase, compared to the traditional method of pollination, the frequency of visit of bees to flowers - 2.3-2.4 times, the intensity of bees with pollen clumps - with 23.1-24.5%, of the amount of

pollen collected - with 46.2-57.4%, of the degree of fertilization of flowers - of 2.1-2.2 or, as a result, a significant increase in fruit yield of at least 15-30%.

When pollinating sunflower (Cebotari et al., 2015c; Cebotari et al., 2017). For directed pollination of sunflower, the load of bees must be at least 4 families/ha. Each family must have at least 7 ranges of bee frames. The hives with the bee families are placed in a row around the chain on all four sides, at a proportional distance from each other. The distance between the hives is calculated by dividing the total length of the perimeter around the hive by the number of hives placed.

Experimental results have shown that bee-directed pollination of sunflower crops by the proposed method ensures an increase in the total mass of seeds (harvest) by 21.3-36.3% - compared to the traditional method and 3.6-8.4 times - compared to isolated pollination.

CONCLUSIONS

Sustainable selection of purebred bee families *Apis mellifera Carpatica*, with the application of innovative methods of instrumental insemination of queens, contributes to the preservation of the bee population at the highest level of development of morpho-productive characters and their breeding value, ensuring confidence in the superior quality of the beekeeping parent material proposed for reproduction.

The conservation of bee populations requires their protection from the residues of some pesticides, which are more and more often attested in the flowers of some entomophilous agricultural plants. Knowledge of the most dangerous and widespread pesticides, as well as the identification of clean sites and biotopes of pesticide residues, are necessary actions of technology for the protection and conservation of bee populations.

The maintenance of bee families in comfortable and ecological hives is one of the technological methods that ensure the conservation of the species and race of bees *Apis mellifera Carpatica*. The hives of vertical models have comfort advantages for bees, compared to the horizontal ones, and ensure an increase of the prolificacy of the queens - by 3.5% (td = 2.07; P <0.05), of the family power - by 6, 0% (td =

2.41; $P < 0.05$) and honey production by 19.1% ($td = 5.33$; $P < 0.001$) and an economic efficiency of at least 23.8 euros per family of bees. Feeding bee families during periods of inadequate harvesting in nature with carbohydrate nutritional supplements, enriched with biologically active substances of various organic origin, contributes to strengthening the vital activity capacity of bee families during these periods, ensuring the increase of queens prolificacy and the number of brood 7.7-45.9%; family power by 9.3-16.9%; flight intensity of bees by 6.8-7.7%; disease resistance by 5.0-8.4%; winter hardiness by 10.5%; the amount of wax raised in the nest by 36.7-39.3%; the amount of pasture with 23.3-27.6% and the amount of honey accumulated in the nest with 19.6-38.9%.

Rational use of *Apis mellifera Carpathica* species and bee race can be achieved by exploiting bee families not only for obtaining bee products, but also for their use in directed pollination, through innovative technologies, of entomophilous agricultural crops that contribute to the growth of fruit crops. orchards with 15-30% and sunflower seeds with 21.3-36.3%.

ACKNOWLEDGEMENTS

Scientific research was carried out within the State Program, project 20.80009.7007.12 “Diversity of hematophagous arthropods, zoo and phytohelminths, vulnerability, climate tolerance strategies and development of innovative procedures for integrated control of species of socio-economic interest”, funded from the state budget.

