The Journal of Animal & Plant Sciences, 28(1): 2018, Page: 252-263
ISSN: 1018-7081

COMPARISON OF QUALITY CHARACTERISTICS IN HONEY USING GREY RELATIONAL ANALYSIS AND PRINCIPAL COMPONENT ANALYSIS METHODS

M. Topal[1*]and A. M. Yağanoğlu[2]

[1]Department of Biostatistics, Faculty of Medicine, Kastamonu University, Kastamonu, Turkey.
[2]Departmant of AnimalScience, College of Agriculture, Atatürk University, Erzurum,Turkey
Correspondingauthor email:mtopal@kastamonu.edu.tr

ABSTRACT

Composition characteristics are taken into account to determine authenticity and quality of honey. This study used grey relational analysis and principal component analysis methods to identify the most important variables affecting quality of honey.Honey specimens from 20 different producers were obtained to determine quality characteristics. C4 % (0.787), glucose (0.753), moisture (0.731), F+G (0.712), fructose (0.685), acidity (0.605), brix (0.581), conductivity (0.580), δ13C honey (0.576), proline (0.571), pH (0.530), δ13Cprotein-honey (0.527), δ13C protein (0.516), Fructose-glucose ratio (0.507) and diastase number (0.490) were found to be the most important variables on quality of honey based on the mean values of grey relational coefficients of quality characteristics. Grey relational grade (GRG) calculated by using eight values obtained from principal component analysis of quality parameters showed that S6 (0.690) was the honey with the highest quality, while S16 (0.501) was the honey with the lowest quality.

Keywords: Honey quality, Carbon isotope ratio ($^{13}C/^{12}C$), PCA, Grey relational analysis.

INTRODUCTION

Honey is a natural sweet substance which is produced by honey bees from the nectar of plants (blossom honey) or from secretions of living parts or excretions of plant-sucking insects on the living parts of plants (honeydew) that bees collect, transform and place in honeycombs to mature (Codex Stan,1981; Turkish Food Codex, 2005).

Composition and quality of honey varies according to production method used by the producer, ecologic characteristics of the region, processing, storage conditions and flora which is the source of nectar (White,1978; Anklam, 1998; Guler *et al*., 2008; Tosun, 2013; Guler *et al*., 2014). Criteria such as moisture, fructose+glucose, fructose/glucose, free acidity, electrical conductivity, diastase number, proline,$\delta^{13}C$ (protein-honey) are taken into account to determine the authenticity and quality of honey (Codex Stan,1981; Turkish Food Codex, 2005).

Various statistical methods have been used to determine the relationship between quality characteristics in honey. Principal component analysis (PCA), linear discriminant analysis (LDA) and cluster analysis (CA) methods have been used to classify honey according to regions (Silvano *et al*., 2014; Gok *et al*., 2015). PCA and LDA have also been used to classify honey according to physicochemical characteristics (Chakir *et al*., 2011). There are also reports on the use of PCA to classify honey specimens (Chua *et al*., 2012) and together with CA methods to classify honey according to mineral compositions (Yücel and Sultanoğlu, 2013). LDA and PCA methods have been used to evaluate analysis results of pure honey and adulterated honey that contain various sugar syrups at different ratios (Subari *et al*., 2012). In addition, PCA method has been exploited determine source of flora and mineral composition of honey using near infrared spectroscopy (Escuredo *et al*., 2015).

Various assumptions such as homogeneity of variance, fit of distribution of data to normal distribution and adequate sampling size have to be valid to use parametrical statistical analysis methods. Non-parametrical methods or parametrical methods in which normal distribution assumption is not valid are used when these assumptions are not valid. Grey relational analysis method is another method that is used when sampling size is not adequate. The assumption that data fits any distribution is not valid in this method (Deng, 1982; Xia *et al*., 2012). Furthermore, both discrete data, that is, the data obtained according to grading scale and continuous data, that is, the data obtained according to range and ratio scales can be used in this method. Grey relational analysis method is especially appropriate in cases with a limited sampling size or in cases which require the selection of studied specimens. When it is necessary to select specimens or individuals by considering a certain or a group of characteristics, this method allows making comparisons between individuals or groups by performing individual or group grading and classification when required. Grey relational grade (GRG) which is calculated by simple mathematical operations and is easy to interpret, can be used to choose good and poor examples in individual comparisons especially in quality analyses. Grey relational analysis has been used in evaluation of water quality (Zhu and Hao, 2009; Liu *et* *al*., 2011; Cheng *et al*., 2011), classification of tobacco leaves (Mao *et al*., 2010) and in the sensory analysis results of the samples obtained from the Longissimus dorsi, Triceps brachi and Semitendinosus muscles of the Awassi and Red Karaman sheep breeds (Topal *et al*., 2016).

The purpose of this study was to determine the variables having the most significant impact on determining quality of honey using grey relational analysis and principal component analysis methods and to determine the highest quality honey by comparing honey specimens supplied from twenty different honey production enterprises in a region with the same environment and flora.

MATERIALS AND METHODS

Pure honey specimens produced in Erzurum were used in the study. The specimens were collected from different honey producers at the end of production season and were subjected to analyses at room temperature in laboratory. The specimens were collected in the form of extracted honey from twenty different producers and were not subjected to any processing or thermal procedure. Analyses were conducted shortly after the collection of specimens.

Variation in each variable was analyzed by calculating coefficient of variation (CV) descriptive statistics values of each variable. Correlation analysis was used to determine level of relationship between the variables.

