Scientific Paper

Potentialities of the utilization of zootechnical additives in Cuban apiculture

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Abstract

Objective: To evaluate the potentialities of the utilization of zootechnical additives in apiculture in Cuba.

Materials and Methods: Papers available in online data bases (PubMed, Scopus, Web of Science and Google Scholar), related to the use of zootechnical additives in honey bees (*Apis mellifera*), were reviewed. A search was made for the keywords *zootechnical additives*, *probiotics*, *prebiotics* and *phytobiotics*, *plant extract*, and then the word *bees* was added. All the searches were carried out between 2019 and 2020. There was no exclusion of any period and no language restrictions were applied.

Results: The utilization of antibiotics in the control and prevention of bacterial infections in apiculture causes changes in the rearing of this species, and thus their use is limited in many countries. In the face of this reality, the use of zootechnical additives constitutes an alternative, due to their capacity to modulate the immunological system and the intestinal microflora in bees; besides having an antagonist function against pathogens. In the last decades it is noted that probiotics and plant extracts are the most widely used to improve health and productive indicators, for which work is done to achieve their standardization in Cuba.

Conclusions: Obtaining and evaluating zootechnical additives in bees is a growing strategy worldwide, which is focused on minimizing or eliminating the use of chemicals. In Cuba, the use of these additives constitutes a sustainable proposal to improve health indicators, and thus maintain the quality of honeys.

Keywords: bees, plant extracts, prebiotics, probiotics

Introduction

The bees of the species *Apis melífera* are among the most important insects of the earth ecosystem, because they act as pollinators of different plant species, which allows to sustain infinite crops that are part of man's trophic chain (Pufal *et al*., 2017; Hung *et al.,* 2018).

The increasing demand for honey and the development of technologies for their exploitation favor the growth of apicultural production in recent years (Magaña *et al*., 2016), which in Cuba represents one of the exportable items of the Agroforestry Entrepreneurial Groups of the Ministry of Agriculture (ProCuba, 2019).

The average apicultural production of the last ten years oscillates around 8 000 t of honey. In 2019 six thousand 900 t were exported, from the activity of 204 000 beehives, with average yield of 49 kg per hive. However, the existing potential is much higher, for which work is done on a development program that includes important investments, among which are bee breeding, health, enhancement of melliferous flora and diversification of products in the search for higher added value, stand out (Pérez-Piñeiro, 2017).

Approximately 70 000 beekeepers, associated to the diverse state and non-state productive forms, are exposed to the effects of climate change, intensive agriculture, pests and agrochemicals. In Cuba the situation is different from other countries; yet, the ecosystemic resources should be maintained and the chain should be prepared to act against the influence of these factors and not lose the quality and innocuousness of products, because the possibility that they are produced in some of the zones of higher exposure is not excluded (Larson *et al*., 2020).

At present, many pathogen agents, such as viruses, fungi, bacteria and protozoa, threaten bee colonies. In the world strategies are used that include the use of antibiotics and pesticides to control or minimize the effect of such pathogens. Nevertheless, the negative consequences of the residues of these substances in the food are known,

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for which they constitute a great concern and threat for human health in the planet (Aidara-Kane *et al*., 2018) and, in turn, for farmers they represent the challenge of surviving in the market of foodstuffs.

In the face of these conditions, an alternative would be to avoid the use of antimicrobials as growth promoters or routine prophylactics (Aidara-Kane *et al*., 2018). Among the strategies to reduce their utilization improved management practices and the use probiotics and medicines that increase the capacity of the immune system to face pathogen agents, are proposed. The natural and organic variants constitute the most interesting options, as well as the microorganism species and plant-derived products, capable of promoting growth, improve the health of farm animals and control diseases, among which bees are included (Tonello, 2019). Hence the objective of this study was to evaluate the potentialities of the use of zootechnical additives in apiculture in Cuba.

Materials and Methods

Papers available in online databases were reviewed (PubMed, Scopus, Web of Science and Google Scholar), linked to the use of zootechnical additives in honey bees (*Apis mellifera*). The search was carried out from the key words *zootechnical additives*, *probiotics*, *prebiotics*, *phytobiotics*, *plant extract*, which were combined with *bee*. All the searches were carried out between 2019 and 2020. There was no exclusion of periods during the quest, and no language restrictions were applied. The abstracts of all the papers were carefully read and studies that evaluated the action of zootechnical additives on bee health and the safety protocols for a zootechnical additive candidate, were included.

