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Thermal properties of honey as affected by the addition of sugar syrup

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ABSTRACT

Ensuring the authenticity of honey is a priority for producers and regulatory authorities. The aim of this work was to evaluate the thermal properties (using a Differential Scanning Calorimeter "DSC") of ten types of sugar syrup, six types of honey and the mixtures of sunflower honey with all these syrups at different proportions simulating the adulteration of honey (ratio honey/syrup: 80/20; 90/10; 95/05). The glass transition temperature (Tg midpoint) ranged from 60.2 ºC to 67.3 ºC in honey samples and from 32.8 ºC to 95.8 ºC in syrup samples. The differences in sugar composition of the syrups mainly affect their thermal properties. In the adulterated samples, the glass transition temperature was affected by the type of syrup, proportionally to the adulteration level. These results offer compelling evidence that the DSC can be used for the identification of addition of syrup to honey, although to be conclusive a greater number of honey types must be considered.

Keywords: differential scanning calorimetry, adulteration, glass transition, honey, syrup.

1. Introduction

Food fraud is the economically motivated adulteration of any edible product for financial gain. Many food fraud databases reporting these incidents in Europe in recent years have highlighted that honey is highly vulnerable to food fraud as it represents about

the 90% of all entries related to sweeteners (Food Fraud Database, 2016; FoodSHIELD, 2016; RASFF, 2016). Honey adulteration has to be seen from different perspectives: (1) Public Health, as it involves the presence of uncontrolled ingredients that can cause serious health problems when the adulterant is toxic, or allergenic in sensitive people (Everstine et al., 2013); (2) Legal, as it is strictly forbidden to add anything to honey; this requirement is established in Codex Alimentarius and has been adopted by E.U. legislation and some U.S. states (Codex Alimentarius Commission, 1981; Europa, 2010; United States Food and Drug Administration, 2011); and (3) Economic, by unfair competition involving the industry, distributors and the livelihood of beekeepers, leading to a destabilization of markets. Therefore, guaranteeing the authenticity of honey has become a very imperative matter for the international honey market (processors, retailer, beekeepers), regulatory authorities and consumers.

Honey can be exposed to fraud worldwide. One of the most common types of adulteration of honey involves its dilution with other less expensive (three to five fold) sugar syrups such as corn, cane, agave and specially rice syrup, among others. Rice syrup is widely used in some Asian countries, which are the origin of most of the European and U.S. imports (United States International Trade Commission, 1994). The importance of detecting the presence of this kind of syrup in honey is proven by the existence of classical analytical techniques that are used specifically for this syrup.

In recent years, a large number of analytical methods have been used to differentiate genuine honey from adulterated. Among them, SCIRA (stable carbon isotope mass spectrometry) and NMR spectroscopy are the most recognized (Elflein & Raezke, 2008; Bertelli et al., 2010; De Oliveira et al., 2014). These techniques are very expensive, requiring highly specialized equipment and are time-consuming. Moreover, in order to get conclusive results for one sample it would be necessary to use the results obtained by

applying the combination of several of these techniques. The industrial laboratories do not have this instrumental capability; therefore, the major bottleneck in the application of these techniques is the limited number of samples that can be analysed in specialized laboratories due to both, time and financial restrictions.

The new tendency in analysis is focused on the development of alternative analytical procedures that not only enable rapid screening, but are also cheaper and greener than the traditional ones (Reference). Among them, Differential Scanning Calorimetry (DSC) has some advantages over other classical detection methodologies; it is a relatively fast technique that does not require any solvent and thus it is environmental friendly technique. Moreover, this technique uses a very small amount of sample and little preparation. Several investigations have already used DSC to study the adulteration of different kinds of food since this technique facilitates the analysis of various food components such as proteins, fats and carbohydrates (Dahimi et al., 2014; Tomaszewska-Gras, 2016). The use of DSC to assess the authenticity of sweeteners is based on the fact that each of them has its intrinsic characteristics and composition. However, there is very limited data in the literature about using melting curves for the assessment of honey authenticity; among them, the work reported by Cordella et al. in 2002, stands out. This paper proved that DSC could be a powerful technique for detecting the presence of beet and cane syrup in honey samples. Nevertheless, to be conclusive, it would be necessary to increase the number type of samples analysed both for honey and syrups. In addition, previous published studies (Cordella et al., 2002; Lupano, 1997) did not take into account the possible artefact provoked by the presence of water in the sample, since the plasticizer effect of water can distort the results of the thermal properties. In this sense, the present work presents an improvement over previous studies since samples were submitted to lyophilization to remove the water content for samples.