In this study, grey relational analysis was used to determine the variables that affect quality of honey and to identify honey with the highest quality. Grey relational analysis is a sub-title of grey system theory and is a method of grading, classification and decision making (Baş and Çakmak, 2012; Topal *et al*., 2016). Grey system theory was developed by Ju-Long Deng in 1982 and is used to eliminate uncertainty in data sets especially in those with small sampling size that does not estimate any distribution assumption (Deng, 1982; Xia *et al*., 2012). Grey systems can be divided into three types including the black system, the white system and the grey system. Color black represents that information is unknown; color white represents that information is exactly known, while color grey represents that information is not exactly known, in other words, it is between black and white. Grey relational analysis is an influential procedure for solving inter-relationships among multiple-performances (Sudeepan *et al*., 2014). Grey relation coefficient is used form grading, classification and decision-making in data set. Grey relation coefficient takes a value between 0 and 1. A coefficient value close to 1 means high relationship between real values and reference values, while a value close to 0 means that the relationship is low (Hsu and Huang 2006; Deng 1989). Grey relational analysis is used to determine the relationship between the series in grey relational grade (GRG). If the two series are the same, GRG becomes equal. In this study, weight values w_k were obtained from principal component analysis (PCA) (Mehat *et al*., 2014). Xuerui *et al*. (2007) and Xiaojun *et al*. (2010) defined the lower and upper limits of the grey relational grade (GRG).

RESULTS AND DICUSSION

Coefficient of variation (CV%) values of quality characteristics and raw data obtained from twenty specimens are presented in Table 1. The correlation coefficients between the quality characteristics are given in Table 2.

As the measurement units of quality characteristics were different in Table 1, variation coefficient was used as distribution measurement to compare the variation of quality characteristics. According to variation coefficient, the highest variability in quality characteristics were observed in δ13C protein-honey ratio (CV=175.50), C4% (adulteration) (CV=139.40%) and diastase number (CV=25.82), while the lowest variability was observed in δ^{13}C honey (CV=1.09), δ^{13}C protein (CV=1.11), brix (CV=1.22) and F/G ratio (CV=3.64) respectively. Variation coefficient can also be used to determine the stability of quality characteristics. Lower variation coefficient shows more stability in a variable.

According to correlation coefficients (Table 2), there is a direct and statistically significant correlation between fructose and F+G (0.989), glucose and F+G (0.986) and fructose and glucose (0.951). It can be stated that an increase in any of these variables positively affects the other. It was observed that there was a negative significant relationship between brix and moisture (-0.486) and brix and δ^{13}C protein (-0.506), which means that an increase in brix value causes a decrease in moisture and δ13C protein value. There was a positive significant relationship between conductivity and proline (0.678). However, there was a negative significant relationship between conductivity and diastase number (-0.518). Thus, it can be stated that a change in conductivity of honey has a linear effect on proline and a reverse effect on diastase number. A negative and statistically significant relationship was observed between C4% and F/G ratio (-0.636) and C4% (adulteration) and δ13C protein-honey (-0.868). There was a negative correlation between δ^{13}C protein-honey and δ^{13}C protein (-0.611) and a positive correlation between δ^{13}C protein-honey and F/G ratio (0.634). An increase in δ^{13}C protein-honey value causes a decrease in δ^{13}C protein value and an increase in F/G ratio. In a previous study carried out on honey obtained from bees that feed on different sugar groups, a positive relationship was found between δ^{13}C value of honey and C4% (r =

0.997) (Guler *et al*., 2014). A negative correlation was found between conductivity and water amount (r = -0.6130). It was reported that conductivity value decreases when water amount increases (Chua *et al*., 2012). In another study, a positive correlation was observed between water content of honey and conductivity, color and glucose content and a negative correlation was observed between saccharose, HMF and free acidity. The same study also found a negative correlation between conductivity and free acidity (Silvano *et al*., 2014).

Since the units of quality characteristics in data set are different, measurement units of quality characteristics should be converted to the same unit and normalization should be performed before conducting grey relational analysis. Normalized values for quality parameters and the difference value between comparability sequence and reference sequence are presented in Table 3.

Normalization decreases variation in sequence. Values vary between 0 and 1 after normalization. "The minimum approach" normalization method is used if the values in the series are desired to take small values. In linear normalization, points that take small values take values close to "1", while great values take values close to "0". If the values in the series are desired to take great values, "the maximum" normalization method is used. The points that take great values take values close to "1", while those taking small values take values close to "0". The objective approach method was used in normalization operation of brix and F/G ratio variables. On the other hand, the minimum approach was used in normalization operation of moisture, conductivity, pH, acidity δ^{13}C honey and δ^{13}C protein variables. The maximum approach method was used in normalization of proline, diastase number, fructose, glucose, F+G, δ^{13}C protein-honey and C4% (adulteration) variables.

Grey relational coefficients calculated for each quality characteristic are presented in Table 4. Considering mean grey relation coefficients of quality characteristics in honey presented in Table 4, the most significant quality criteria were found to be C4% (adulteration) (0.787), glucose (0.753), moisture (0.731), F+G (0.712), fructose (0.685), acidity (0.605), brix (0.581), conductivity (0.580), δ^{13}C honey (0.576), proline (0.571), pH (0.530), δ^{13}C protein-honey (0.527), δ^{13}C protein (0.516), Fructose/glucose ratio (0.507) and diastase number (0.490) respectively for the example data set. As a result, these variables (C_4% (adulteration), glucose, moisture, F+G and fructose) can be taken as the most important quality characteristics to determine quality of honey (Table 4). The larger grey relational grade value corresponds to high quality performance (Sing *et al*., 2014). In a previous study found thatthe C4%, vitamin C, F/G ratio, viscosity, invertase, Δδ13Cp-h werefoundto be successful in discrimination of honeysamples in theStepwisemethod (Önder *et al*., 2016).

