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USE OF CHROME TRACE FOR VITAL ACTIVITIES FUNCTIONS STIMULATION OF APIS MELLIFERA BEE COLONIES

Valentina CEBOTARI[1], I. TODERAS[1], I. BUZU[1], V. RUDIC[2]

[1]Zoology Institute of Science Academy from Moldova, 1, Academiei street, MD 2028, Chisinau, Republic of Moldova, Chisinau, Moldova
[2]Microbiology and Biotechnology Institute of Science Academy of Moldova, 3/2, Academiei street, Chisinau, Republic of Moldova, Chisinau, Moldova

Corresponding author email: valentinaceb@yahoo.com

Abstract

The goal of the work was to determine the biostimulation effect of a nutritional supplement of sugar syrup enriched with biomass extract of cyanobacteria Spirulina platensis strain, grown on remedy of chromium alum and potassium, on vital activities functions of Apis mellifera bee colonies. In special experiences has been tested in bees food, an energy-mineralo-protein nutritional supplement „Apispir+Cr", composed of sugar syrup in proportion 1:1, enriched with biomass extract of cyanobacteria Spirulina platensis strain, grown in the presence of coordinate compound of chromium and potassium alum-KCr (SO4) 2·12H2O administered in nutrient medium componence, in the amount of 30...35 mg/L in the first three days of cultivation. The extract of the cyanobacteria Spirulina platensis stem biomass has been obtained by repeated extraction of biologically active substances with alcohol solution and solution of NaOH, by centrifuging, by combinig the supernatants and by making the dialysis up to pH 7.5...8.5 with getting the final extract. Nutritional supplement 'Apispir+Cr' was tested on two similar batches with bee colonies, with 10 families in each batch, including:-Ist batch-witness, where the bees were fed with sugar syrup 1:1, in amount of 100...130 mL to a frame with bees, at every 2 days during two weeks;-II batch-experimental, where bees were fed with sugar syrup 1:1, in the same amount, enriched with the 'Apispir+Cr' supplement under 500:1, and syrup with extract. In early may, before the base picking, were examined the main morphoproductive characters and qualities of bee colonies. Test results have shown that feeding the bees with a nutritional supplement 'Apispir+Cr' in early spring, during the poor harvest in nature, contributed to a significant increase in the value of the main morphoproductive characters of bee colonies. Thus, bee colonies from experimental batch, who received in food the nutritional supplement enriched with 'Apispir+Cr', exceeded their fellows: after the prolificity – with 1390 eggs/24 hours, or with 74.4% ($P < 0.001$), quantity of covered brood-with 167 hundreds of cells, or with 74.5% ($P < 0.001$), the power of the family-with 0.56 kg or 25.3% ($P < 0.001$), resistance to disease-with 10.8 percentage, or 13.4% ($P < 0.001$), the amount of wax accumulated in nest-with 1.10 kg, or with 8.,6% ($P < 0.05$), the amount of honey in the nest-2.44 kg, or 90.0% ($P < 0.001$), the quantity of pasture accumulated in the nest-with 36.4 hundreds of cells, or with 106.7% ($P < 0.001$).

Key words: *biostimulation, „Apispir+Cr", productivity, bees, families.*

INTRODUCTION

After the winter, bee colonies are usually convalescent, as a result of the various weather's action.

In Republic of Moldova, at poor periods harvest in nature, especially, early spring (March-April), before the main harvest, in feeding bees persist a deficiency of complete nutrients, such as protein, vitamins, trace elements etc. In the bees body, there is a deficiency of essential indispensable nutrients, which stagnates their vital activity and inhibit subsequently, the food accumulation processes in the nest.

A special role has feeding bees with micronutrients, such as bioactive catalysts, which determines the enzymes activity and serve as a substrate for cell regeneration at living organisms. Microelements influence refers also to digestion processes and nutrients assimilation. Particularly important are the trace elements for the oxygen transport, regulating the body hydrological regime, dissimilation products neutralizing as a result of oxidation processes.