74 The aim of this work was to apply DSC to evaluate adulteration of honey by the
75 addition of different types of syrup.

76 **2. Materials and methods**

77 2.1. Materials

78 Six types of raw honey harvested in 2016 in different areas of Spain, provided by the
79 company Melazahar (Montroy, Valencia), were used in this study: sunflower (*Helianthus*
80 *annuus*); orange blossom (*Citrus spp*.), rosemary (*Rosmarinus officinalis*), heather (*Erika*
81 *spp*.), polyfloral honey and forest. These botanical categorization was performed by
82 means of pollen analysis, which was quantified following the recommendations of the
83 International Commission for Bee Botany (Von Der Ohe et al., 2004). Furthermore, in
84 the present study syrups from different origins were used: agave (Natural Bioaprica,
85 Spain), maple (Maple Joe, Canada), sugar cane (Ingenio Nuestra Señora del Carmen,
86 Spain), barley (La Finestra sul Cielo, Italy); corn (Roquette Laissa, Spain); five types of
87 rice syrup from different brands: Arroz biocesta, Spain (Rice I); Danival, France (Rice
88 II); Mandolé, Spain (Rice III), La Finestra sul Cielo, Italy (Rice IV); and husked rice
89 (Mitoku Macrobiotic, Japan).

90 The samples evaluated in the present work were: 6 types of pure honey, 10 types of
91 pure syrup and the mixture of sunflower honey with all the types of syrup at different
92 proportions simulating the adulteration of honey (ratio honey/syrup: 80/20; 90/10; 95/05).

93 2.2. Moisture evaluation

94 Water content of samples was determined using a refractrometer (Abbe-type model
95 T1 Atago, USA) and the Chataway tables in accordance with the Harmonized Methods
96 of the European Honey Commission (Bogdanov, 2009). The residual moisture of samples
97 was obtained by calculating the weight difference before and after lyophilization using
98 an analytical balance (PB303-L, Mettler Toledo).

99 2.3. Sugar Analysis

100 Fructose, glucose, sucrose and maltose were analyzed as described by Bogdanov et
101 al., (1997) using a Liquid Cromatograph (Agilent Technologies modelo 1120 Compact
102 LC, Germany) with an Evaporative Light Scattering Detector (Agilent Technologies
103 modelo 1200 Series, Germany) and a Waters Carbohydrate column (4.6 x 250 mm, 4
104 μm). The separation of the different sugars was performed in isocratic mode with water
105 and acetonitrile (20/80) at a flow rate of 0.8 mL/min. The elution was finished in 14
106 minutes. Detector conditions were: temperature 50 °C, gas pressure (N_2) 3.5 bars and gain
107 = 6. The analysis of the data was performed with the software EZChrom Elite.
108 Quantification of sugars was carried out using the calibration curves of the corresponding
109 external standards. The quantification limits of the four sugars studied were 0.1 g/100 g
110 honey.

111 2.4. Protein content

112 Protein content was measured by Kjeldahl procedure (AOAC, 2000). In order to avoid
113 the interference that pollen could cause in the quantification of proteins, honey samples
114 were previously centrifuged.