In determining grey relation grades (GRG) PCA was applied to grey relational coefficients presented in Table 4 to determine weight (w_k) values of each quality characteristic. Null hypothesis correlation matrix is formed as unit matrix to test the applicability of PCA to data set. Bartlett's sphericity test was used to test the applicability of PCA on data set, in other words, to test whether the correlation coefficients matrix was a unit matrix. Bartlett's sphericity test value was found to be 337.265 ($P<0.001$). It can be stated that correlation matrix was not unit matrix, in other words, correlations between some variables in the correlation matrix were significant and thus principal component analysis can be applied to our data set. Principal component analysis results of grey relational coefficients of quality characteristics in twenty honey specimens are presented in Table 5. The total variation in the data set are described by 85.96% in the first six factors.

The components with an eigenvalue greater than 1 were considered as significant. Eigenvalues of principal components were found to be 3.93, 3.037, 2.114, 1.607, 1.179, 1.025, 0.862, 0.438, 0.421, 0.195, 0.084, 0.063, 0.042, 0.001 and 0.000 respectively. 85.96% of total variation in data set was explained by first six factors. 26.21% of total variance was explained by the first factor; 20.24% of total variance was explained by the second factor; 14.10% was explained by the third factor; 10.71% was explained by the fourth factor; 7.86% was explained by the fifth factor and 6.83% was explained by the sixth factor. Other nine components were found to explain 14.04% of total variation. These components were not considered as significant.

Square of eigenvectors show the contribution of quality characteristic that corresponds to principal components. Contribution shares of quality characteristics that were obtained by taking the squares of each eigenvector are presented in Table 5. For example, contribution of brix was $0.1138^2=0.013$ for the first component; $0.1088^2=0.0118$ for the second component; $0.0989^2=0.0098$ for the third component; $0.5875^2=0.3452$ for the fourth component;$-0.2327^2=0.0541$ for the fifth component and $0.3778^2=0.1427$ for the sixth component. In this study, the first six principal components obtained from quality characteristics explained 85.96% of total variance.

It is understood from Table 5 that the variables that most contributed to the first component were glucose (0.2318), F+G (0.2264), fructose (0.1957), proline (0.0768), F/G ratio (0.0624) and δ^{13}C protein (0.0584) respectively. The variables that most contributed to the second component were found to be C4% (0.2943), δ13C protein-honey (0.2357), δ13C honey (0.1575), δ13C protein (0.1186) and pH (0.0620). Diastase number (0.3118), conductivity (0.2347), proline (0.1341), moisture (0.1039) and F/G ratio (0.0718) were found to be the variables that most contribute to the third component. The variables that most contributed to the fourth component were brix (0.3452), acidity (0.3211) and pH (0.1794). Conductivity (0.2378), moisture (0,1804), pH (0.1621), proline (0.1173) and acidity (0.0902) quality characteristics made the most contribution to the fifth component, while moisture (0.3036), $\delta^{13}C$ honey (0.1742), brix (0.1427) and $\delta^{13}C$ protein (0.1388) variables made the most contribution to the sixth component. According to principal component analysis, 85.96% of total variation in data set was explained by the first six factors. Average of weight values of each variable calculated from six components gives weight value (W_k) of each quality variable. For example, weight value of brix was found as;

w_1=(0.0130+0.0118+0.0098+0.3452+0.0541+0.1427)/6 = 0.096.

The highest weight values were found to be moisture (0.104), brix (0.096), conductivity (0.092), acidity (0.081), pH (0.077), diastase number (0.077), $\delta^{13}C$ protein (0.072), proline (0.069), $\delta^{13}C$ honey (0.068), C_4% (0.052), glucose (0.045), F+G (0.044), $\delta^{13}C$ protein-honey (0.043), fructose (0.042) and F/G ratio (0.036) respectively.

In comparisons according to grey relational grades (GRG), the specimen with a GRG value that is close to 1 is expressed to have the highest quality. GRG varies between 0 and 1. GRG values that are closer to 1 show that there is a high relation between real values and reference values, while GRG values that are close to 0 show a low relation. For each variable, the reference values are taken as the highest quality value (1). Accordingly, it can be stated that the closer GRG values are to 1, the higher is the quality of the honey; or the closer GRG values are to 0, the lower is the quality of honey. Grey relational grade (GRG) obtained for each specimen from raw data by taking the average of grey relational coefficients in Table 4, standard deviations of GRGs, lower and upper limits are presented in raw data column in Table 6.

However, in real practice, the effect of each factor or variable is not definitely the same. Therefore, weight values of quality characteristics should also be taken into account to calculate grey relational coefficients. In this study, mean square of quality characteristics in each eigenvector, in other words, the mean of contributions, was taken as the weight value of the related quality characteristic. Weight values that were obtained according to principal component analysis of quality variables w_1, w_2, …, w_{15} are presented in W_k column of Table 5. Grey relational grades (GRG) that were recalculated into account the weight values obtained for each quality variable, standard deviations of GRGs, lower limits and upper limits are presented in PCR column of Table 6.

