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ANTIBIOTICS RESIDUES AS LIMITING FACTOR OF HONEY QUALITY

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Abstract

Residues of veterinary drugs represent a significant risk to the health of honey consumers. Antibiotics can get into honey by using the antibiotics for treatment and prevention of bees diseases but also through the plant nectar and pollen. In Serbia, the use of antibiotics in beekeeping for bacterial diseases treatment is prohibited and accordingly there is no prescribed maximum permissible concentration for them in honey. The aim of this paper is to monitor the presence of antibiotic residues in honey which necessarily indicate their illegal and uncontrolled use. The presence of antibiotic residues in honey was screened for microbiological method "Modified method 4 plates" (EUR 15127-EN). The total of 135 samples of different honey types has been examined. Five of them (3.7%) were positive to antibiotic residues. The presence of antibiotic residues was found in the acacia honey (0.31%), linden honey (0.33%), sunflower honey (0.19%), mixed honey (0.17%) and honeydew honey (0.10%). Such unprofessional, unconscionable and unlawful use of antibiotics leads to their presence in honey and other bee products, as well as in the highly desirable and valuable products making them unusable.

Key words: *antibiotics, honey, quality, residue*

Introduction

Within the healthy food that nature gives to man, on the list of quality and value, bee products undoubtedly take special place. Honey and other bee products are a real treasure of natural medicinal-prophylactic ingredients (Babic, 2012). Honey is generally considered as a natural and healthy product (Reybroeck, 2003). It has long been known that honey has an antimicrobial activity. Recently the presence of antimicrobial peptide-defensin in honey has been proven which could someday be used for the treatment of antibiotic-resistant infections (Kwakmann et al., 2010). Risks to consumers of honey are highly dependent on the degree of control exercised by the producers, buyers, processors, retailers and authorized bodies for control of honey which prevent or minimize the risk to an acceptable level. The last few decades, which are characterized by a significant increase in industrial production, increased and uncontrolled use of pesticides and antibiotics in agriculture and intense international traffic have increased environmental contamination caused by various pollutants and toxic substances such as heavy metals, pesticides, radionuclides, and antibiotics (Barišićet et al., 1999; Mujić et al., 2011; Roman et al., 2011). Antibiotics can get into honey by using the antibiotics for treatment and prevention of bee diseases, but also through the plant nectar and pollen (Roman, 2005; Roman et al., 2007). Because of that, content of these substances in honey and bee organism is a very good bioindicator of

the use of antibiotics in beekeeping and of their presence in the environment (Celli and Maccagnani, 2003; Porini et al., 2003). Antibiotics, such as streptomycin, tetracyclines and sulfonamides are often used in beekeeping as preventive or therapeutic treatment to protect apiary (Iancu et al., 2012). Other antibiotics such as erythromycin, lincomycin, monensin, enrofloxacin and alike are also used in beekeeping (Johnson et al., 2010). In the EU the use of antibiotics for bacterial infections has been banned, except for the European foulbrood via the "cascade" system, but taking honey from those hives is banned. So there is no MRL (Maximum Residue Limits) for the presence of antibiotics in honey (EEC Regulation 2377/90 and amendments). Codex Alimentarius Standard for Honey (12-1981) in section 4.2. with the defined MRL for pesticides and veterinary drugs does not define the MRL for antibiotics (Johanson et al., 2010). In Serbia, the use of antibiotics in beekeeping for combating bacterial diseases is prohibited and accordingly there is no prescribed maximum permissible concentration of them in honey (Regulation of concentration of pesticides, metals and metalloids and other toxic substances, chemotherapeutics, anabolics and other substances that can be found in food (FRY Official Register, 5/92, 11/92, 32/2002). Although our beekeeping public knows about this the beekeepers still reach for antibiotics in the process of "saving" the American and European foulbrood what results in uncontrolled presence of antibiotic residues in honey and spread of disease. Some countries outside Europe have legalized the use of tetracyclines, sulfonamides and other antibiotics for the treatment of American foulbrood. Systematic use of tetracycline in Canada and the United States has led to resistance strains of Paenibacillus larvae subsp. larvae to tetracycline. Sulfonamides in some countries are used in the prevention of Nosemosis. Chloramphenicol is used in beekeeping in China, and the positive tested samples for chloramphenicol indicate that the honey is of Chinese origin or honey mixed with the Chinese honey (Reybroeck, 2003). Low concentrations of streptomycin can also be found in fruit honey obtained from the nectar collected from the pear orchards treated during flowering with preparations of streptomycin against fire blight (Reybroeck, 2003). Antibiotic residue exhibits a relatively long half–life and can have direct toxic effects on the consumers, such as allergic response and the induction of drug-resistant strains of bacteria (Gunes et al., 2009; Johnson et al., 2010).