Generalities of additives

Additives are substances that, intentionally incorporated to feedstuffs, can influence positively their characteristics or animal production. They can be divided into five categories (Rodríguez, 2019):

i. *Technological additives*. They are products added to the diets with technological purposes, which include preservatives, antioxidants, anticaking agents, stabilizers, gelling agents, binders, substances for the control of contamination by radionuclides, acidity regulators, additives for silage and denaturants.
ii. *Sensorial or organoleptic additives*. They are substances added to foodstuffs with the intention of improving or modifying their organoleptic properties or visual characteristics, for example: colorants (they add or return color to food) and flavoring agents (they increase the smell or palatability of foodstuffs).
iii. *Zootechnical additives*. They are capable of influencing positively the improvement of animal performance. Among the additives there are different functional groups: the digestive ones, which facilitate the digestion of foodstuffs by acting on certain raw material, such as enzymes; stabilizers of the intestinal flora, which are those microorganisms or substances that have a positive effect on the intestinal flora, like probiotics, prebiotics, symbiotics and organic acids, among others.
iv. *Nutraceuticals or functional foods*. They are foodstuffs and components that provide benefits to health and participate in the prevention and control of diseases; besides fulfilling the traditional nutritional requisites. Vitamins, minerals, amino acids and oligoelements are included among the functional groups.
v. *Coccidiostatic and histomonostatic additives*. They constitute medicinal substances that are used for the prevention of coccidiosis widely used in diets aimed at young monogastric animals.

Main zootechnical additives and their utilization in bees

The strategies that can be implemented to reduce the use of antibiotics in apiculture are varied. The effect of zootechnical additives on the yield and health of animals, among which bees are included, is known, because they gain increasingly more attention due to their beneficial role. Among the additives that are given priority in bees are probiotics, prebiotics and phytobiotics (Maruščáková *et al*., 2020), which help stimulate the immune system and regulate the intestinal microbiota, thus reducing the negative impacts of the first larval phases, and other environmental challenges faced by the insect.

Use of probiotics in apiculture

The World Health Organization defined probiotics as microorganisms that administered alive and in adequate quantities, confer benefits to the host's health. Nevertheless, in the field of animal nutrition, especially for farm animals, probiotics are used to protect the animals against specific pathogen bacteria, besides having beneficial effects on the yield of the species (Markowiak and Śliżewska, 2018).

In general, lactic acid bacteria are used as probiotics, including lactobacilli, streptococci, bifidobacteria, enterococci and fungi species, such as *Saccharomyces* and *Aspergillus*. Normally, the action mechanisms of probiotics are the modulation of the balance of microbiota in the gastrointestinal tract, improvement of digestion, absorption of nutrients and stimulation of immunity to maintain animal health. Through competitive inclusion these microorganisms secrete substances that inhibit growth or kill and alter the gene expression of pathogen agents.

The probiotics used in apicultural production and health are included, mainly, as supplement in the syrup and pollen, and the genus *Lactobacillus* is the most widely used one (Mudroňová *et al*., 2011). In *in vitro* tests the inhibiting effect of *Bacillus* sp. strains, isolated from samples of honey and bee intestine against larvae of *Paenibacillus larvae* and *Ascosphaera apis*, is reported (Audisio, 2017).

In essay with *Lactobacillus johnsonii* CRL1647, isolated from the intestinal tract of bees and selected for its high production of lactic acid, as monoculture to test the behavior of the insect colonies, has been reported. The strain was administered during three months in the syrup and significant differences were found in the open and sealed brood areas in the treated group with regards to the control. In addition, regarding the initial number, higher percentage of bees was found in the treated group (54 %) than in the control (18 %). Likewise, the honey harvest was higher (40 and 19 %) for the treated groups with regards to the control, respectively (Audisio and Benítez-Ahrendts, 2011).

With a similar scheme, through the strain *B. subtilis* subsp. *Subtilis,* Audisio (2017) found similar beneficial results. The counts of *Nosema* spp. and *Varroa* spp. spores in the treated beehives were lower than in those of the control group. These results in the experimental apiaries indicate that *B. subtilis* subsp. *subtilis* Mori2 favored the performance of bees. First, because the microorganism stimulated oviposition by the queen, which was translated into a higher number of bees and, consequently, more honey. Second, because it reduced the prevalence of two important diseases of bees throughout the world: nosemosis and varroasis.

