ГОДИШНИК НА СОФИЙСКИЯ УНИВЕРСИТЕТ „СВ. КЛИМЕНТ ОХРИДСКИ“

ФАКУЛТЕТ ПО МАТЕМАТИКА И ИНФОРМАТИКА
Том 102

ANNUAL OF SOFIA UNIVERSITY „ST. KLIMENT OHRIDSKI“

FACULTY OF MATHEMATICS AND INFORMATICS
Volume 102

CLASSIFICATION OF DIFFERENT TYPES BEE HONEY ACCORDING TO PHYSICO-CHEMICAL CHARACTERISTICS

KRASTENA T. NIKOLOVA, DOYCHIN T. BOYADZHIEV

The physicochemical parameters (refractive index, water content, β-carotene, color parameters and content of glucose, fructose, sucrose and oligosaccharides for 14 types of bee honey have been investigated. They are grouping according to the following parameters:

1. Geographic region 1 – valley-mountain
2. Geographic region2 – North or South Bulgaria
3. Year of producing – 2008 or 2009
4. Botanical origin – honeydew, multiflorous, sunflower, lime.

Analysis of the data gives the opportunity for characterizing the samples of bee honey by using discriminant analysis. The models correctly present geographic region, year of producing, botanical origin and it can be used for determining the type of unidentified samples.

Keywords: Bee honey, physicochemical properties, discriminant analysis, mathematical modeling

2000 Math. Subject Classification: 62P30

1. INTRODUCTION

Bee honey contains a variety of different sugars, more than 180 ingredients such as enzymes, organic acids, vitamins, minerals, polyphenols, carotenoids, antioxidants, flavonoids, etc [1, 2, 3]. As is well known, one of the parameters to estimate the quality of honey is the contents of sugars. The most common glucose, fructose and saccharose are contained in honey in proportions as follows: 31.3%, 38% and 8% [4]. The variety of components of bee honey are an important criterion for the quality and mark some particular features of the corresponding sample. A number of authors have sought to identify the most significant parameters in order to classify bee honey. Models to classify citrus and eucalyptus honey by studying the water content, electric conductivity, pH factor, contents of glucose, saccharose and fructose have been proposed [5]. Color coordinates x, y and lumineance L are of essential significance for the classification of 15 types of Spanish honey – forest, lavender, eucalyptus, rosemary, citrus etc. [6]. Data about step-by-step discriminatory analysis, principal factor analysis for Spanish, Italian, Iranian and African honeys have been reported [7, 8]. There is comparatively little data on Bulgarian honeys such as multi-flower, acacia, lime, sunflower, forest honeydew.

The objective of this work is to test discriminatory models using the analyzed indicators to discern the geographic origin (field, mountain or Northern–Southern Bulgaria), year of production (2008–2009) and botanic origin (multiflorous or sunflower).

The objective defined requires the solution of the following problems:

- Creation of database, including types of bee honey of different botanic origin and region of cultivation;
- Determination of physical-chemical parameters (color coordinates a^* and b^*, x and y in two colorimetric systems SIE Lab and XYZ, correspondingly, luminance L^*, content of pigments such as β-carotene and chlorophyll, water content, index of refraction, sugar content).
- Establishing of significant differences in the parameters under study.
- Modeling and analysis of the groups by types of honey, yield, and regional origin.
- Test of the obtained model by using independent samples.

2. MATERIALS AND METHODS

2.1. SAMPLES

The basic data includes 14 types of bee honey of field and mountain regions in Northern and Southern Bulgaria. The samples were purchased from producers and suppliers, from two years – 2008 and 2009. Four samples of multi-floral honey with commercially available sweeteners were used to test the models.

2.2. METHODS

The color parameters of two different colorimetric systems – XYZ (aimed at large color differences) and CIE Lab (aimed at small color differences) [9] are measured. A colorimeter Lovibond PFX 880 (UK) and a cuvette with a 10 mm thickness are used. To determine the water content the refractive index is measured using an Abbe refractometer (Carl Zeiss, Germany) at $20 \pm 0.5\,^{\circ}$C The equivalent water content is determined from a table, given in Official Methods of Analysis [10]. After the honey solution is filters, the sugar content is determined using liquid chromatography with an IR detector (Waters). The parameters of the methods are: column Aminex HPX-87H; detector Differential refractometer R401, (Waters); temperature of the column and the detector is 30°C, the volume of the sample injected was $10\,l$, speed – $0.5\,ml/min$. The software "Statistica" to process the data was used. Their distribution is normal according to the Kolmogorov–Smirnov criterion [11, 12]. To establish the statistically significant differences between the indicators for the different sorts Tukey criterion for multiple comparisons was applied [13].

Discriminatory analysis is used to model the group with a priori equal probabilities to fall into the groups [14].