Comparison of honey specimens according to grey relational coefficients that were calculated by also talking into account the weight values obtained from principal component analysis of quality variables showed that the highest quality honey specimens were S6 (0.690), S20 (0.688), S9 (0.655), S10 (0.641), S12 (0.635), S13 (0.629), S5 (0.624), S19 (0.621), S8 (0.620), S17(0.614), S15 (0.600) S4 (0.599), S1 (0.590), S14 (0.589), S2 (0.578), S7 (0.576), S11 (0.571), S3 (0.558), S18 (0.529) and S16 (0.501) respectively. Thus, it can be stated that S6 is the highest quality honey specimen, while S16 is the lowest quality honey specimen. The fact that grey relational grades of S6, S20, S9, S10, S12, S13, S5, S19, S8 and S17 were greater than mean GRG (0.605) reveals that these honey specimens are higher quality than the mean. However, a significantly high difference between GRG values while making a comparison according to GRG values between each honey specimen definitely determines the difference between the two honey specimens. It can be stated that the difference between GRG values of S6 and S17 honey specimens (0.690-0.614=0.076) are very low and thus there was no significant quality difference between S6, S20, S9, S10, S12, S13, S5, S19, S8 and S17. However, it can be stated that S6 is higher quality than S17. The fact that the difference between GRG values of S6 which has the highest GRG value and S16 which has the lowest GRG value was high (0.690-0.501=0.189) shows that there is a significant quality difference between S6 and S16 (Table 6).

Lower and upper limits of grey relational grades (GRG) show whitening range of the variable. A coefficient that is closer to 0 means it is getting darker, in other words, there is a low relationship between real values and reference values. On the other hand, a coefficient that is closer to 1 means that it is getting whiter, in other words, there is a high relationship between the real value and the reference value. The closer the lower and upper limits of GRG are to 1, the more significant is the variable of that coefficient. Besides GRG values, lower and upper limits of GRG values should also be great to talk about certain superiority in GRG values (Xuerui *et al*., 2007). Comparison of honey specimens according to lower and upper limits of GRGs revealed that the highest quality specimens were S9, S10, S7, S13, S11, S20, S6, S8, S14, S17, S15, S3, S5, S12, S2, S11, S18, S19, S4 and S16 according to lower limit and S20, S6, S4, S19, S9, S17, S13, S1, S5, S8, S15, S12, S10, S2, S14, S3, S11, S7, S16 and S18 according to upper limit. This indicates that quality grading of honey specimens is insignificant, in other words, there is no difference between the qualities of honey. Comparison of honey specimens according to lower and upper limits of GRGs that were calculated by also taking into account weight values revealed that quality ranking was S6, S20, S9, S12, S10, S13, S5, S19, S8, S17, S15, S4, S14, S1, S2, S7, S11, S3, S18 and S16 in terms of upper limits and S6, S20, S9, S10, S13, S12, S8, S5, S19, S17, S15, S4, S14, S1, S7, S2, S11, S3, S18 and S16 in terms of lower limits. The fact that also lower limits of grey relational coefficients of S6, S20, S9, S10, S13 and S12 honey specimens were higher than mean GRG shows that these honey specimens are higher quality than general mean among all honey specimens. Considering the grading of GRGs that were calculated for each specimen also taking into account weight values obtained from principal component analysis of quality variables in honey and grading of lower and upper limits of GRGs, nearly a similar ranking occurs in all three cases. This indicates that quality grading of honey specimens is significant, in other words, there is a difference between the qualities of honey. As a result, it can be stated that there is a significant superiority and inferiority between the qualities of honey specimens (S1, …, S20) according to GRG values.

It can be stated that grey relational grades (GRG) that were calculated by taking into account weight values obtained from principal component analysis of quality parameters provide more robust and consistent results than GRG values that were calculated over raw data without taking weight values into account. Each quality characteristic does not have the same effect on GRG values. It can be stated that the use of weight values of quality characteristics to calculate GRG values of each honey specimen can help to yield more real-like results in grading and classification of honey specimens according to quality and to choose higher quality honey.

Table 1. Raw data of each quality characteristics for twenty comparability sequences and their descriptive statistics values.