Strains of bifidobacteria (AcjBF), isolated from the intestinal tract of the Japanese bee *Apis cerana japonica*, were studied for their possible applications as probiotic agent against *Melissococcus plutonius*, the causative agent of the European foulbrood (EFB). The results of the *in vitro* inhibition essays revealed that the free supernatants of cells from all the AcjBF isolates, exhibited antagonist effects on the growth of *M. plutonius*, for which they could be potentially used as a new natural biological agent to control the EFB (Wu *et al*., 2014). Nevertheless, no beneficial effect was found in field trial with specific lactic acid bacteria (hbs-LAB) of bees to control *P. larvae*, in spite of having effect on individual insects (Stephan *et al*., 2019).

In supplementation of colonies with beneficial bacteria such as *Lactobacillus plantarum* Lp39 improvements were obtained in the innate immune response and resistance to opportunistic infections, which suggests that these bacteria could be useful to approach directly some of the pathogen agents involved in the decrease of the populations of honey bees (Berríos *et al*., 2018).

In field essays and controlled experiments in laboratory, Daisley *et al*. (2020) referred that a consortium of lactobacilli strains, isolated from the intestinal tract, can improve the survival of honey bees to the infection by *P. larvae*, directly inhibit the *P. larvae* cells *in vitro*, and modulate beneficially innate immunity and other response genes of the host during the experimental infection. Although it was observed that the evaluated lactobacilli in this study are beneficial under infectious conditions, more essays will be needed to determine their long-term impact on beehives of healthy bees.

Daisley *et al*. (2020) evaluated the effect of a probiotic additive of lactobacilli (BioPatty) in beehives infected with American foulbrood (AFB). These authors proved with their results that in the treated beehives there was a significantly lower pathogen burden when comparing them to the control. The above-cited authors used bee larvae in experiments at laboratory level and proved that the same bacterial consortium, integrated by *Lactobacillus plantarum* Lp39, *Lactobacillus rhamnosus* GR-1 and *Lactobacillus kunkeei* BR-1 (contained in BioPatty), reduced the population of pathogens, increased the expression of key immunity genes, and improved the survival of larvae during the infection by *P. larvae*.

In field experiment, the application of a mixture of microorganisms with probiotic potential, isolated from the intestine of healthy bees, decreased the infestation level of the pathogens *N. ceranae* and *V. destructor*, although it did not modify the strength of the beehive or the infection by the virus of acute paralysis or the virus of deformed wings (Añón, 2018).

In a trial at laboratory level species of *Bacillus* ssp. and *Brevibacillus* ssp. were used, associated with honey bees, as natural alternative for the control of LA and chalk offspring. In such experiment favorable results are reported, showing inhibition against the pathogens. This constitutes the first study of associations between the presence of genes related to the synthesis of antimicrobial peptides and their antagonism against *P. larvae* and *A. Apis*.

Arredondo *et al*. (2018) evaluated in larvae and adult bees the beneficial action of a mixture of four *Lactobacillus kunkeei* strains, isolated from the intestinal microbial community of the insect. Their administration in controlled laboratory models decreased the mortality associated to the infection by *P. larvae* in the larvae and counts of *N. ceranae* spores of adult honey bees. These results suggest that the mixture of this beneficial microorganism can be an attractive strategy to improve bee health. Nevertheless, the above-referred authors suggest conducting field studies to evaluate its effect on naturally infected colonies. Against this same etiological agent beneficial effects of other probiotic strains are reported (Ptaszyńska *et al.,* 2016).

At present the study of the function of the bee microbiota is continued, due to its contribution to the health and productivity of this species, and because of the influence that probiotics can have on its stabilization and on the stimulation of the immune system of bees (Al-Ghamdi *et al*., 2020).

Use of prebiotics in apiculture

Initially, prebiotics are defined as a non-digestible food ingredient, which produces a beneficial effect on the host, by stimulating selective growth or metabolic activity of a limited number of bacteria in the colon. At present, they are conceptualized as a substrate that is selectively used by the microorganisms of the host, and which confer it beneficial health. Thus, the concept of this term is broadened, in which different substances from carbohydrates (polyphenols and polyunsaturated fatty acids) and their application on body sites that are not the gastrointestinal tract, are included. However, the requisite that they are selective mechanisms mediated by the microbiota, as well as the condition of the beneficial effects that are documented on the host's health, is maintained (Gibson *et al*., 2017).