Of trace elements, a special role has the chromium as a biologically active substance. This is contained in bigger quantity in beekeeping products, especially, in Royal Jelly and bees' bread.

Minerals, including trace elements, get into the bee body through water, pollen, nectar. The presence of trace elements in these bee foods determines the vital activity of the body, and their content in beekeeping products.

To fill the deficit of biologically active substances, including trace elements, in bee foods and unlocking their vital activity processes, during poor periods of harvest, beekeepers apply different procedures and stimulating means of bees vital functions (Tuktarav et al., 2010; Еремич, 1986; Кузин, 2003; Панин, 2001; Таранов, 1986).

It is known an accelerating method of bee colonies development, which consists of feeding them with sugar syrup, enriched with trace elements introduced in form of salts and $COCl_2$, $MnSO_4$, proteins in form of pollen collected from bees balls, calcium caseinate, made from skimmed cow's milk and medical growth stimulators, containing sulphonamides and antibiotics. The disadvantages of this technique are that it is expensive, and the milk proteins and trace elements introduced in form of salts are hard digestible for bees, and being easily oxidable, they cause disturbances in the digestive tract of their functions. Moreover, according to EU and national rules, the use of pharmaceuticals containing sulphonamides and antibiotics, in the treatment, prevention and stimulating the bees, is strictly prohibited.

The above problems can be solved in part, through the use in bees food of nutritional supplement of sugar syrup 1:1 enriched with the biomass extract of cyanobacteria Spirulina platensis stem CNB-CB-02 (Bulimaga, 2006; Rudic et al., 2004; Toderaş et al., 2003). Biomass extract of this stem has a wide spectrum of biologically active substances, but, at the same time, contains little protein and trace elements, particularly, chromium.

In this context, the development and testing of new nutritional supplements for bees food in poor harvest periods is an actual problem.

MATERIALS AND METHODS

At the experimental apiary of Zoology Institute, in collaboration with the microbiology and biotechnology Institute of Science Academy from Moldova, was developed and tested in feeding bees with a mineral energo-protein nutritional supplement, composed of sugar syrup in proportion of 1:1, enriched with the biomass extract of cyanobacteria Spirulina platensis stem, grown in the presence of coordinativ compound of chromium and potassium alum - $KCr(SO_4)_2 \cdot 12H_2O$, administred in composition of nutrient medium, in amount of 30...35 mg/L in the first three days of cultivation, as a result, the biomass gained a high content of biologically active substances, including trace elements, particularly of Cr (Toderaş et al., 2012).

The biomass extract of cyanobacteria Spirulina platensis stem has been obtained by extraction of biologically active substances with alcoholic solution of 20...30%, by its shaking and centrifugation with separation of supernatant sediment. After this, the sediment was parched at the temperature of 40...45^oC, was extracted with 0.45% NaOH solution by shaking for 60 min, centrifugation, sediment separation, and repeated extraction of biologically active substances with 0.45% NaOH solution by shaking it for 30 min, centrifugation, adding of obtained supernatants and making the dialysis up to pH 7.5...8.5 with obtaining of final supplement.

This extract, called by us 'Apispir + Cr', presents a liquid green color with a yellowish tinge, whose dry substance contains 55...65% protein, contained the entire set of essential and non essential amino acids. The extract contains as a bioactive component part, chromium in amount of 0.2...0.4%, one of the main catalysts and antioxidant element of regenerative processes of lacto sinthesing and reproductive tissues' cells of the bees.

To estimate the efficiency of this nutritional supplement 'Apispir+Cr', during the poor harvest period (early April) have been carried out comparative testing experiences on bee colonies from two similar batches, with 10 families in each batch, of which:

- Ist batch – witness, where bees was receiving as food sugar syrup 1:1, in amount of 100...130

mL at one bee comb, each 2 days**,** during 2 weeks.

- IInd batch – experimental, where the bees received as food sugar syrup 1:1 in same amount, enriched with „Apispir+Cr" supplement in proportion of 500:1, respectively syrup and extract.

