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SPECIFIC QUALITY INDICATORS OF MONOFLORAL LINDEN HONEY

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ABSTRACT

The research was aimed at developing a comprehensive approach and identifying criteria for assessing the quality of monofloral linden honey based on the determination of its organoleptic, physicochemical indicators, pollen analysis and establishing compliance with the requirements of quality standards. 32 samples of honey labeled as linden honey were analyzed. Evaluation criteria for monofloral linden honey in Ukraine have been established. The degree of monoflorality of linden honey can be from 30.0% of linden pollen and higher. According to organoleptic indicators, honey from linden is characterized by a peculiar delicate aroma of linden flowers, mainly has a color from light yellow to white shades, consistency depending on the season (liquid, viscous, very viscous, dense). According to physical and chemical parameters: moisture - 18.5%, proline - 308.12±34.18 mg/kg, electrical conductivity - 0.35± 0.04 M/cm, fructose to glucose ratio - not lower than 1.2, diastasis not less than 11.0 units. Gote, the content of reducing sugars is 85.6±2.47%; sucrose in the range of 2.8-3.9%. It has been established that the quality indicators of monofloral linden honey meet the requirements of the national standard of high-grade honey and the existing EU requirements; the content of dominant linden pollen should not be lower than 30%. It has been proven that the ratio of fructose to glucose for monofloral linden honey should not be lower than 1.2, the proline content is not less than 300.0 mg/kg, and the electrical conductivity is 0.63Ms/cm. A comprehensive approach to the identification and evaluation of the quality of monofloral linden honey has been developed and the main criteria for the degree of monoflorality, organoleptic and physicochemical indicators have been determined.

Keywords: linden honey, quality indicators, monofloracy, organoleptic indicators, proline, pollen analysis, electrical conductivity, diastase.

INTRODUCTION

The variety of plants of different climatic zones and geographical origins cause differences in the fodder base and, accordingly, the presence of different types of honey. In recent years, monofloral honey has attracted the increased interest of consumers and, especially, the attention of specialists in the field of medicine due to the presence of phytochemical substances that directly affect certain indicators of human and animal health, in particular, promote wound healing, are characterized by antioxidant, antitumor and anti-inflammatory activity. In the works of many authors, the physico-chemical properties, mineral profiles and antioxidant activity and ecological assessment of individual monofloral honeys are given, depending on their botanical and geographical origin (Adamchuk et al, 2019; Aumeeruddy et al, 2019; Bertonecelj et al, 2011; Choi Suk-Ho et al, 2020; El Sohaimy et al, 2015; Razanov et al, 2022). Different analytical methods are used to identify original honey. New profiles may include parameters such as aliphatic organic acids, amino acids, volatiles, flavonoids, carbohydrates, phenolic acids and proteins, not just sugar profiles (Mateo et al, 2021; Truzzi et al, 2014; Ozcan-Sinir et al, 2020; Nyuk Ling Chin et al, 2020). For the successful promotion of domestic honeys on the internal and foreign markets, their compliance with the requirements of domestic and international standards is a mandatory condition. To determine the botanical origin of honey, the classic methodology is palynological analysis, which is based on counting pollen grains. The botanical origin of the samples is established with the help of pollen analysis supplemented with organoleptic assessment. Physico-chemical parameters such as electrical conductivity, pH, water content, carbohydrates and color of monofloral honey are used to build the data set (Arnauta et al, 2013; Mateo et al, 2021). Scientists of different countries pay a lot of attention to the research of monofloral honeys. Thus, to determine the botanical origin of Polish monofloral honey, samples of aqueous extracts of six types of honey were examined using NMR spectroscopy, namely heather (*Calluna vulgaris* L.), buckwheat (*Fagopyrum esculentum* L), linden (*Tilia* L), rapeseed (*Brassica napus* L. var. *napus*), acacia (*Acacia* Mill.) (Aboud et al, 2011; Zieliński et al, 2015; Chirsanova et al, 2021; Gül Banu Çiçek et al, 2021).

Portuguese scientists investigated the profiles of 51 samples of 12 labeled monofloral varieties of Portuguese honey from several regions of mainland Portugal and the Azores (Zieliński et al, 2021). Melissopalynology, antioxidant capacity, and the content of mineral and toxic elements were analyzed in eight types of Hungarian honey. The monofloral origin of each sample was confirmed by pollen analysis. Honey absorption was positively correlated with antioxidant capacity determined by three different methods (TRC, DPPH, ORAC) and mineral content (Bodó et al, 2021). In recent years, employees of the laboratory of methods for assessing the quality and safety of beekeeping products of the NSC "Institute of Beekeeping named after P.I. Prokopovich", a wide range of research was conducted to assess the quality of honey of various botanical origins (Lazarieva et al, 2021; Lazarieva et al, 2021), which enters the official trade network.