115 2.5. Differential scanning calorimetry (DSC)

116 2.5.1. Sample preparation

117 Before determination of the thermal properties of the samples, the first step was to
118 remove their moisture since previous studies demonstrate that moisture greatly interferes
119 with the measurements of these properties (Kántor et al., 1999). It was possible to remove
120 more than 98% of water by lyophilization (LyoAlfa, Telstar, Spain). Since honey and
121 syrups do not have freezable water, it was necessary to dilute them in distilled water (1 g
122 sample/10 g water) before lyophilization (Ospina, 2014). Diluted samples were placed in
123 aluminium containers (5 mL in each container) and frozen at -40 °C for 24 h, at 130

mmHg. In order to remove the residual moisture, lyophilized samples were introduced in a desiccator with P_2O_5 (Panreac, Barcelona, Spain) to reach constant weight.

2.5.2. DSC determination

Thermal properties of the samples were obtained by means of a Differential Scanning calorimeter (Mettler Toledo, DSC1, Suiza) equipped with an intracooler. Nitrogen (99.99% purity at 20 mL/min) was the purge gas used. The equipment was calibrated with indium (ΔH_f = 28.5 J/g) and zinc (ΔH_f = 103.7 J/g). Dehydrated samples of 9-10 mg were weighed into aluminium pans (40 µL, ME-26763, AL-CRUCIBLES) covered and sealed on the sample platform and then micro-perforated. All samples were subjected to the following temperature cycle: from 25 ºC to -40 ºC (rate of 10 ºC/min); from -40 ºC to 110 ºC (rate of 10 ºC/min) and held for 5 min. After that a cooling scan was applied from 110 ºC to -40 ºC and finally the temperature was increased to 120 ºC.

The glass transition temperature at the beginning (Tg onset) and in the middle (Tg midpoint) of each sample was obtained using Mettler Toledo DSC STARe SW 9.20 software. The analysis of each sample was carried out in triplicate.

2.6. Statistical Analysis

An analysis of variance (ANOVA) using Statgraphics Centurion 16.1 was applied to study the influence of the type of honey, syrup and their mixtures on the thermal properties (Tg onset and Tg midpoint) and sugar content of the samples. LSD (least significant difference) at significance level α = 5% was used to analyse the differences between samples.

In addition, the data were analysed using principal components analysis: PCA, applying the software Unscrambler X.10. The variables analysed by PCA were centered and weighted in order to compensate for the different scales of the variables. Statistical assumptions for this analysis were checked previously, which indicated that PCA analysis

149 was suitable for the dataset (KMO > 0.8, Barlett's statistic p > 0.001). For all the PCA
150 analyses carried out in this study, the internal consistency and reliability of each
151 component was assessed using Cronbach's alpha (a > 0.9).

152 **3. Results and Discussion**

153 3.1. Sugar content and glass transition temperature of pure honey and pure syrup samples

154 Table 1 shows the content (g/100g dry matter) of main sugars (fructose, glucose,
155 sucrose and maltose), and glass transition temperature obtained for the different
156 representative types of raw pure honey: three monofloral honey samples (sunflower,
157 orange blossom and rosemary), two honeydew honey samples (forest and heather) and
158 one polyfloral honey. The same parameters were analysed in ten types of pure syrup
159 samples from different sources: rice from different brands (I, II, III, IV), husked rice,
160 corn, maple, barley, sugar cane and agave. In addition, this table shows the ANOVA
161 results (F-ratio and significant differences) obtained for the factors "type of honey", "type
162 of syrup" and "honey samples and syrup samples".

163 As expected, fructose was the most dominant sugar followed by glucose in all cases
164 (Persano-Oddo & Piro, 2004). In this study, the monofloral honey samples (rosemary,
165 orange blossom and sulflower) had the highest fructose level (average = 55.3, 54 and 46
166 g/100 g dm, respectively). The same occurs for glucose, but in this case sunflower honey
167 showed a significant high concentration (average = 40.5 g/100 g dm), whereas rosemary
168 honey samples and orange blossom showed average values of 34 g/100 g dm. It is
169 common to find high levels of glucose in sunflower honey samples, even if they came
170 from different countries (Juan-Borrás et al., 2014). The sucrose content was less than 0.05
171 g/100 g dm in all cases. In general, the level of this sugar is not important in honey
172 although some specific types of honey such as acacia and hedysarum honey could contain
173 values above 2.5 g/100 g and 3 g/100 g, respectively (Persano-Oddo et al. 1995; Juan-