REFERENCES

- Agreement Paris - United Nations Framework Convention on Climate Change (2016). Retrieved 17.12.2018 from <https://eur-lex.europa.eu/content/paris-agreement/html?Locale=ro>
- Alaux, C. et al. (2010). Interactions between *Nosema* microspores and a neonicotinoid weaken honeybees (*Apis mellifera*). *Environmental Microbiology*, 12, 774-782.
- Cebotari, V. (2006). Beekeeping and breeding program in the Republic of Moldova. Achievements and perspectives in animal husbandry. *International Scientific Symposium*, Chişinău, 328-330.
- Cebotari, V., & Buzu, I. (2010). Zootechnical norms regarding the honeybee colonies evaluation, breeding and certification of genetic material in beekeeping. Contemporary Science Association. *Proceedings of the 1st International Animal Health Science Conference: The Beekeeping Conference*, Addleton Academic Publishers, Library of Congress Control Number: 2010918363, p. 26-30.
- Cebotari, V., & Buzu, I. (2011). Biologic particularities of *Apis mellifera Carpathica* bee from Republic of Moldova. In: Actual problems of protection and sustainable use of the animal world diversity. *International conference of zoologists dedicated to the 50th anniversary from the foundation of Institute of Zoology of ASM*, Ed. “Continental Grup”, Chişinău, p. 91-93.
- Cebotari, V., & Buzu, I. (2012). Bee colonies comfort in different types of hives. *Scientific Papers. Series D. Animal Science*, LV, 149 – 153.
- Cebotari, V., Toderaş, I., Buzu, I. et al. (2013). Invasion of the Varroa jacobsoni parasite in the *Apis mellifera Carpathica* bee families. *Bulletin of the Academy of Sciences of Moldova, Life Sciences*, 3 (321), 4-13.
- Cebotari, V., Buzu, I. et al. (2015a). Genetic amelioration of some populations of *Apis mellifera Carpathica* bees from area of forests of Moldova. *Scientific papers. Animal Science*, 64(20), 22-33.
- Cebotari, V., & Buzu, I. (2015b). The plum tree conducted pollination with the bees help. *Scientific Papers Animal Science*, 63(20), 164-172.
- Cebotari, V., Buzu, I. (2015c). The bee families use at sun flower pollination. *Scientific Papers, Animal Science*, 63(20), 173-181.
- Cebotari, V., Buzu, I. (2015d). Testing of the bee queens by the qualities of descendants. *Scientific Papers. Series D. Animal Science*, LVIII, 32-41.
- Cebotari, V., Buzu, I., Gliga, O. et al. (2017). Estimation of the efficiency of pollination by bees of sunflower culture for hybrid seed production. *Scientific Papers. Series D. Animal Science*, LX, 212-216.
- Cebotari, V., & Buzu, I. (2018). Efficiency of bee queen rearing depending on organization way of the nest in cell starter colonies. *Scientific Papers. Series D. Animal Science*, LXI(2), 124-131.
- Cebotari, V., Buzu, I., & Postolachi, O. (2019a). Pesticide residues in melliferous flowers from sites with diverse anthropic impact. *Scientific papers. Animal Science*, 71(24), 123-131.
- Cebotari, V., Buzu, I., & Postolachi, O. (2019b). Impact of climate change of air temperature on vital activity of the bee families. *Scientific Papers. Series D. Animal Science*, LXII(1), 249-255.
- Cebotari, V., Buzu, I., Postolachi, O., Siceanu, A., & Căuia, E. (2019c). Particularities of natural varroa-resistance of honey bees population of *Carpathian* race from the Codri of the Republic of Moldova. *Scientific papers. Series D. Animal Science*, LXII(1), 27-35.
- Cebotari, V., & Buzu, I. (2020). Conformity of rapi, peas and maize flowers, concerning pesticide residues for organic beekeeping. *Scientific Papers. Series D. Animal Science*, LXIII(1), 415-421.
- Cebotari, V., & Buzu, I. (2021a). The morpho-productive particularities of queens *Apis mellifera Carpathica* inseminated instrumentally. *Scientific Papers. Series D. Animal Science*, LXIV(2), 25-34.