3. RESULTS AND DISCUSSION

The database includes 14 types of bee honey from different regions (valley-mountain or southern-northern Bulgaria). For each of the samples studied, four independent measurements have been performed. The Scheffe criterion shows significant statistical differences in the studied types of honey. The presence of considerable difference in the physicochemical characteristics of honey provides the reason for a subsequent modeling of its origin. To model the honeys by region valley-mountain a step-by-step linear discriminatory analysis was used.

A model with grouping parameter "extraction area" was obtained and it includes the following parameters by the order of introduction into the model: x, oligosaccharides and refractive index. The classification of the different sorts according to the extraction area is 100% (Table 1).

TABLE 1. Classification of the samples by the model Valley–Mountain

Group	Percent correct	Mountain $p = 0.357$	Valley $p = 0.643$
Mountain	100	20	0
Valley	100	0	36
Total	100	20	36

It has been attempted to discern the samples by the geographical region with a grouping variable Geographic area 2: Northern-Southern Bulgaria. With a classifying parameter "Geographical area 2" (Northern or Southern Bulgaria) we observe a

76.8% correct classification, with all samples from two types from southern Bulgaria and one from Northern Bulgaria were incorrectly classified.

Except by region, the samples are subdivided by the year of extraction. More parameters are included in the new model – the color coordinates, luminance and beta carotene. The presence of more variables in the model is easy to explain because the content of sugars is decisive for the crystallization of honey, while the refractive index (water content) – for the development of microorganisms in the product. The indicated parameters are related to the kinetics of the process in the bee honey during storage. The classifying parameter (Year of production) (2008 and 2009) 94.64% of the samples are recognizable, of them only three fall into a wrong group (Table 2).

TABLE 2. Classification of the samples by the year of production

Group	Percent correct	2008 $p = 0.286$	2009 $p = 0.714$
2008	87.50	14	2
2009	97.50	1	39
Total	94.64	15	41

With a classifying parameter "Botanical origin" two models are possible: with color parameters included and physicochemical indicators arranged as luminance, parameter a, saccharose, or only with color parameters: y, L, a, b and refractive index.

TABLE 3. Modeling of botanical origin by color and physicochemical indicators

Group	Percent correct	Honeydew p=0.273	Multi-floral p=0.455	Sunflower p=0.273
Honeydew	100.00	12	0	0
Multi-floral	80.00	0	16	4
Sunflower	100.00	0	0	12
Total	90.91	12	16	16

For the classifying parameter "Botanical origin" both model have the same sample identification capability of 90.91%, with four samples in the first model move from multi-floral group into the sunflower, while in the second model three multi-floral samples (wrongly identified in the first model as well) go into the sunflower group, while one sunflower sample was identified as multi-floral. The results are presented in Tables 3 and 4.

TABLE 4. Modeling the botanical origin by color indicators

Group	Percent correct	Honeydew p=0.273	Multi-floral p=0.455	Sunflower p=0.273
Honeydew	100.00	12	0	0
Multi-floral	85.00	0	17	3
Sunflower	91.67	0	1	11
Total	90.91	12	18	14

For a better visualization of the results a subsequent canonical analysis was performed. On the basis of the first two canonical variables, the position of the separate samples for the model with included color parameters and physicochemical indicators is presented in Fig. 1, the four wrongly identified samples being marked.

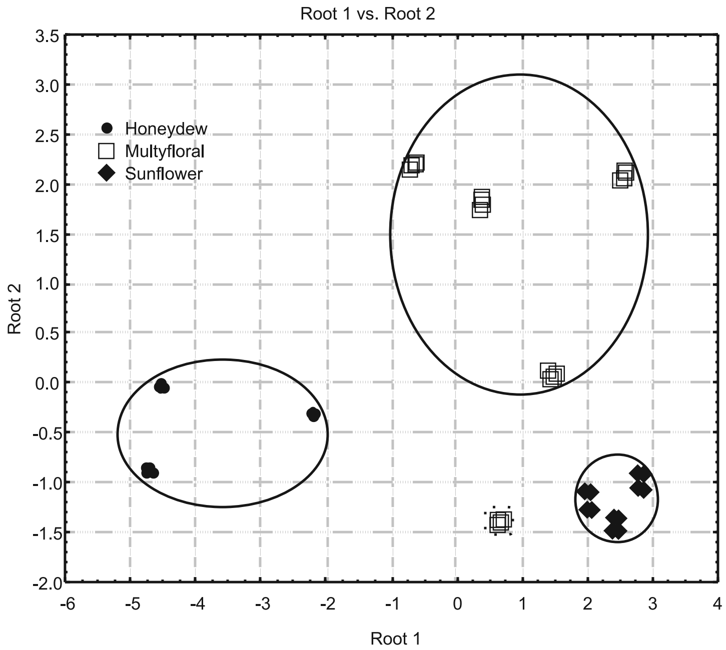

Fig. 1. Disposition of the three sorts of honey in the plane of the first two canonical variables

The figure confirms the stated hypothesis for the presence if significant differences between the separate types of honey. The analysis of the Mahalanobis distances between the three basic groups shows that the sunflower and the multi-floral honeys are close to each other and are relatively far from the honeydew. This is clearly seen from the figure shown if we trace the projections of the clouds of the various sorts upon the first canonical variable which plays an important role in discrimination of the groups – the sunflower and multi-floral are projected on the positive, while honeydew is projected on the negative direction.