Sumple No	◦Brix	Moisture (%)	Conductivity mS/cm	pH	Freeacidity meq/kg	Proline mg/kg	Diastase number	Fructose	Glucose	F+G	F/G Rate	δ13C honey	δ13C Protein	δ13C Protein-honey	C4(%) (adulteration)
S1	83.76	19.30	0.202	2.65	30.00	594	13.9	38.0	29.2	67.2	1.30	-26.1	-25.7	0.4	0.0
S2	81.60	15.80	0.189	3.15	25.00	519	10.9	39.5	31.8	71.3	1.24	-25.6	-26.2	-0.6	3.6
S3	81.73	20.60	0.193	3.07	22.50	557	13.9	39.5	31.8	71.1	1.24	-25.5	-25.7	-0.2	1.3
S4	82.80	15.70	0.199	0.08	27.50	660	13.9	43.4	32.8	76.2	1.32	-25.4	-25.2	0.2	0.0
S5	84.06	14.40	0.182	2.65	27.50	483	17.9	38.4	29.0	67.4	1.32	-25.7	-25.6	0.1	0.0
S6	82.00	16.30	0.150	3.15	20.00	513	17.9	38.0	28.1	66.1	1.35	-25.8	-25.6	0.2	0.0
S7	82.57	16.20	0.180	2.98	22.50	574	17.9	38.2	30.5	68.7	1.25	-25.2	-25.4	-0.2	1.3
S8	82.85	15.40	0.160	3.17	25.00	557	17.9	41.0	32.0	73.0	1.28	-25.4	-25.4	0.0	0.0
S9	83.38	14.90	0.165	3.04	22.50	477	13.9	41.2	32.8	74.0	1.26	-25.9	-25.8	0.1	0.0
S10	82.29	15.90	0.204	2.70	22.50	545	13.9	38.8	29.0	67.8	1.34	-25.9	-25.7	0.2	0.0
S11	83.07	15.20	0.170	3.00	27.50	479	17.9	38.1	30.7	68.8	1.24	-25.5	-25.7	-0.2	1.3
S12	82.00	14.80	0.199	2.99	20.00	524	8.3	37.3	27.8	65.1	1.34	-25.7	-25.8	-0.1	0.6
S13	82.80	15.40	0.156	3.03	25.00	443	13.9	36.9	29.9	66.8	1.23	-25.8	-25.1	0.7	0.0
S14	80.59	18.20	0.167	2.75	22.50	494	23.0	37.6	30.3	67.9	1.24	-25.2	-25.5	-0.3	1.9
S15	82.78	15.30	0.206	3.17	27.50	715	13.9	39.5	32.5	72.0	1.22	-25.6	-25.8	-0.2	1.2
S16	84.16	14.80	0.174	3.04	25.00	298	8.3	23.1	17.0	40.1	1.36	-25.7	-25.3	-0.6	3.6
S17	83.34	15.50	0.172	3.10	30.00	480	13.9	41.2	31.0	72.2	1.33	-26.1	-25.9	0.2	0.0
S18	83.77	14.90	0.196	3.11	25.00	623	13.9	37.6	28.7	66.3	1.31	-25.4	-25.7	-0.3	1.9
S19	84.17	14.60	0.225	3.24	20.00	692	8.3	39.1	29.1	68.2	1.34	-25.9	-25.6	0.3	0.0
S20	84.42	14.80	0.165	2.68	20.00	447	13.9	35.9	27.6	63.5	1.30	-26.1	-25.5	0.6	0.0
CV (%)	1.22	10.22	11.11	6.39	13.26	17.91	25.82	10.34	11.48	10.70	3.64	1.09	1.11	175.40	139.40

CV: Coefficient of variation

Table 2.The correlation coefficients between the quality characteristics.

	◦Brix	Moisture %	Conductivity mS/cm	pH	Freeacidity meq/kg	Proline mg/kg	Diastase number	Fructose (F)	Glucose (G)	F+G	δ13C honey	δ13C Protein	F/G rate	δ13C Protein-honey
◦Brix	1													
Moisture %	-0.486*	1												
Conductivity mS/cm	0.120	0.074	1											
pH	-0.096	-0.215	0.047	1										
Freeacidity meq/kg	0.230	0.073	0.064	-0.069	1									
Proline. mg/kg	-0.083	0.132	0.678**	0.286	0.066	1								

Diastase number	-0.367	0.286	-0.518*	-0.320	0.128	-0.012	1							
Fructose (F)	-0.275	0.127	0.157	0.138	0.130	0.646**	0.313	1						
Glucose (G)	-0.367	0.212	0.083	0.159	0.164	0.600**	0.387	0.951**	1					
F+G	-0.321	0.168	0.124	0.149	0.148	0.633**	0.351	0.989**	0.986**	1				
δ^{13}C honey	0.400	-0.314	0.180	-0.086	-0.155	-0.133	-0.397	-0.290	-0.571**	-0.425	1			
δ^{13}C Protein	-0.506*	0.143	-0.040	0.236	-0.040	0.203	0.444*	0.071	0.194	0.129	-0.419	1		
F/G rate	-0.068	0.068	-0.178	-0.131	-0.100	0.307	0.473*	0.455*	0.435	0.451*	-0.200	0.225	1	
δ^{13}C Protein-honey	0.346	-0.058	-0.113	-0.294	-0.049	0.088	0.031	0.313	0.200	0.264	0.170	-0.611**	0.634**	1
C_4 % (adulteration)	-0.241	0.065	0.040	0.209	0.034	-0.274	-0.174	-0.548*	-0.392	-0.482*	-0.222	0.444	-0.636**	-0.868**

*:P<0.05; **:P<0.01

Table 3. Normalized values of the responses and the difference values between the comparability sequence and reference sequence