In order to organize beekeeping in a way that would reduce the risks of contamination of bee products by harmful substances to a minimum it is necessary to implement a system of self-control in chain "from farm to fork." According to the chain, beekeepers should be included in the introduction of guidelines "good beekeeping practices" that promote technological development and productivity of beekeeping. The best result of this is getting consumer confidence which is the most important on the global market today. Considering the clinical findings and the epizootic situation, the assumption is that the beegarden still uses the antibiotics. The aim of this paper is to monitor the presence of antibiotic residues in honey which necessarily indicate their illegal and uncontrolled use.

Material and methods

In the last 2013,135 samples of different honey types were collected and examined at the Institute of Veterinary Medicine "Novi Sad" in Novi Sad ("NIV NS") Serbia. Honey samples were collected from two sources: honey samples that were brought by the owners at the "NIV NS" and honey samples collected by experts from "NIV NS" on-site production. All the samples were properly packaged in glass or plastic jars and properly labeled. The presence of antibiotic residues in honey was screened for microbiological method "Modified method 4 plates" (EUR 15127-EN). The total of 135 samples of

different honey types has been examined. If the sample contains active antimicrobial residues there will not be growth of the test microorganism in the diffusion zone, ie. it will be a zone of inhibition. The width of the zone of inhibition is measured from the edge of the hole in the agar to the limits of growth of test microorganisms. The results have been statistically analyzed and presented by means of descriptive statistics.

Results and discussion

The total of 135 samples of different honey types has been examined. Five of them (3.7%) were positive to the presence of residues of antibiotics with inhibition of growth of more than 4 mm. The presence of antibiotic residues was found in the acacia honey (0.31%), linden honey (0.33%), sunflower honey (0.19%), mixed honey (0.17%) and honeydew honey (0.10%). Analysis of differences in the frequency of the number of positive samples of different types of honey indicates that there is no statistically significant difference in the frequency of positive samples between different types of honey ($p>0.05$).

The results of our research correspond to the references and indicate that the use of antibiotics is still present in beekeeping. Such unprofessional, unconscionable and unlawful use of antibiotics leads to their presence in honey and other bee products and make highly desirable and valuable products unusable (Plavša et al., 2005).The antibiotics, such as gentamycin, erythromycin, penicillin, tetracycline, streptomycin, ofloxacin and sulphonimides are also reportedly used in beekeeping residues. These antibiotic residues have toxic acute and chronic effects on human health and reduce the efficacy and quality of honey (Zai et al., 2013). "Modified method 4 plates" is a screening method. This method cannot be used for the detection of the types and amounts of antibiotics in honey. Different techniques were used for the detection and quantification of these antibiotics in honey, mostly Biochip array Technology and Thin Layer Chromatography (TLC) were used for the detection, like Elisa method. The latest research has developed valid, simple and rapid method for antibiotics by HPLC method, mass spectrometry and LC/MS (Liquid chromatography–mass spectrometry). These techniques were found sensitive, reproducible and very useful for antibiotics and others drugs detection because by means of these techniques we are able to quantify the presence of a very low amount of drugs in sample. The type detection of antibiotics in honey by using HPLC is fast, valid and specific (Zai et al., 2013). LC / MS method is simple, rapid, reliable and sensitive enough for routine use in laboratory work (Krivoklavek et al., 2005). ELISA technique is simple, sensitive and represents a specific powerful tool for selective detection of a very low amount of substances in physiological, biological and environmental samples (Jeon et al., 2008). Therefore, this method enables the efficient determination of target molecules in a complex sample without extraction of the sample (Jeon et al., 2008).

Conclusion

Considering the facts stated in the introduction of this paper to ban the use of antibiotics in beekeeping, the results indicate that there is still insufficiently controlled use of products in beekeeping and insufficiently developed awareness among beekeepers. The presence of antibiotics residues is a limiting factor in the quality of honey and makes it unfit for human consumption. Food safety is the top priority both in research and in the legislative field. Natural conditions, moderate continental climate and the wealth of flora and fauna are virtually ideal conditions for beekeeping and for getting quality and safe bee products. The potential for obtaining such high valued products is necessary in order to use and store brand quality honey both on the Serbian and foreign market.

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