In early may, before the main harvest, were studied the main morph productive characters and features of bee colonies, in accordance with zootechnic Norm regarding bee colonies evaluation, raising and certification of beekeeping genitor material.

The data obtained in the experiments were processed statistically using computer software 'STATISTICS-6' and evaluated their certainty, according to variational biometric statistics, after the methods of Плохинский Н. А., 1969.

RESULTS AND DISCUSSIONS

Test results have shown that feeding bees with the nutritional supplement 'Apispir+Cr' in early spring, during the poor harvest in nature, contributed to a significant increase in the value of the main morph productive characters of the bee families (table).

Thus, if at he beginning of experiences, the queens' prolificacy of bee colonies from both batches was at the same level, then over a month, this character at bee colonies from IInd batch, which received in food the nutritional supplement enriched with 'Apispir+Cr', has increased significantly compared to the witness batch, with 1,390 eggs/24 hours, or with 74.4% ($P < 0.001$).

Same growth regularities was observed also at the covered brood quantity. It was found that this character has increased directly proportional with the prolificacy, surpassing the witness batch with 167 hundreds of cells, or with 74.5% ($P < 0.001$).

Increased activity of the bees breeding functions from IInd batch, compared with the witness batch, has led to increasing the amount of working bees in the nest. Thus, the bee colonies from IInd batch, which received as food nutritional supplement enriched with 'Apispir+Cr', exceeded significantly the bees from witness batch by their strength with 0.56 kg or 25.3% ($P < 0.001$).

It was established that feeding bees with the nutritional supplement enriched with 'Apispir+Cr' contributes to strengthening their resistance to disease, observed from the standard test of hygienic instinct of dead brood removal. Thus, disease resistance of bee colonies fed with the nutritional supplement enriched with 'Apispir+Cr' increased compared to the witness batch, with 10.8 percentual units, or with 13.4% ($P < 0.001$).

Results of research demonstrate that the nutritional supplement enriched with 'Apispir+Cr, administered in bees' food, exerts a general cumulative influence on their vital activity functions. It was found that at worker bees from IInd batch, the wax glands significantly are increased, compared to the bees from witness batch. Thus, the wax quantity filed by the bee colonies from IInd batch, was significantly higher, compared to the witness batch, with 1.1 combs, or with 84.6% ($P < 0.05$).

Morph productive performance of bee colonies from experimental batch compared to the witness batch, are reflected more obvious in the diagram (fig.), in which the level of morph productive characters' value of bee colonies from witness batch is located on the abscissa conventionally at order 0 (zero) percent.

Table 1. Morph productive characters′ values of bee colonies from experimental batches

Specification	Ist batch $M_1 \pm m_1$	IInd batch $M_2 \pm m_2$	$M_2 - M_1$	% compared to Ist batch	td
Queens prolificacy, eggs/24 hours	1868 ± 117	3258 ± 101	1390	174.4	8.99^{***}
Quantity of covered brood, hundreds cells	224.0 ± 14.1	391.0 ± 12.1	167	174.5	8.99^{***}
Colonies′ strength, kg	2.21 ± 0.08	2.77 ± 0.08	0.56	125.3	4.95^{***}
Resistance to disease,%	80.4 ± 1.0	91.2 ± 1.0	10.8	113.4	7.66^{***}
Wax quantity, built combs	1.30 ± 0.33	2.40 ± 0.31	1.10	184.6	2.43^{*}
Quantity of honey, kg	2.71 ± 0.30	5.15 ± 0.21	2.44	190.0	6.67^{***}
Quantity of bee bread, hundreds cells	34.1 ± 4.25	70.5 ± 3.53	36.4	206.7	6.59^{***}