Sumple No	◦Brix	Moisture (%)	Conductivity mS/cm	pH	Freeacidity meq/kg	Proline mg/kg	Diastase number	Fructose (F)	Glucose (G)	F+G	δ^{13}C honey	δ^{13}C Protein	F/G rate	δ^{13}C Protein-honey	C_4 (%) (adulteration)
Reference sequence	**1**	**1**	**1**	**1**	**1**	**1**	**1**	**1**	**1**	**1**	**1**	**1**	**1**	**1**	**1**
S1	0.273	0.210	0.307	1.000	0.000	0.710	0.381	0.734	0.772	0.751	1.000	0.500	0.375	0.769	1.000
S2	0.835	0.774	0.480	0.153	0.500	0.530	0.177	0.808	0.937	0.864	0.444	0.917	0.750	0.000	0.000
S3	0.888	0.000	0.427	0.288	0.750	0.621	0.381	0.798	0.937	0.859	0.333	0.500	0.750	0.308	0.639
S4	0.669	0.790	0.347	0.271	0.250	0.868	0.381	1.000	1.000	1.000	0.222	0.083	0.250	0.615	1.000
S5	0.149	1.000	0.573	1.000	0.250	0.444	0.653	0.754	0.759	0.756	0.556	0.417	0.250	0.538	1.000
S6	1.000	0.694	1.000	0.153	1.000	0.516	0.653	0.734	0.703	0.720	0.667	0.417	0.063	0.615	1.000
S7	0.764	0.710	0.600	0.441	0.750	0.662	0.653	0.744	0.854	0.792	0.000	0.250	0.688	0.308	0.639
S8	0.649	0.839	0.867	0.119	0.500	0.621	0.653	0.882	0.949	0.911	0.222	0.250	0.500	0.462	1.000
S9	0.430	0.919	0.800	0.339	0.750	0.429	0.381	0.892	1.000	0.939	0.778	0.583	0.625	0.538	1.000
S10	0.880	0.758	0.280	0.915	0.750	0.592	0.381	0.773	0.759	0.767	0.778	0.500	0.125	0.615	1.000
S11	0.558	0.871	0.733	0.407	0.250	0.434	0.653	0.739	0.867	0.795	0.333	0.500	0.750	0.308	0.639
S12	1.000	0.935	0.347	0.424	1.000	0.542	0.000	0.700	0.684	0.693	0.556	0.583	0.125	0.385	0.833
S13	0.669	0.839	0.920	0.356	0.500	0.348	0.381	0.680	0.816	0.740	0.667	0.000	0.813	1.000	1.000
S14	0.417	0.387	0.772	0.831	0.750	0.470	1.000	0.714	0.842	0.770	0.000	0.333	0.750	0.231	0.472
S15	0.678	0.855	0.253	0.119	0.250	1.000	0.381	0.808	0.981	0.884	0.444	0.583	0.875	0.308	0.667
S16	0.107	0.935	0.680	0.339	0.500	0.000	0.000	0.000	0.000	0.000	0.556	1.000	0.000	0.000	0.000
S17	0.446	0.823	0.707	0.237	0.000	0.436	0.381	0.892	0.886	0.889	1.000	0.667	0.188	0.615	1.000
S18	0.269	0.919	0.387	0.220	0.500	0.779	0.381	0.714	0.741	0.726	0.222	0.500	0.313	0.231	0.472
S19	0.103	0.968	0.000	0.000	1.000	0.945	0.000	0.788	0.766	0.778	0.778	0.417	0.125	0.692	1.000
S20	0.000	0.935	0.800	0.949	1.000	0.357	0.381	0.631	0.671	0.648	1.000	0.333	0.375	0.923	1.000
					The difference values between the comparability sequence and reference sequence										
S1	0.727	0.790	0.693	0.000	1.000	0.290	0.619	0.266	0.228	0.249	0.000	0.500	0.625	0.231	0.000

S2	0.165	0.226	0.520	0.847	0.500	0.470	0.823	0.192	0.063	0.136	0.556	0.083	0.250	1.000	1.000
S3	0.112	1.000	0.573	0.712	0.250	0.379	0.619	0.202	0.063	0.141	0.667	0.500	0.250	0.692	0.361
S4	0.331	0.210	0.653	0.729	0.750	0.132	0.619	0.000	0.000	0.000	0.778	0.917	0.750	0.385	0.000
S5	0.851	0.000	0.427	0.000	0.750	0.556	0.347	0.246	0.241	0.244	0.444	0.583	0.750	0.462	0.000
S6	0.000	0.306	0.000	0.847	0.000	0.484	0.347	0.266	0.297	0.280	0.333	0.583	0.938	0.385	0.000
S7	0.236	0.290	0.400	0.559	0.250	0.338	0.347	0.256	0.146	0.208	1.000	0.750	0.313	0.692	0.361
S8	0.351	0.161	0.133	0.881	0.500	0.379	0.347	0.118	0.051	0.089	0.778	0.750	0.500	0.538	0.000
S9	0.570	0.081	0.200	0.661	0.250	0.571	0.619	0.108	0.000	0.061	0.222	0.417	0.375	0.462	0.000
S10	0.120	0.242	0.720	0.085	0.250	0.408	0.619	0.227	0.241	0.233	0.222	0.500	0.875	0.385	0.000
S11	0.442	0.129	0.267	0.593	0.750	0.566	0.347	0.261	0.133	0.205	0.667	0.500	0.250	0.692	0.361
S12	0.000	0.065	0.653	0.576	0.000	0.458	1.000	0.300	0.316	0.307	0.444	0.417	0.875	0.615	0.167
S13	0.331	0.161	0.080	0.644	0.500	0.652	0.619	0.320	0.184	0.260	0.333	1.000	0.188	0.000	0.000
S14	0.853	0.613	0.227	0.169	0.250	0.530	0.000	0.286	0.158	0.230	1.000	0.667	0.250	0.769	0.528
S15	0.322	0.145	0.747	0.881	0.750	0.000	0.619	0.192	0.019	0.116	0.556	0.417	0.125	0.692	0.333
S16	0.893	0.065	0.320	0.661	0.500	1.000	1.000	1.000	1.000	1.000	0.444	0.000	1.000	1.000	1.000
S17	0.554	0.177	0.293	0.763	1.000	0.564	0.619	0.108	0.114	0.111	0.000	0.333	0.813	0.385	0.000
S18	0.731	0.081	0.613	0.780	0.500	0.221	0.619	0.286	0.259	0.274	0.778	0.500	0.688	0.769	0.528
S19	0.897	0.032	1.000	1.000	0.000	0.055	1.000	0.212	0.234	0.222	0.222	0.583	0.875	0.308	0.000
S20	1.000	0.065	0.200	0.051	0.000	0.643	0.619	0.369	0.329	0.352	0.000	0.667	0.625	0.077	0.000

Table 4.Grey relational coefficient (GRC) for twenty comparability sequences.