Another important aspect of this definition is that prebiotics are no longer limited to human feeding, but they can be also considered in other categories, such as animal nutrition. The main prebiotic components are fructooligosaccharides (FOS), manane oligosaccharides (MOS), inulin, isomalto-oligosaccharide (IMO), polydextrose, lactulose and resistant starch.

Oligosaccharides, such as those of soybean (SOS), galactooligosaccharides (GOS) and xylooligosaccharides (XOS), are also prebiotic agents (Cheng *et al*., 2014) which participate in the stimulation of the intestinal microbiota, especially in the colon, and produce a fermentation status in the population of *Lactobacillus* and *Bifidobacterium*. These bacteria promote the production of short chain fatty acids, among whose most outstanding effects the decrease of the intestine pH and the control of bacterial communities that can be deleterious for ecological niches, stand out.

Another effect of these populations is the decrease of the time of intestinal passage, which causes the increase of the volume of the fecal bolo and the frequency of depositions (Corzo *et al*., 2015). The most widely studied prebiotics as additives in the diets for non-ruminant animals, and which function differently, are short-chain oligosaccharides of simple sugars, especially FOS, GOS and MOS.

In the consulted databases there was no access to a substantial reference of papers that approach this topic, if it is compared with the available information about probiotics and phytobiotics (Di Gioia and Biavati, 2018). Nevertheless, the utilization of prebiotic and probiotic products can represent an alternative solution for the prophylaxis and treatment of diseases in bees (Pătruică *et al*., 2013). The utilization of organic acids (lactic and acetic) incorporated to the syrup is also reported. It could also be noted in a work by Pătruică *et al.* (2011) that the experimental groups that were fed with acidifying substances recorded statistical differences in terms of the number of offspring at 14 and 21 days, with regards to the control group.

The utilization of lactic acid and cider vinegar, as prebiotics in symbiosis with probiotics, to test their influence on the development of the insect's glands showed a positive result when compared with the control group, according to Pătruică *et al*. (2012). In a similar experimental design, but with lactic and acetic acid as prebiotics, in symbiosis with probiotics, these authors described that in the histological studies of the intestine of worker bees after three weeks of evaluation, there was a close correlation with the absorption of nutrients and with the good development of bee colonies during the active season (Pătruică *et al*., 2013).

The economic and health analyses of the apiaries fed with the above-described prebiotics showed positive balances, expressed in a higher honey production, from 14,7 to more than 45,5 % compared with the control group (Pătruică and Hutu, 2013), as well as a more favorable health status and bioproductive index in the colonies of bees that consumed these products.

The supplementation of the diet of honey bees with probiotics and prebiotics (inulin) did not show beneficial effect against the infection by the pathogen *Nosema ceranae*. In this regard, Ptaszyńska *et al*. (2016) argue that the additive can affect the immune system of the insect, and increase significantly the mortality of bees. These authors evaluated the biological effects of porphyrins (10 µM and 100 µM) against the microsporidium *Nosema ceranae* in bees, for which experiments were developed in cages. As a result they reported the significant reduction of the number of spores (from 2,6 to 5 times) of the experimental group (bees infected with *Nosema* with a diet of syrup with sucrose-porphyrin) with regards to the control bees. In another essay, Juhász *et al*. (2019) recorded increase in the number of *Lactobacillus* spp. with the inclusion of inulin in syrup of bees, and concluded that it was necessary to study further the topic.

Ibarra-Navarrete (2019) evaluated the effect of four levels of oxalic acid (75, 100, 125, 150 g) and the chemical treatment based on amitraz on the control of the mite *Varroa destructor* in *Apis mellifera* bees. This author concluded that the concentration of 150 g of oxalic acid was the one that showed higher effectiveness in the pathogen control, compared with the other treatments. From the economic point of view, it was more cost-effective than the chemical treatment.

Jack *et al.* (2020) evaluated the efficacy of the vaporization of oxalic acid (OA) and the interruption of rearing as controls of *Varroa* with the utilization of different schemes. These authors proved that the colonies treated with amitraz were healthier and had better survival than those treated with the vaporization of OA, which suggests the need to work on the improvement of non-conventional control methods and include cost-effective treatments, which can be easily used by beekeepers.

Plant extracts, phytogenic or nutraceutical

Phytogenic additives are defined as substances from different plant parts, which are incorporated to the diet to improve the animal productivity, by promoting its productive development and improving the quality of animal derived foodstuffs. Phytogenics are classified into herbs (flowers and no-ligneous plants), botanical products (whole plants or parts that exemplify the use of roots, leaves and bark), essential oil (hydrosoluble extraction of volatile plant compounds) and oily resins (extracts based on a non-aqueous solvent) (Diaz-Sanchez *et al.*, 2015).