174 Borrás et al., 2014). Maltose is also a minor sugar; its content in different types of
175 monofloral and forest honey is quite low, not exceeding 1.5 g/100 g (Persano-Oddo et al.
176 1995). In this work the maltose content in polyfloral honey (0.21 g/100 g dm) and heather
177 honey (1.44 g/100 g dm) was higher than the amount found in the rest of the samples. In
178 general, the sugar composition of the honey samples analyzed in the present study are in
179 the usual range, considering that sugar content strongly depends on the type of
180 flowers/plant secretions used by the bees, and therefore varies with the type of honey.

181 As expected, proteins were present in low concentrations in all honey samples
182 (Mohammed & Kamran, 2012). The total protein content ranged from 0.18 g/100 g dm
183 in rosemary honey samples to 0.71 g/100 g dm in honeydew honey.

184 In general, the analysed syrup samples showed significant differences in terms of
185 sugar content as compared to honey samples, especially in the case of fructose. Unlike
186 honey, all rice syrup samples showed significantly low fructose content. On the contrary,
187 agave syrup showed a significantly high fructose content, which is two times higher than
188 the typical level found in honey samples. There was a wide range of variability in terms
189 of glucose content: from 0.05 g/100 g dm in maple syrup to 51.1 g/100 g dm in Rice IV
190 syrup sample. In general, glucose concentration in rice syrup samples was in the same
191 range that the amount found in honey samples, except for husked rice (3.99 g/ 100 g dm).
192 As shown for honey samples, and with the exception of sugar cane and agave syrup,
193 sucrose was present in negligible amounts in syrup samples (< 0.05 g/100 g dm). On the
194 contrary, sucrose-rich syrup samples showed negligible amounts of maltose (< 0.05 g
195 /100 g dm).

196 Maltose content in the syrup samples, was significantly higher than the above
197 mentioned content of this sugar in pure honey samples. The level of maltose was

especially important in husked rice syrup (52 g/100 g dm) and barley syrup (61 g/100 g dm).

The protein content of syrup samples was very similar or even lower than the protein content of honey samples, except for barley syrup (1.1 g/100 g dm).

Figure 1 shows the typical DSC thermograms and the glass transition obtained for honey and syrup samples (1^{st} and 2^{nd} heating scan). The glass transition temperature obtained in the second scan was slightly higher, which points to a loss of some residual water during the first heating of the sample. Thus, the values that are shown in Table 1 were obtained from the second scan, in which all the samples are supposed to be almost completely anhydrous. As shown in Table 1, the highest values of glass transition temperature (Tg onset and Tg midpoint) were found for monofloral honey samples, which are also the ones that showed the highest fructose level. Sunflower honey showed intermediate values of Tg midpoint.

Syrup samples showed a wider range of variability of glass transition temperature values (Tg midpoint ranging from 32.8 ºC to 95.8 ºC) than those obtained for honey samples (Tg midpoint ranging from 60.2 ºC to 77.3 ºC); this is because the syrup samples, unlike honey samples, have a very different sugar composition.

Sunflower and orange blossom honey samples showed intermediate behaviour in terms of Tg and sugar composition. However, sunflower is more common and can be found worldwide (Juan-Borrás et al., 2014), thus this type of honey was the one chosen to evaluate the impact of the adulteration with different amount of syrup samples.

3.2. Effect of syrup on the thermal properties of honey

Figure 2 shows, as an example, different typical DSC thermograms of sunflower honey samples adulterated with 20% of rice, sugar cane and hushed rice syrups. While in

all the pure honey samples evaluated in the present study only a glass transition temperature was detected, in some of the adulterated samples, two glass transition temperatures appeared. Thus, in order to evaluate the effect of the addition of syrup on the thermal behaviour of the samples, the temperature of the second glass transition was chosen in all cases.