Remark: **B > 0.95 (P < 0.05);** *** **B > 0.999 (P < 0.001).**

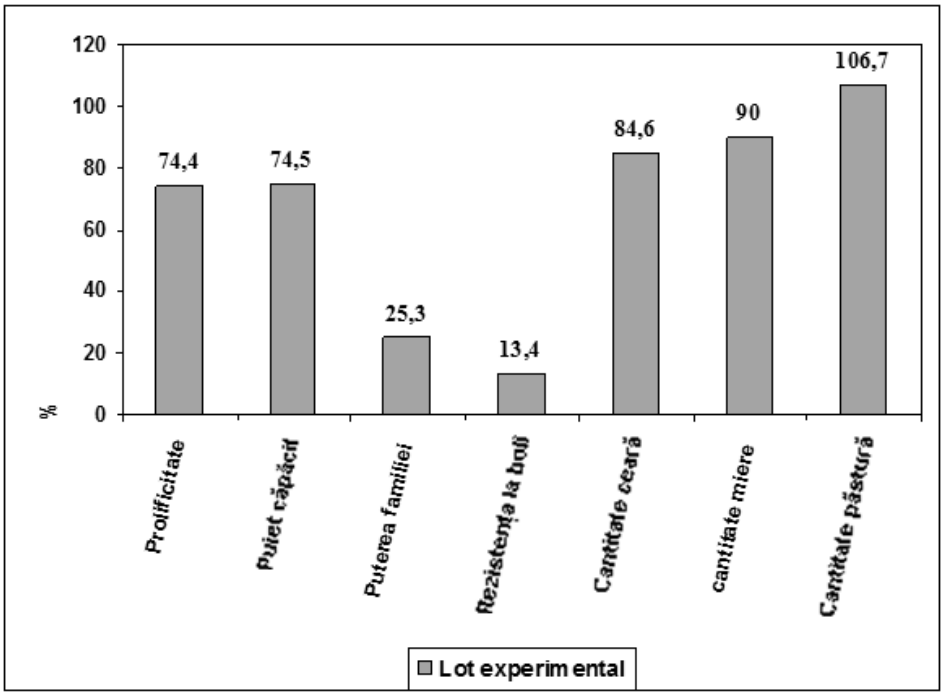

Figure 1. Morph productive performances of bee colonies from experimental batc, compared to witness batch

Due to activation of physiological functions of queens' and working bees' bodies, bee colonies from IInd batch, which received in food the nutritional supplement enriched with 'Apispir+Cr', have accumulated higher production volumes in the nest. Thus, the quantity of honey accumulated in nest by bee colonies from IInd batch was significantly higher, compared to bee colonies from witness batch, with 2.44 kg, or 90.0% ($P < 0.001$). Also, the amount of bee bread accumulated in nest by bee colonies from Iind batch was higher compared to those from witness batch, with 36.4 hundreds of cells, or with 106.7% ($P<0.001$).

Generalized result achieved, as a result of feeding bee colonies with nutritional supplement enriched with 'Apispir+Cr', is due to the presence in it, of biologically active substances, such as: amino acids in increased quantity, peptides, vitamins, pigments and trace elements, in particular, chromium in increased quantity, as catalysts of some important functions of regeneration of the queens' ovarian tissues cells, and of lactogene glands of worker bees. Chromium has high stimulating and antioxidant properties and is part of queens' milk composition, determining the quality and the level of permanent nutrition of the queen, and of brood larvae in first days.

All this has led to the intensification of the vital physiological functions of working bees, in particular, of queens prolificacy, bee colonies' resistance to disease, wax activity of working bees, which have contributed to increase of production volumes accumulated in the nest.

CONCLUSIONS

1. To fill the deficit of complete nutrients in feeding bees during poor harvest period in nature, can be used as a nutritional supplement, composed from sugar syrup in proportion of 1:1 with 'Apispir+Cr (patent MD 476 Z 2012.09.30).

2. Biologically active substances contained in the nutritional supplement 'Apispir+Cr', especially chromium, helps to stimulate the vital activity of bees, accelerating the reproduction rate of brood, increase of bee colonies strength, their resistance to disease and ability to accumulate production in the nest.

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