These additives from plants can vary according to the origin, plant composition, influence of pre-harvest conditions, climate conditions, geographical position, exogenous stress, harvest and post-harvest procedures, such as the processing and extraction of the biological principle, and are used in solid, dry and ground forms, and in liquids as extracts and essential oils (Madhupriya *et al*., 2018).

Plants have an unlimited capacity to synthesize compounds, among which are alkaloids, phenols, flavones, essential oils and related compounds, which makes them an important natural source of substances that have biological properties. In general, from these groups more than 200 000 metabolites are described, and others that continue to be discovered and explored by several scientific specialties (Ncube and Van Staden, 2015).

Consumers prefer phytogenic additives and are aligned with the clean, green and ethical concept that is applied in livestock in general. Clean refers to the decrease of the use of synthetic compounds, green, to the reduction of the impacts generated on the environment, and ethical, to the effects on animal welfare (Stevanović *et al.,* 2018).

One of the mentioned mechanisms to show the antimicrobial activity of plant extracts and essential oils in general, is the hydrophobic characteristic, which favors the interaction of the active principle with the cell membrane, generating disturbance of it, its rupture and damage on its internal structures and permeability, which causes on the pathogen agent extravasation of intracellular material, physiological alteration and changes of virulence, by inhibiting genetic regulation (Rivera-Calo *et al.,* 2015).

Use of plant extracts in bees

Hydrosoluble extracts of ten plant species (*Achyrocline satureioides, Chenopodium ambrosioide, Eucalyptus cinerea, Gnaphalium gaudichaudianum, Lippia turbinata, Marrubium vulgare, Minthostachys verticillata, Origanum vulgare, Tagetes minuta* and *Thymus vulgaris*) were tested as growth inhibitors on *Paenibacillus* ssp, the causative agent of American foulbrood. These extracts showed antibacterial activities and inhibited the growth of almost all the analyzed *P. larvae* strains.

Several extracts of natural organic and aqueous products increase the survival of bees and reduce the burden of spores after the oral treatment. Natural compounds are reported, particularly flavonoids in several plant extracts that show anti-microsporidiosis activity in honey bees, although it has not been confirmed that flavonoids are the source of such activity. Arismendi *et al*. (2018) reported anti-*Nosema* activity *in vivo*, similar to fumagillin in hydrosoluble extracts of essential oil (EO) of *Cryptocarya alba* leaves. These authors observed that the monoterpenes selected from the extract (β-phellandrene, eucalyptol and α-terpineol) also inhibited *N. ceranae*.

The use of nutraceuticals or plant extracts (thymol and carvacrol), included in the sugar syrup to control *N. ceranae* and *N. apis* in honey bees, did not reduce the level of *Nosema* spp. spores although it did decrease the mortality of bees. Nevertheless, in other essays beneficial effects are reported (van den Heever *et al*., 2016).

The supplementation of bees with curcumin, an antimicrobial compound from curcuma (*Curcuma longa*), reduced the burdens of *Nosema* spp. spores and produced higher survival of the infected bees (Strachecka *et al*., 2015). Although they were not proven in this study, promising results were also recorded with polysaccharides from algae and oxalic acid (Nanetti *et al*., 2015) and porphyrins (Ptaszyńska *et al*., 2018). In all the essays the burden of *N. ceranae* spores was reduced, when the bees were fed sugar syrup.

In a study to test the effect on nosemosis of a solution of oxalic acid 0,25 M, administered to bees as organic additive in the sugar syrup, Nanetti *et al.* (2015) observed in laboratory trials that the number of spores was significantly lower in the treated group than in the control one. In the field essays they detected that the prevalence of the infection decreased in young and adult bees. These authors concluded that the application of the oxalic acid syrup can be included among the alternative strategies for the management of this species.

The commercial phytopharmacological additive Nozevit R against *Nosema ceranae* proved to have beneficial effects by decreasing the burden of spores of the colonies. Yet, this same product in cage essays did not have any effect (van den Heever *et al*., 2016).

Another supplement based on sea algae HiveAlive TM caused decrease in the spore burden of colonies and increased the population of beehives with regards to the controls, after the administration of two half-yearly treatments (Charistos *et al.,* 2015).