Sumple No	◦Brix	Moisture (%)	Conductivity mS/cm	pH	Freeacidity meq/kg	Proline mg/kg	Diastase number	Fructose (F)	Glucose (G)	F+G	$\delta^{13}C$ honey	$\delta^{13}C$ Protein	F/G rate	$\delta^{13}C$ Protein-honey	C_4(%) (adulteration)
S1	0.407	0.388	0.419	1.000	0.333	0.633	0.447	0.653	0.687	0.667	1.000	0.500	0.444	0.684	1.000
S2	0.752	0.689	0.490	0.371	0.500	0.515	0.378	0.722	0.888	0.786	0.474	0.857	0.667	0.333	0.333
S3	0.818	0.333	0.466	0.413	0.667	0.569	0.447	0.712	0.888	0.780	0.429	0.500	0.667	0.419	0.581
S4	0.602	0.705	0.434	0.407	0.400	0.791	0.447	1.000	1.000	1.000	0.391	0.353	0.400	0.565	1.000
S5	0.370	1.000	0.540	1.000	0.400	0.473	0.590	0.670	0.675	0.672	0.529	0.462	0.400	0.520	1.000
S6	1.000	0.620	1.000	0.371	1.000	0.508	0.590	0.653	0.627	0.641	0.600	0.462	0.348	0.565	1.000
S7	0.680	0.633	0.556	0.472	0.667	0.597	0.590	0.661	0.775	0.706	0.333	0.400	0.615	0.419	0.581
S8	0.587	0.756	0.789	0.362	0.500	0.569	0.590	0.809	0.908	0.849	0.391	0.400	0.500	0.481	1.000
S9	0.467	0.861	0.714	0.431	0.667	0.467	0.447	0.822	1.000	0.891	0.692	0.546	0.571	0.520	1.000
S10	0.807	0.674	0.410	0.855	0.667	0.551	0.447	0.688	0.675	0.682	0.692	0.500	0.364	0.565	1.000
S11	0.531	0.795	0.652	0.457	0.400	0.469	0.590	0.657	0.790	0.709	0.429	0.500	0.667	0.419	0.581
S12	1.000	0.886	0.434	0.465	1.000	0.522	0.333	0.625	0.612	0.619	0.529	0.546	0.364	0.448	0.750
S13	0.602	0.756	0.862	0.437	0.500	0.434	0.447	0.610	0.731	0.658	0.600	0.333	0.727	1.000	1.000
S14	0.462	0.449	0.688	0.747	0.667	0.485	1.000	0.636	0.760	0.685	0.333	0.429	0.667	0.394	0.486
S15	0.608	0.775	0.401	0.362	0.400	1.000	0.447	0.722	0.963	0.811	0.474	0.546	0.800	0.419	0.600
S16	0.359	0.886	0.610	0.431	0.500	0.333	0.333	0.333	0.333	0.333	0.529	1.000	0.333	0.333	0.333
S17	0.475	0.738	0.630	0.396	0.333	0.470	0.447	0.822	0.814	0.819	1.000	0.600	0.381	0.565	1.000

S18	0.406	0.861	0.449	0.391	0.500	0.694	0.447	0.636	0.658	0.646	0.391	0.500	0.421	0.394	0.486
S19	0.358	0.939	0.333	0.333	1.000	0.901	0.333	0.702	0.681	0.693	0.692	0.462	0.364	0.619	1.000
S20	0.333	0.886	0.714	0.908	1.000	0.438	0.447	0.575	0.603	0.587	1.000	0.429	0.444	0.867	1.000
Mean	0.581	0.731	0.580	0.530	0.605	0.571	0.490	0.685	0.753	0.712	0.576	0.516	0.507	0.527	0.787
GRC	(7)	(3)	(8)	(11)	(6)	(10)	(15)	(5)	(2)	(4)	(9)	(13)	(14)	(12)	(1)

Table 5. Eigenvectors of the principal component analysis on the physicochemical parameters and contribution of each individual physicochemical quality characteristic for the principal component.