Therefore, it is expedient to develop methods of comprehensive assessment of monofloral honeys, which belong to elite varieties and are in great demand on the world market. In this way, a wide range of research was carried out to assess the quality of honey from linden trees produced in different regions of Ukraine, and significant experimental material was developed, which makes it possible to identify and justify the basic indicators of honey quality as criteria for a comprehensive assessment of its monoflorality (Lazarieva et al, 2021).

MATERIALS AND METHODS

The research was carried out within the framework of the state theme "Development of methods for assessing the quality of monofloral honeys No. 0121U108509" on the basis of the NSC "Institute of Beekeeping named after P.I. Prokopovich". The material for the study was 32 samples of linden honey from different regions of Ukraine, which were sent to the laboratory for the purpose of certification. All samples were declared by the manufacturers as monofloral linden honey. Sampling of honey, analysis of organoleptic and physicochemical indicators was carried out by the methods specified in SSU 4497:2005 "Natural honey. Specifications". Organoleptic indicators were determined by consistency, taste, aroma, degree of crystallization, and physico-chemical indicators by diastase number, hydroxymethylfurfural (HMF) content, mass fraction of water, mass fraction of reducing sugars, and mass fraction of sucrose, proline content and electrical conductivity. Equipment was used for this: a spectrophotometer

ULAB-102 (UNICO, China), refractometer RBH-90 ATC (Republic of China), conductometer HI 98303DiST 3 (HANNA Instruments, made in Mauritius), centrifuge MPW-251 (Poland), pH- meter PH-150 MH (Russia), water bath Labexpert (Ukraine), electronic scales AXIS (Poland). The study of the ratio of the amount of fructose to glucose was carried out using test systems D-Glucose/D-Fructose UV method (r-Biopharm, Germany). Determination of the species composition of pollen grains was carried out by the microscopic method. The identification of honeydew pollen grains was carried out using the atlas of honeydew plants and the use of electronic pollen databases (Karpovych et al, 2015). Identification was carried out using a microscope PZO Warszawa (Poland). The obtained data were processed statistically using the program "Microsoft Excel 15.0" with the calculation of the arithmetic mean (M), standard error (m) (Mazur, 1997).

RESULTS AND DISCUSSION

Natural linden honey is a valuable source of biologically active compounds, as it is enriched with carbohydrates and is therefore easily and quickly absorbed by the body, and is transformed into plastic and energy substances (Lysenko et al, 2012; El Sohaimy et al, 2015; Ramsay et al, 2019). At the first stage of determining the botanical origin of honey, it is important to evaluate the organoleptic parameters with the determination of properties specific to monofloral linden honey: aroma, color, taste, consistency. Honey from linden should have a specific taste, possess a peculiar pronounced aroma of linden flowers. By the aroma of honey, you can judge to a certain extent about its grade and quality. Honey obtained from bees that are fed with sugar syrup does not contain organic volatile substances and therefore does not have the aroma characteristic of flower honey. It is less sweet than floral.

When analyzing linden honey according to organoleptic parameters, a light shade (from light yellow to white) was noted, the honey irritated the mucous membrane of the oral cavity, had a specific taste and had a peculiar delicate aroma of linden flowers. Organoleptic tests (Fig. 1) established that out of the 32 samples of "linden honey" provided, all samples corresponded to the typical characteristics of linden honey in terms of consistency, 11 samples did not correspond in color (had a dark color), 21 were classified as linden honey in terms of taste sample (11 samples had a less taste or no specific aroma for linden honey at all).

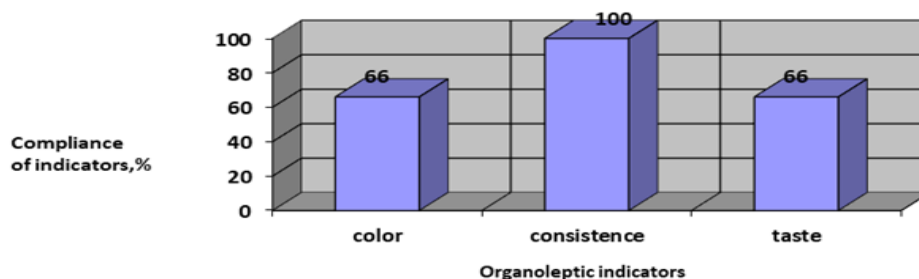


Figure 1. Correspondence of organoleptic quality indicators of linden honey, %.