Essays with methanol extracts from leaves of native plants (*Ugni molinae*, *Aristotelia chilensis* and *Gevuina avellana*) and from propolis, included in the diet at different concentrations for the control of the burden of *N. ceranae* and bee survival, showed their antiparasite effect when the bees were treated with extracts as they were infected with the pathogen. *A. chilensis* (8 %), *U. molinae* (2 and 8 %) and propolis (8 %) significantly decreased the burden of *N. ceranae* and improved the survival of bees. On the other hand, when the bees were treated with extracts first, and were then infected with *N. ceranae*, they showed higher intake of the diet. In this case, all the extracts significantly decreased the burden of the parasite, but only those from *U. molinae* (2 and 8 %) and LR propolis (8 %) maintained high survival of bees infected with *N. ceranae* (Arismendi *et al*., 2018).

Utilization of zootechnical additives in Cuba

In Cuba there is large experience in the utilization of zootechnical additives in several animal species with beneficial effects on health and bioproductive indicators. The most widely used additives are probiotics, among them *Lactobacillus, Bacillus* and yeasts (Hernández-García *et al*., 2019). To a lower extent, they are also used as prebiotics and phytobiotics.

In spite of the experiences with the use of additives and the suggestions that are offered in the review of patents, it is in recent years that it begins to be approached in the topic of bees, for which the promotion of research lines that guarantee the use of probiotics to eliminate the residual effect of antibiotics in honey and other apicultural products that constitute one of the most important exportable items of Cuba, are declared as technological niches (Amaral, 2008).

From the zootechnical additives described for *Apis mellifera*, in Cuba work is done *in vitro* with probiotic strains, isolated from the intestine of adult *Apis mellifera* bees (Hernández-García *et al*., 2020). These studies conducted by the Reference Laboratory for Apicultural Research and Health (LARISA for its initials in Spanish) and the University of Sancti Spiritus show non-concluded promising results. Likewise, the University of Granma develops laboratory and field essays with plant extracts to control the mite *Varroa destructor*.

Both projects are associated to a national program led by the National Center of Agricultural Health (CENSA, for its initials in Spanish). Researchers from this institution have obtained essential oils with potentialities for their use against pathogens that affect bees. The University of Matanzas develops a territorial project of the Ministry of Science, Technology and Environment (CITMA) for obtaining a probiotic biopreparation from the microbiota of the digestive tract of *Melipona beecheii*, in order to inhibit the development of pathogens and increase the productivity of bees. These incipient studies are supported on the need to have natural alternatives that maintain the integral control program of bee health, characterized by the exclusion of chemicals in Cuba.

Conclusions

Obtaining and evaluating zootechnical additives in bees is a strategy worldwide that allows to minimize or eliminate the use of chemicals, protect the environment and the ecosystemic resources, ensure food innocuousness and guarantee the benefits for the insect health as well as productivity. The results that have been reached in this field are supported by much research. However, it is necessary to study the topic further as there is variability in some works, which can be associated to the diversity of factors that participate in the effectiveness of additives, among which those of the additive in question, the ones related to the insect, and those regarding the characteristics of the diets that are supplemented, stand out.

The main additives aimed at honey bees are, first, probiotics (lactobacilli), secondly, plant extracts and, to a lower extent, prebiotics (inulin).

In Cuba, the utilization of zootechnical additives in bees constitutes a sustainable alternative to improve health and productive indicators, and thus maintain the quality of the honeys that are produced free from chemicals. The country has important contributions to the knowledge of zootechnical additives in various animal species in different institutions, but the study in bees is still incipient.

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Conflict of interest

The authors declare that there are no conflicts of interests among them.

Authors' contribution

- Juan Emilio Hernández-García. Participated in the genesis of the idea, bibliographic search, information interpretation and analysis, as well as in the preparation of the manuscript.
- José Antonio Rodríguez-Díaz. Participated in the genesis of the idea, bibliographic search, information interpretation and analysis, as well as in the preparation of the manuscript.
- Osmaida Estrada-Cutiño. Participated in the design of the study, information interpretation and analysis, as well as in the preparation of the manuscript.
- Yovanni Solenzal-Valdivia. Participated in the bibliographic search, information interpretation and analysis, as well as in the preparation of the manuscript.
- Ken Jact Fernández-León. Participated in the bibliographic search, information interpretation and analysis, as well as in the preparation of the manuscript.
- Ana Julia Rondón-Castillo. Participated in the information interpretation and analysis, as well as in the preparation of the manuscript.

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