Quality characteristic	Eigenvectors of Principal Component						Contribution of each individual physicochemical quality characteristic						
	1	2	3	4	5	6	1	2	3	4	5	6	W_k
◦Brix	0.1138	-0.1088	0.0989	0.5875	-0.2327	0.3778	0.0130	0.0118	0.0098	0.3452	0.0541	0.1427	0.096
Moisture (%)	-0.1631	0.0735	-0.3223	0.0797	0.4247	-0.5510	0.0266	0.0054	0.1039	0.0064	0.1804	0.3036	0.104
ConductivitymS/cm	-0.0732	0.1257	0.4845	0.2428	0.4876	-0.0103	0.0054	0.0158	0.2347	0.0590	0.2378	0.0001	0.092
pH	-0.1683	0.2489	0.1710	-0.4236	-0.4026	0.0608	0.0283	0.0620	0.0292	0.1794	0.1621	0.0037	0.077
Freeaciditymeq/kg	-0.1527	0.1173	0.0295	0.5667	-0.3004	-0.1945	0.0233	0.0138	0.0009	0.3211	0.0902	0.0378	0.081
Proline mg/kg	0.2771	-0.0068	-0.3662	-0.0157	0.3425	-0.2954	0.0768	0.0000	0.1341	0.0002	0.1173	0.0873	0.069
Diastasenumber	0.1094	-0.0415	0.5584	-0.1655	-0.1537	-0.2954	0.0120	0.0017	0.3118	0.0274	0.0236	0.0873	0.077
Fructose (F)	0.4424	0.1715	-0.1285	0.0317	0.0753	0.0753	0.1957	0.0294	0.0165	0.0010	0.0057	0.0057	0.042
Glucose (G)	0.4815	0.0127	-0.0056	-0.0468	0.1589	0.0993	0.2318	0.0002	0.0000	0.0022	0.0252	0.0099	0.045
F+G	0.4758	0.1055	-0.0737	-0.0053	0.1182	0.0887	0.2264	0.0111	0.0054	0.0000	0.0140	0.0079	0.044
$\delta^{13}C$ honey	-0.1783	0.3969	-0.1496	-0.1449	0.0492	0.4174	0.0318	0.1575	0.0224	0.0210	0.0024	0.1742	0.068
$\delta^{13}C$ protein	-0.2417	-0.3444	-0.2247	-0.0782	0.2394	0.3726	0.0584	0.1186	0.0505	0.0061	0.0573	0.1388	0.072
F/G rate	0.2498	-0.2064	0.2680	-0.1501	0.1336	0.0129	0.0624	0.0426	0.0718	0.0225	0.0178	0.0002	0.036
$\delta^{13}C$ protein-honey	-0.0604	0.4855	0.0824	0.0107	0.1011	0.0259	0.0036	0.2357	0.0068	0.0001	0.0102	0.0007	0.043
C_4 (%) (adulteration)	0.0664	0.5425	-0.0454	0.0915	0.0423	0.0102	0.0044	0.2943	0.0021	0.0084	0.0018	0.0001	0.052
Total							1.0	1.0	1.0	1.0	1.0	1.0	1.0

Eigenvalue of each principal components

Component	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
Total	3.931	3.037	2.114	1.607	1.179	1.025	0.862	0.438	0.421	0.195	0.084	0.063	0.042	0.001	0.000
% of Variance	26.21	20.24	14.10	10.71	7.86	6.83	5.75	2.92	2.81	1.30	0.56	0.42	0.28	0.01	0.00
Cumulative (%)	26.21	46.45	60.55	71.26	79.12	85.96	91.70	94.63	97.43	98.73	99.29	99.71	99.99	100.00	100.00

Table 6. Grey relational grade and its order for twenty comparability sequences.

Honey No	According to Raw Data γ_i	Order	S_{γ_i}	$\gamma_i \pm S_{\gamma_i}$	According to W_k of PCA γ_i	Order	S_{γ_i}	$\gamma_i \pm S_{\gamma_i}$
S1	0.618	12	0.230	0.388 - 0.848	0.590	13	0.016	0.570 - 0.606
S2	0.584	15	0.193	0.391 - 0.777	0.578	15	0.018	0.560 - 0.596
S3	0.579	16	0.169	0.410 - 0.748	0.558	18	0.014	0.544 - 0.572
S4	0.633	7	0.260	0.373 - 0.893	0.599	12	0.015	0.583 - 0.614
S5	0.620	11	0.219	0.401 - 0.839	0.624	7	0.023	0.601 - 0.647
S6	0.666	3	0.228	0.438 - 0.894	0.690	1	0.026	0.664 - 0.716
S7	0.579	17	0.124	0.455 - 0.703	0.576	16	0.015	0.561 - 0.591
S8	0.634	6	0.204	0.430 - 0.838	0.620	9	0.018	0.603 - 0.638
S9	0.673	2	0.201	0.472 - 0.874	0.655	3	0.017	0.638 - 0.672
S10	0.639	5	0.172	0.467 - 0.811	0.641	4	0.018	0.622 - 0.659
S11	0.576	18	0.132	0.444 - 0.708	0.571	17	0.016	0.556 - 0.588
S12	0.609	13	0.213	0.396 - 0.822	0.635	5	0.026	0.609 - 0.661
S13	0.647	4	0.203	0.444 - 0.850	0.629	6	0.018	0.611 - 0.647
S14	0.593	14	0.178	0.415 - 0.771	0.589	14	0.017	0.571 - 0.606
S15	0.622	10	0.212	0.410 - 0.834	0.600	11	0.017	0.583 - 0.617
S16	0.465	20	0.214	0.251 - 0.679	0.501	20	0.024	0.477 - 0.525
S17	0.633	8	0.221	0.412 - 0.854	0.614	10	0.017	0.597 - 0631
S18	0.525	19	0.141	0.384 - 0.666	0.529	19	0.018	0.511 - 0.546
S19	0.627	9	0.253	0.374 - 0.880	0.621	8	0.023	0.598 - 0.644
S20	0.682	1	0.241	0.441 - 0.923	0.688	2	0.024	0.664 - 0.712
Mean	0.610		0.203	0.407 - 0.813	0.605		0.019	0.586 - 0.624

S_{γ_i} : Standart Deviation; $\gamma_i \pm S_{\gamma_i}$: Lower and Upper Limit

Acknowledgments: We declare that we have no financial and personal relationships with other people or organizations that can inappropriately influence our work; there is no professional or other personal interest of any nature or kind in any product, service, and/or company that could be construed as influencing the position presented in the manuscript entitled "Comparison of quality characteristics in honey using grey relational analysis and principal component analysis methods "This article does not contain any studies with human or animal subjets. We would like to thanks Mahir Murat Cengiz and Murat Tosun for their contribution in the compilation of this study.

Conflict of Interest: Mehmet TOPAL and Aycan Mutlu YAĞANOĞLU declare that they have no conflict of interest.

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