The samples used are for the control of the adequacy of the created model for the description of the botanical origin of honey. From the remaining three types of honey with a known botanical origin, the samples which are insufficient to form separate groups two-lime and acacia can be classified as multi-floral, while that of thistle – as sunflower. This can be explained with the different seasons during which they are collected – the former two in the spring while the third in the summer. Honeydew is classified correctly, lime honey and acacia honey are in the group of multi-floral honey. The results from the classification according to the obtained models are presented in Table 5.

TABLE 5. Verification of the model for botanical origin with independent samples

Sort	Classified as
Lime	Polyfloral
Acacia	Polyfloral
Honeydew	Honeydew
Thistle	Sunflower
With sweetener	Polyfloral, Honeydew

4. CONCLUSIONS

The analysis of the data base give the opportunity to characterize different types of bee honey by using the discriminant analysis. It provides an efficient tool for the qualitative distinction of natural bee honey and adulterated honey containing admixtures from sugar or glucose. The models and the associated Mahalanobis distances enable the classification of unknown samples or samples with admixtures.

5. REFERENCES

1. Gheldorf, N., Wang, X. H., Engeseth, N. J.: Identification and quantification of antioxidant components of honey from various floral courses. *Journal of Agricultural and Food Chemistry*, **50**, 2002, 5870–5877.
2. Aljadi, A. M., Kamaruddin, M. Y.: Evaluation of the phenolic content and antioxidant capacities of two Malaysian floral honey. *Food Chemistry*, **85**, no. 4, 2004, 513–518.
3. Al-Mamary, Al-Meeri, Al-Habori: Antioxidant activities and total phenolics of different types of honey. *Nutrition research*, **22**, no. 9, 2002, 1041–1047.
4. Al-Khalifa, Al-Arify: Physicochemical characteristics and pollen spectrum of some Saudi honeys. *Food Chemistry*, **67**, 1999, 21–25.
5. Serrano, S., Villarejo, M., Espejo, R., Jodral, M.: Chemical and physical parameters of Andalusian honey: Classification of Citrus and Eucalyptus honeys by discriminant analysis. *Food Chemistry*, **87**, 2004, 619–625.
6. Mateo, R., Bosch-Reig, F.: Classification of Spanish Unifloral Honeys by Discriminant Analysis of Electrical Conductivity, Color, Water Content, Sugars, and pH. *J. Agric. Food Chem.*, **46**, no. 2, 1998, 393–400.
7. Baroni, M. V., Chiabrando, G. A., Costa, C., Wunderlin, D. A.: Assessment of the Floral Origin of Honey by SDS-Page Immunoblot Techniques. *J. Agric. Food Chem.*, **50**, no. 6, 2002, 1362–1367.
8. Iglesias, T., Martín-Álvarez, P. J., Carmen, P. M., de Lorenzo, C., Pueyo, E.: *Journal of Agricultural and Food Chemistry*, **54**, no. 21, 2006, 8322–8327.

9. Commission Internationale de I Elairage Recommendations on uniform color spaces, color difference equations, psychometric color terms. CIE publication no. 15 (F.1.3.1.), 1971, supplement 2. Burean central. de la Commission Internationale de I Eclairage. Vienna, 1978.

10. AOAC, Official methods of Analysis, Association of Official Analytical Chemists, Arlington, VA, 1995.

11. McLachlan, G. J.: *Discriminant Analysis and Statistical Pattern Recognition*. John Wiley & Sons. Inc, 1992.

12. Vandeginste, B. G. M., Massart, D. L., Buydens, L. M. C., De Jong, S., Lewi, P. J., Smeyers-Verbeke, J. (ed.): *Handbook of Chemometrics and Qualimetrics, Part A, Part B*. Elsevier, 1998.

13. Kalinov, K.: *Statistical methods in behavioral and social sciences*. New Bulgarian University, Sofia, 2001 (in Bulgarian).

14. Lakin, G.: *Biometrics*. Vyshaya shkola, Moscow, 1990 (in Russian).

Received on 3 March, 2015

Krastena T. Nikolova
University of Food Technologies
26 Maritsa Blvd, 4000 Plovdiv
BULGARIA
e-mail: kr.nikolova@abv.bg

Doychin T. Boyadzhiev
Plovdiv University "Paisii Hilendarski"
236 Bulgaria Blvd, 4003 Plovdiv
BULGARIA
e-mail: dtb@uni-plovdiv.bg