- Cebotari, V., & Buzu, I. (2021b). Impact of climate change of atmospheric precipitations on the vital activity of bees families. *Scientific papers - Animal Science Series*, 75(26), 87-97.
- Cebotari, V., Buzu, I. (2021c). Conformity of lavender flowers and sunflower on pesticide residues for organic beekeeping. *Scientific papers. Animal Science*, 76(27), 145-154.
- Cîrnu, I., & Cociu, V. (1973). The efficiency of entomophilic pollination in apples. Bee pollination. Bucharest, RO: Apimondia Publishing House, 113-118.
- Consequences of climate change (2018). Retrieved 17.12.2018 from <https://ec.europa.eu/clima/change/consequences.ro>.
- Coman, R. (2012). Plant pollination. In: Beekeeping, ACA, ICDA, Ploiești, RO: LVS CREPUSCUL Publishing House, 49-55.
- Curennoi, N.M. (1973). The importance of honey bees for the regular fruiting of the apple. Bee pollination. Bucharest, RO: Apimondia Publishing House, p. 137-140.
- Decline of bees. Technical report of Greenpeace research laboratories (2013), 48 p.
- EU Council Decision 2016/1841. JOL 282, 19.10.2016, p. 1-3.
- Falaleev, N.A., et al. (1973). Increasing sunflower production in East Kazakhstan through pastoral movements of apiaries for harvesting and pollination. In Pollination with bees. Bucharest, RO: Apimondia Publishing House, p. 230-233.
- Frediani, D. (1973). The role of bees in sunflower (*Helianthus annuus* L.) pollination in Central Italy. In Bee pollination. Bucharest, RO: Apimondia Publishing House, p. 228-230.
- Furgala, B. (1973). Sunflower pollination - a neglected field of research (in USA). In Bee pollination. Bucharest, RO: Apimondia Publishing House, p. 233-237.
- Gerster, F. (2013). Plan for the sustainable development of beekeeping in France (Part VII). *Beekeeping*, 10, 6-7.
- Gill, R. et al. (2012). Combined pesticide exposure severely affects individual and colony-level traits in bees. *Nature*, 491, 105-108. doi: 10.1038/nature11585.
- Henry, M., et al. (2012). A Common Pesticide Decreases Foraging Success and Survival in Honey Bees. *Science*, 1215039 Published online 29 March 2012 [DOI:10.1126/science.1215039].
- <http://www.beyondpesticides.org/programs/bee-protective-pollinators-and-pesticides/chemicals-implicated>. Retrieved 22.06.2016.
- Kremen, C., Williams, N., Aizen, M. et al. (2007). Pollination and other ecosystem services produced by mobile organisms: a conceptual framework for the effects of land-use change. *Ecology Letters*, 10, 299-314.
- Magdici, M. (2005). Pollination with bees or poverty. *Beekeeping Romania*, 6, 14-15.
- Memmott, J., Craze, P., Waser, N. et al. (2007). Global warming and the disruption of plant-pollinator interactions. *Ecology Letters*, 10, 710-717.
- Mondet, F., Căuia, E., Cebotari, V., et al. (2020). Evaluation of suppressed mite reproduction (SMR) reveals potential for *Varroa* resistance in European honey bees (*Apis mellifera* L.). *Journal insects*, 11, 175-192.
- Oliveira, R.A. et al. (2013). Side-effects of thiamethoxam on the brain and midgut of the Africanized honeybee *Apis mellifera*. *Environmental Toxicology*, 11, 125-132.
- Sanitary regulation regarding the maximum allowed limits of residues of phytosanitary products from / or on food and feed of plant and animal origin, approved by the Government Decision of the Republic of Moldova no.1191 of 23.12.2010. (O.M. nr.5-14 from 14.01.2011, art.03).
- Sanitary-veterinary norm regarding the taking of official samples from live animals and products of animal origin, approved by the Decision of the Government of the Republic of Moldova no. 782 of 01.09.2010 (O.M. nr. 160-162 from 07.09.2010, art. 871).
- Sparks, T., Langowska, A., Glazaczov, A. et al. (2010). Advances in the timing of spring cleaning by the honeybee *Apis mellifera* in Poland. *Ecological Entomology*, 35, 788-791.
- Toderaș, I., Rudic, V., Cebotari, V., Buzu, I. et al. (2012a). *Process for feeding Apis mellifera bee families*. Patent no. MD 475 Z 2012.09.30. AGEPI of the Republic of Moldova. BOPI, nr. 2, Chișinău, p. 28.
- Toderaș, I., Rudic, V., Cebotari, V., Buzu, I. et al. (2012b). *Process for feeding Apis mellifera bee families*. Patent no. MD 476 Z 2012.09.30. AGEPI of the Republic of Moldova. BOPI, nr.2, Chișinău, p. 28-29.
- Toderaș, I., Rudic, V., Cebotari, V., Buzu, I. et al. (2012c). *Process for feeding Apis mellifera bee families*. Patent no. MD 477 Z 2012.09.30. AGEPI of the Republic of Moldova. BOPI, nr.2, Chișinău, p. 29-30.
- Toderaș, I., Cebotari, V., Gulea, A., Buzu, I. et al. (2014). *Process for feeding bee families Apis mellifera (Co, Bi)*. Patent no. MD 850 Z 2015.08.31. AGEPI of the Republic of Moldova. BOPI, nr.12, Chișinău, p.29-30.
- Toderaș, I., Cebotari, V., Ungureanu, L., Buzu, I., Gheorghită, C. (2016a). *Process for feeding Apis mellifera bee families*. Patent no. MD 1061 Y 2016.08.31. AGEPI of the Republic of Moldova. BOPI, nr.8, Chișinău, p. 33-34.
- Toderaș, I., Cebotari, V., Ungureanu, L., Buzu, I., Gheorghită, C. (2016b). *Process for feeding Apis mellifera bee families*. Patent no. MD 1062 Y 2016.08.31. AGEPI of the Republic of Moldova. BOPI, nr.8, Chișinău, p. 34.
- Toderaș, I., Cebotari, V., Ungureanu, L., Buzu, I., Gheorghită, C. (2016c). *Process for feeding Apis mellifera bee families*. Patent no. MD 1079 Y 2016.10.31. AGEPI of the Republic of Moldova. BOPI, nr.10, Chișinău, p. 29.
- Toderaș, I., Cebotari, V., Gulea, A., Buzu, I. et al. (2016d). *Process for the production of the compound [tetraoxoethylenediaminetetraacetate dimolybdenum V] bis- (tetrafenylphosphonium) di-semihydrate and process for feeding bees with its use.*

Patent no. MD 4438 B1 2016.10.31. AGEPI. Bul. Of. of Propr. Intellectual, nr. 10, Chişinău, p. 22-23.

Vaissicres, B. (2013). Bees and pollination. *Beekeeping Romania*, 8, 22-24.

Zootechnical norm regarding the crediting of bee families, breeding and certification of beekeeping breeding material, approved by the Decision of the

Government of the Republic of Moldova no. 306 from 28.04.2011 (O.M.nr. 78-81 from 13.05.2011, art. 366).

ПЛЮХИНСКИЙ, Н.А. (1989). *Руководство по биометрии для зоотехников*. Изд. «Колос», Москва, 256 с.