Table 2 shows the glass transition temperature and the estimated sugar and protein content of sunflower honey samples adulterated with different levels of the evaluated samples of syrup. The glass transition temperature was affected by the type of syrup and the adulteration level, being the interaction between these two factors also significant. The addition of increasing amounts of syrup led to a significant decrease in the glass transition value of the samples except for agave, corn and husked rice. Barley and maple syrup addition at 20% led to the highest decrease in the glass transition temperature (approx. 50 ºC). These two types of syrup showed the highest level of maltose and sucrose, respectively.

With the purpose of evaluating from a descriptive point of view, the global effect of the level of adulteration of honey on its glass transition temperature and the composition of adulterated samples (sugars and proteins), a principal component analysis (PCA) was performed (Figure 3) including the results reported in Table 1 and 2. This analysis was carried out using the average values for each sample. This unsupervised procedure permitted to check if there was a spontaneous classification from the data obtained, without previously defining the categories of the samples.

In this PCA plot (Figure 3) pure syrup samples were located just in the opposite side of pure honey samples with the only exception of agave syrup that was placed in the same quadrant that honey. This figure shows that the different types of honey (placed in the inferior right quadrant) and adulterated samples are well differentiated. Samples

containing 20% of maple or barley syrup and 80% of sunflower honey were in the same position as pure syrup samples. In this figure, PC1 is the component that explained the differences among samples: pure syrup, adulterated sunflower honey samples and different types of honey. Glass transition temperature, fructose and maltose content are the variables that had the highest influence on the differences among samples, being fructose and glass transition temperature positively correlated.

In order to discuss, in more detail, the influence of the addition of different proportions of syrup to honey, a second PCA (Figure 4) was carried out considering only the samples located in the highlighted area in Figure 3: mixtures with sunflower honey and different types of pure honey. Figure 4 shows the PCA bi-plot of scores and loading obtained. In this case, three components explained 97% of the total variance. PC1 explained the 91% and was mainly positively correlated with the glass transition temperature, located at the right end of PC1. The second component, PC2, explained the 6% and was positively correlated with fructose and negatively with maltose. In general, adulterated samples are located around pure sunflower honey; the lower the adulteration level the shorter the distance between pure honey and samples containing syrup. The differences of adulteration level (20%, 10% or 5%) are shown in the plot by means of circles. Almost all H80:20 samples (80% pure honey and 20% syrup) are in the external circle; H90:10 samples (90% pure honey and 10% syrup) are at the center circle and H:95:5 samples (95% pure honey and 5% syrup) are very close to pure honey samples. In general, the increase in adulteration level promoted a movement towards the left quadrant, except for agave syrup, which showed an opposite trend due to its high fructose content. Rice syrup behaved in a similar way in spite of the adulteration level and especially RI, RII and RIII samples, which implied their similar behavior in terms of the parameters analyzed.

It is important to point out that for some syrup types the effect of the highest adulteration level in the thermal properties of honey is more marked. This is the case of H80:M20 and H80:B20 samples. Moreover, sugar cane syrup is the one that showed the lowest effect on all the evaluated parameters since all the samples containing this syrup are located near pure honey at all adulteration levels and inside the circle for 5% adulteration level.

4. Conclusions

The addition of sugar syrup promoted significant changes in the thermal properties of adulterated samples as compared to pure honey samples, gradually the adulteration level. The evaluation of the thermal properties of honey by means of Differential Scanning Calorimetry provided information on the possible presence of added sugar syrup in sunflower honey. Further studies are required to validate these results in other types of honey.

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Table 1. Major sugars, total proteins and glass transition temperature (Tg onset and Tg midpoint) of pure honey and syrup samples. Mean values and standard deviation (in brackets).