The obtained results indicate the compliance of the organoleptic characteristics with the typical characteristics of the product under study. From those results presented in fig. 1 is shown that according to organoleptic characteristics, from 66 to 100% of the presented honey samples correspond to the typical characteristics of linden honey. The next task of the research is to determine the physico-chemical parameters characteristic of monofloral linden honey (mass fraction of water, diastase, electrical conductivity, reducing sugars, sucrose content). Moisture content is one of the indicators of its quality. It is estimated by the percentage of water in honey and is regulated in SSU 4497: 2005 "Natural honey. Technical conditions". The moisture content of honey directly depends on its maturity, as well as on storage conditions. The unripe product has high moisture content (above 20%), so it is unsuitable for long-term storage and spoils quickly. Ripe honey contains an average of 18-20% of water. The analysis of the results obtained by us of the studied samples of linden honey (Fig. 2) for the content of the mass fraction of water ranged from 16.7 to 19.6%, which meets the requirements of domestic and international regulatory documents. An excess of water can dramatically reduce the useful qualities and shelf life of honey. The product can "ferment", turning into a foamy mass or harden (crystallize) quickly. Such negative consequences directly depend on the deviation of the normal percentage of water in honey (Mazur, 1997; Polyshchuk et al, 2015).

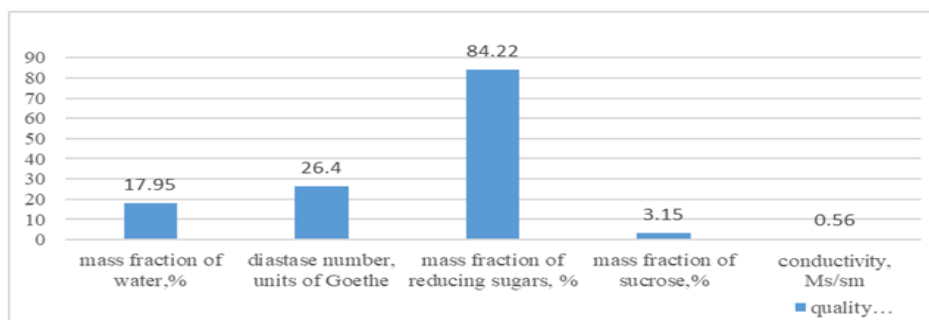


Figure 2. Quality indicators of linden honey (the content of pollen grains is more than 30%).

Diastase is a group of enzymes that includes α - and β -amylases, the amount of which depends on the botanical origin of the nectar and is an indicator of its freshness (Ivashevskaya et al, 2007), in linden honey, the activity of diastase cannot be high, but not less than 11.0 units Goethe. To determine the enzyme activity of honeys, all 32 samples labeled as "linden honey" were examined. It was found out that among the studied samples, the diastase number in linden honey varied widely from 10.1 to 45.8 units Goethe, with an average value of 17.87 ± 4.49 units Goethe (Fig. 2). The ratio of sucrose and reducing sugars in honey characterizes honey in terms of its maturity and quality and can be one of the indicators of the botanical origin of honey. In all types of honey, according to SSU 4497:2005 "Natural honey. Technical conditions", the sucrose content is standardized within the limits of no more than 3.5% for honey of the highest grade and no more than 6.0% for honey of the first grade. While the international requirements for the quality indicator, the sucrose content is no more than 5.0%. The content of reducing sugars in the studied 13 samples ranged from 81.7 to 87.7%, sucrose from 2.8 to 3.9%, with an average value of $3.18 \pm 0.43\%$ (Fig. 2). The result of our studies of the quality indicator of the sucrose content is an average value of 3.15 ± 0.63 , which is within the requirements of both domestic and international regulatory documents. Electrical conductivity is a parameter included in the new international standards regarding the differences between honeydew and flower honey. The limits of this parameter set by the standards are 500 to 800 $\mu\text{S/m}$ for mixed honey, $<500 \mu\text{S/m}$ in the case of pure floral honey with some exceptions. Our research showed that when analyzing linden honey samples, the electrical conductivity ranged from 0.305 Ms/cm to 1.102 Ms/cm. In the case of monofloral linden honey in the presence of linden pollen grains of 30% or more, the average value of the electrical conductivity indicator is $0.63 \pm 0.07 \mu\text{S}$ (Fig. 3).