Sample	Major sugars (g/100 g dry matter)					Glass transition temperature (ºC)	
	Fructose	**Glucose**	**Sucrose**	**Maltose**	**Total Proteins**	**Tg_{Onset}**	**$Tg_{Midpoint}$**
Pure honey							
Forest	37 (4)^{a,5}	23.9 (1.2)^{a,3,4}	<0.05^{1}	<0.05^{a,1}	0.71 (0.08)^{c,10}	50.5 (0.3)^{a,6}	60.2 (0.5)^{a,5}
Heather	44 (2)^{b,6}	28 (3)^{a,b,5}	<0.05^{1}	1.44 (0.09)^{c,1}	0.40 (0.07)^{b,9}	54.6 (0.5)^{b,6,7}	64.5 (0.4)^{b,5,6,7}
Polyfloral	52 (5)^{c,d,7}	31 (3)^{b,c,5,6}	<0.05^{1}	0.21 (0.09)^{b,1}	0.42 (0.03)^{b,9}	57 (2)^{b,7,8}	67 (3)^{b,6,7}
Sunflower	46 (3)^{b,c,6}	40.5 (1.3)^{d,7}	<0.05^{1}	<0.05^{a,1}	0.24 (0.00)^{a,5,6,7,8}	65 (3)^{c,9,10}	72.0 (1.3)^{c,8,9}
Orange blossom	54 (2)^{d,7,8}	34 (2)^{c,6}	<0.05^{1}	<0.05^{a,1}	0.25 (0.03)^{a,6,7,8}	65.23 (0.13)^{c,9}	75.27 (0.13)^{d,9,10}
Rosemary	55.3 (0.6)^{d,8}	34 (3)^{c,6}	<0.05^{1}	<0.05^{a,1}	0.18 (0.07)^{a,2,3,4,5}	69.5 (1.8)^{d,10}	77.3 (0.6)^{d,10}
ANOVA F-ratio	15.49*	15.65*		352.26*	36.89*	57.55*	69.84*
Pure syrup							
Rice I	<0.05^{A,1}	24 (3)^{D,4}	<0.05^{A,1}	32 (4)^{C,D,3}	0.3 (0.3)^{C,D,7,8,9}	59 (2)^{F,8}	68 (3)^{E,7,8}
Rice II	<0.05^{A,1}	34 (4)^{F,6}	<0.05^{A,1}	28 (3)^{B,C,2}	0.15 (0.09)^{A,B,C,1,2,3,4}	33 (5)^{B,C,3,4}	39 (7)^{B,C,2}
Rice III	<0.05^{A,1}	20.4 (0.6)^{C,3}	<0.05^{A,1}	27.3 (1.5)^{B,2}	0.4 (0.2)^{D,8,9}	30 (2)^{B,2,3}	37 (4)^{B,C,1,2}
Rice IV	<0.05^{A,1}	51.1 (1.2)^{H,8}	<0.05^{A,1}	37 (4)^{E,4}	0.093 (0.006)^{A,B,1,2,3}	28 (3)^{B,2}	40 (4)^{A,B,2,3}
Husked Rice	<0.05^{A,1}	3.99 (1.12)^{B,2}	<0.05^{A,1}	52 (2)^{F,5}	0.22 (0.09)^{B,C,D,5,6,7}	88 (3)^{H,11}	95.83 (1.05)^{G,11}
Corn	9 (3)^{C,3}	41.3 (1.7)^{G,7}	<0.05^{A,1}	32.9 (0.7)^{D,3}	0.03 (0.01)^{A,1}	23 (4)^{A,1}	32.8 (1.7)^{A,1}
Barley	3.93 (0.05)^{B,2}	23 (2)^{C,D,3,4}	<0.05^{A,1}	61 (2)^{G,6}	1.10 (0.07)^{E,11}	37 (3)^{C,4}	45 (2)^{C,3}
Maple	<0.05^{A,1}	<0.05^{A,1}	85.94 (1.06)^{C,3}	<0.05^{A,1}	0.07 (0.02)^{A,B,1,2}	44 (2)^{D,5}	53 (2)^{D,4}
Sugar cane	25 (3)^{D,4}	28 (2)^{E,5}	31.73 (1.03)^{B,2}	<0.05^{A,1}	0.10 (0.05)^{A,B,1,2,3}	53.5 (1.6)^{E,6,7}	63 (3)^{E,5,6}
Agave	97 (3)^{E,9}	8 (3)^{B,2}	0.33 (0.13)^{A,1}	<0.05^{A,1}	0.04 (0.01)^{A,1,2}	66.4 (1.6)^{G,9,10}	75.18 (1.09)^{F,9,10}
ANOVA F-ratio	1105.59*	173.79*	10515.39*	306.94*	28.87*	156.55*	107.01*
ANOVA F-ratio (syrup and honey samples)	527.03*	108.84*	10847.8*	485.59 *	29.74*	157.03*	118.75*

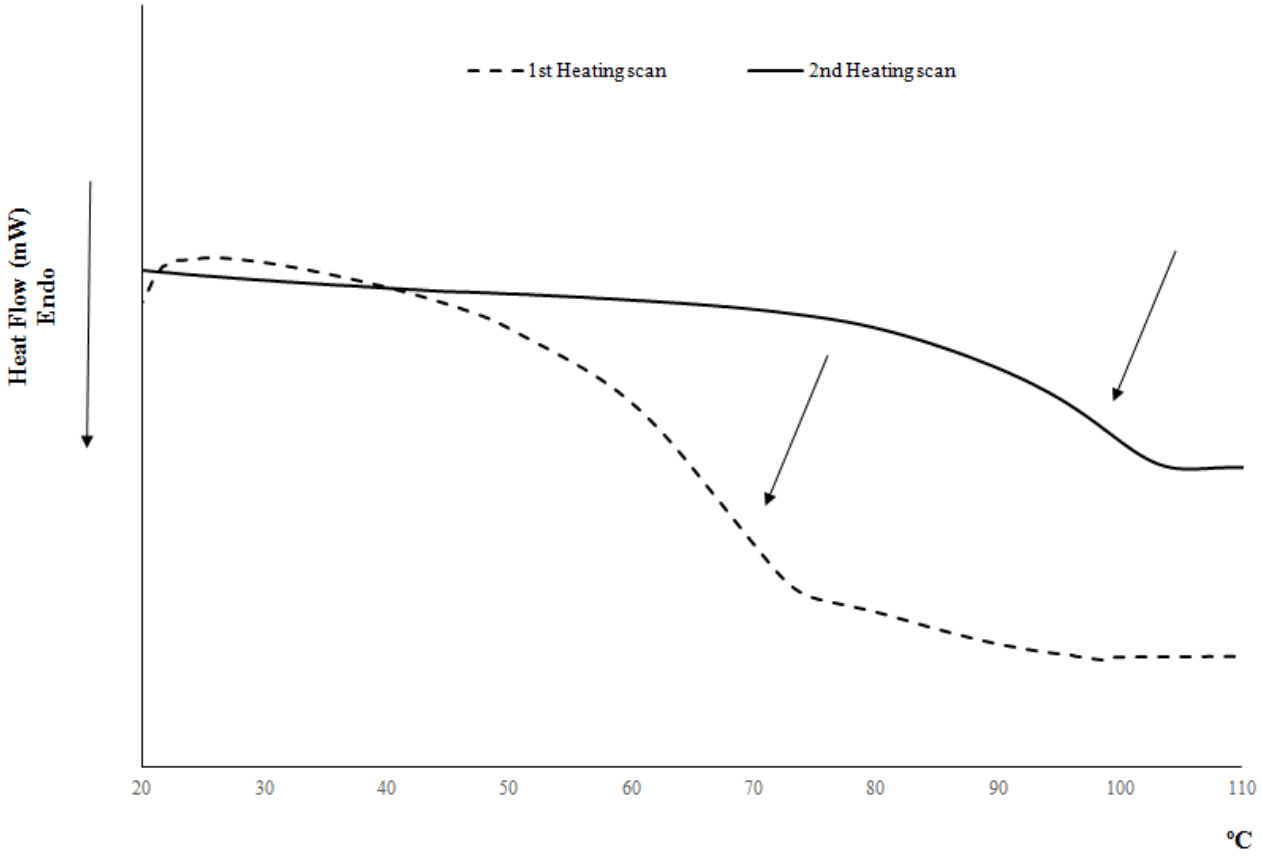
Different letters in the same column indicate differences (*p <0.001) among honey samples (a-d), syrup samples (A-H) or honey samples and syrup samples (1-11)