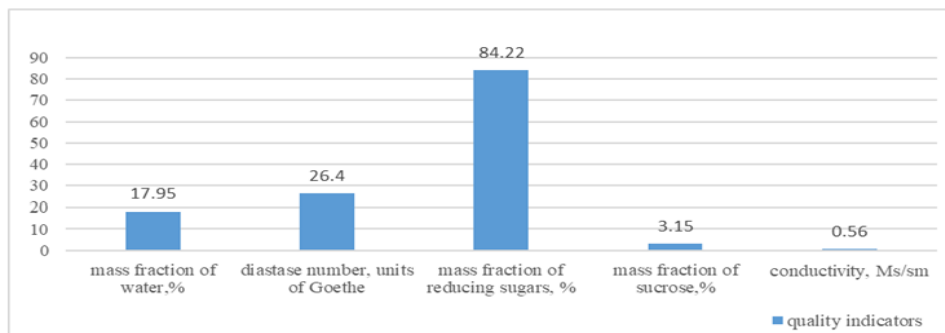


Figure 3. Quality indicators of linden honey (the content of pollen grains is lower than 30%).

The third stage of the study of monofloral honey from linden is the indicators characterizing its naturalness (proline, the ratio of glucose to fructose) and botanical origin according to the indicator of pollen composition. Proline is the main amino acid that enters honey during enzymatic processing and serves as an indicator of honey's naturalness and maturity. It should be noted that according to the requirements of SSU 4497:2005 "Natural honey. Technical conditions" the proline content in honey should be at least 300 mg/kg, according to international regulatory requirements it should be at least 180 mg/kg.

The obtained research results show that the proline content ranged from 239.0 mg/kg to 471.0 mg/kg. When studying samples of linden honey with a pollen grain content of more than 30%, the average value of proline content was 308.12 ± 34.18 mg/kg, and with the presence of pollen grains less than 30%, the average value of proline content was 408.16 ± 31.13 mg/kg (Fig. 4). When studying linden honey samples, where the pollen grain content was 30%, the proline content was 319 mg/kg, when the honey contained 89% linden pollen grains, the proline content decreased to 239 mg/kg, and when 15% pollen grains were present the proline content increased to 471 mg/kg. The obtained data show that the values of proline content in linden honey meet both the requirements of SSU 4497:2005 and the existing EU requirements.

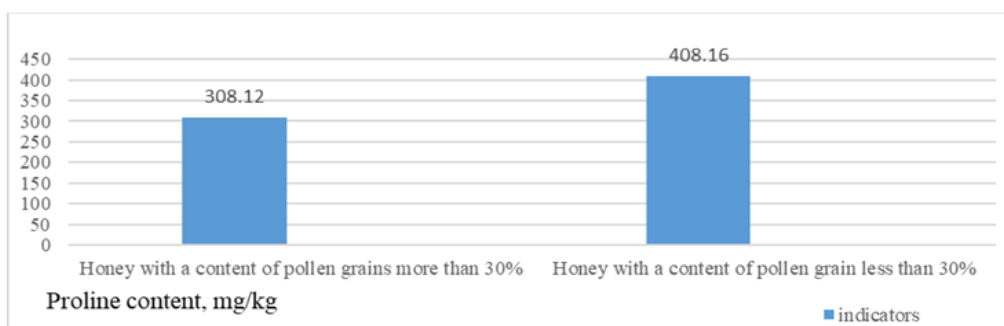


Figure 4. Quality indicators of linden honey characterizing its naturalness (proline, glucose to fructose ratio).

Research results showed that the ratio of the amount of fructose to glucose ranged from 0.88 to 1.28. It was found that the fructose to glucose ratio of honey samples with less than 30% linden pollen ranged from 0.88 to 1.1 with an average value of 0.99 ± 0.10 . In samples of honey where the content of linden pollen grains was more than 30%, the ratio of fructose to glucose ranged from 1.1 to 1.28, with an average value of 1.19 ± 0.047 . After analyzing the obtained data, it was established that the content of linden pollen grains in these samples varied in the range from 3.0 to 89.0%. In 10 samples of linden honey secondary (6.2-29%) there is pollen from the legume family clover (*Trifolium* spp.), mouse nut (*Vicia cracca*) pollen grains of cruciferous (*Cruciferae*), stinging nettle pollen grains (*Urtica dioica* L.), sowing buckwheat (*Fagopyrum esculentum*), sunflower pollen (*Helianthus annuus* L.), the presence of bee fall, willow pollen grains was found (*Salix* spp.) amorphous bush (*Amorpha fruticosa* L.), white acacia (*Robinia pseudoacacia* L.) pollen grains of creeping clover (*Trifolium repens* L.), grunting white *Melilotus albus*, clear white (*Lamium album* L.), pollen grains of meadow clover (*Trifolium pratense*), goldenrod (*Solidago canadensis*), oregano (*Origanum vulgare* L.), mouse nut (*Vicia cracca*), borage plant (*Heracleum sosnowskyi* Manden.) *Veronica* (*Veronica officinalis*), pine (*Pinus sylvestris*), medicinal blood clot (*Sanguisorba officinalis*), cotton wool (*Asclepias*), cornflower (*Centaurea cyanus*), camel thorn (*Alhagi pseudoalhagi*). We found that among the analyzed samples of linden honey (Fig. 5), the diastase number decreases in accordance with the increase of pollen grains from linden amount in the honey.

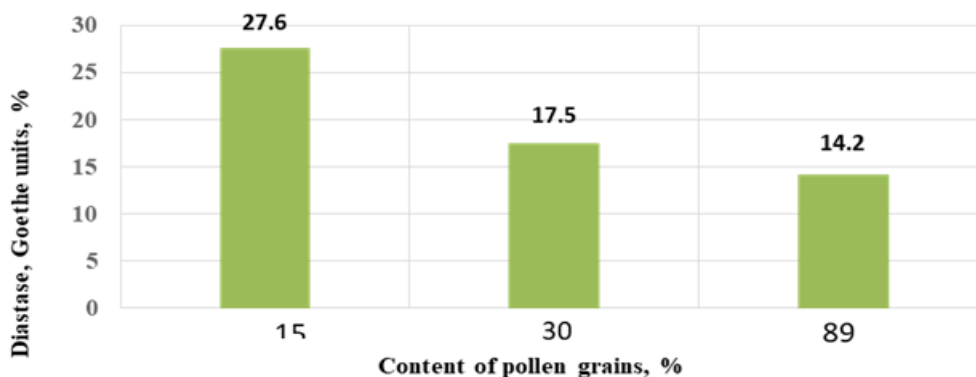


Figure 5. The influence of the content of pollen grains from linden on the activity of diastase, units Goethe.

We proved that with a content of pollen grains from 15 to 30%, the diastase number of linden honey was in the range of 27.6 units Goethe up to 17.5 units Goethe, and with an increase of pollen grains from 30% to 89%, diastase ranged from 17.5 to 14.2 units Goethe.

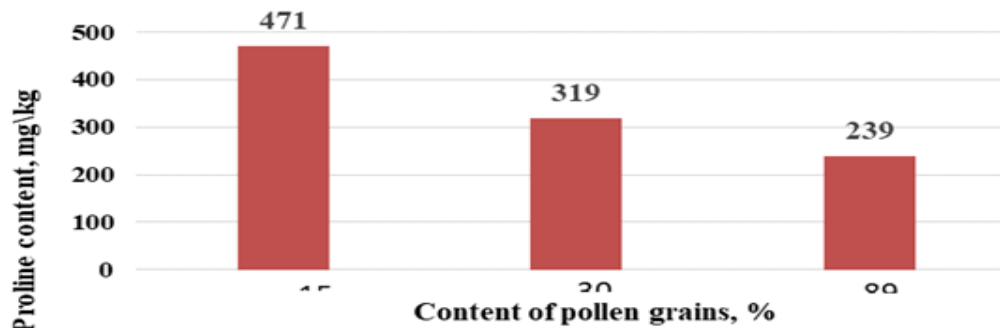


Figure 6. Influence of the content of pollen grains from linden on the content of proline, mg/kg.

At the same time, it should be noted that in the study of linden honey samples (Fig. 6), where the content of pollen grains was 30%, the proline content was 319 mg/kg, with the presence of 89% linden pollen grains in the honey, the proline content decreased to 239 mg/kg, and in the presence of 15% of pollen grains, the proline content increased to 471 mg/kg and it acquires the properties of polyfloral honey (herbs).

CONCLUSIONS

- A comprehensive, step-by-step approach to assessing the quality of monofloral linden honey has been developed, which characterizes its quality and specificity by organoleptic properties, specific physicochemical indicators and indicators of honey naturalness.
- Studies of 32 samples of linden honey from different regions of Ukraine established that its quality indicators meet the requirements of the national standard SSU 4497:2005 "Natural honey. Technical conditions" of high-quality honey and existing EU requirements, except for the indicator of the mass fraction of water.
- According to the pollen analysis indicator, it was established that regardless of climatic conditions, the content of dominant pollen from linden should not be lower than 30%. With a lower content of dominant pollen, the typical characteristics of linden honey change and it acquires the properties of polyfloral honey (various herbs).
- According to research results, it has been proven that the ratio of fructose to glucose for monofloral linden honey should not be lower than 1.2, the proline content should not be less than 300.0 mg/kg, and the electrical conductivity should not be less than 0.63 Ms/cm.

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