Table 2. Glass transition temperature (Tg) and estimated composition of adulterated samples. Mean values and standard deviation, in brackets.

		Glass transition temperature (ºC)		Estimated Composition (g / 100 g dm)				
Syrup	**Sample**	**Tg_{Onset2}**	**$Tg_{Midpoint2}$**	**Fructose**	**Glucose**	**Sucrose**	**Maltose**	**Proteins**
Agave (A)	H80:A20	68.9 (1.6)^{g,1}	77.8 (0.8)^{f,1}	55.67	34.32	0.06	0.00	0.205
	H90:A10	68 (2)^{g,2}	75.7 (1.9)^{f,2}	50.79	37.45	0.03	0.00	0.224
	H95:A5	62.8 (0.1)^{g,3}	70.56 (0.05)^{f,3}	48.38	39.00	0.02	0.00	0.233
Maple (M)	H80:M20	11.8 (1.3)^{b,1}	17.6 (0.5)^{b,1}	38.58	34.00	13.85	0.00	0.215
	H90:M10	69.7 (0.4)^{b,2}	77.9 (0.6)^{b,2}	42.37	37.34	6.76	0.00	0.229
	H95:M5	63.9 (1.9)^{b,3}	73.9 (1.4)^{b,3}	44.20	38.96	3.34	0.00	0.236
Rice IV (R)	H80:R20	66.5 (0.4)^{h,1}	73.84 (0.05)^{g,1}	36.92	42.61	0.00	7.27	0.213
	H90:R10	68.7 (1.7)^{h,2}	76.2 (0.9)^{g,2}	41.46	41.57	0.00	3.63	0.228
	H95:R5	70.9 (1.8)^{h,3}	80.05 (0.04)^{g,3}	43.73	41.05	0.00	1.81	0.235
Rice I (RI)	H80:RI20	53.9 (1.2)^{c,1}	63.6 (1.4)^{c,1}	36.75	37.27	0.00	6.43	0.258
	H90:RI10	54.5 (0.3)^{c,2}	63.6 (0.5)^{c,2}	41.36	38.90	0.00	3.22	0.250
	H95:RI5	59 (3)^{c,3}	69 (3)^{c,3}	43.68	39.72	0.00	1.61	0.246
Rice II (RII)	H80:RII20	58.7 (1.6)^{e,1}	69.9 (1.4)^{e,1}	36.83	39.27	0.00	5.65	0.224
	H90:RII10	60.6 (1.5)^{e,2}	71 (2) ^{e,2}	41.41	39.90	0.00	2.82	0.233
	H95:RII5	59.8 (0.2)^{e,3}	69.7 (0.7)^{e,3}	43.70	40.22	0.00	1.41	0.238
Husked Rice (HR)	H80:HR20	66 (2)^{f,1}	73.8 (0.6)^{e,1}	36.71	33.16	0.00	10.53	0.238
	H90:HR10	57.7 (0.6)^{f,2}	68 (1)^{e,2}	41.34	36.84	0.00	5.27	0.240
	H95:HR5	61.9 (0.8)^{f,3}	71.6 (0.5)^{e,3}	43.66	38.69	0.00	2.64	0.241
Rice III (RIII)	H80:RIII20	57.0 (1.3)^{d,1}	65 (2)^{d,1}	36.82	36.52	0.00	5.43	0.268

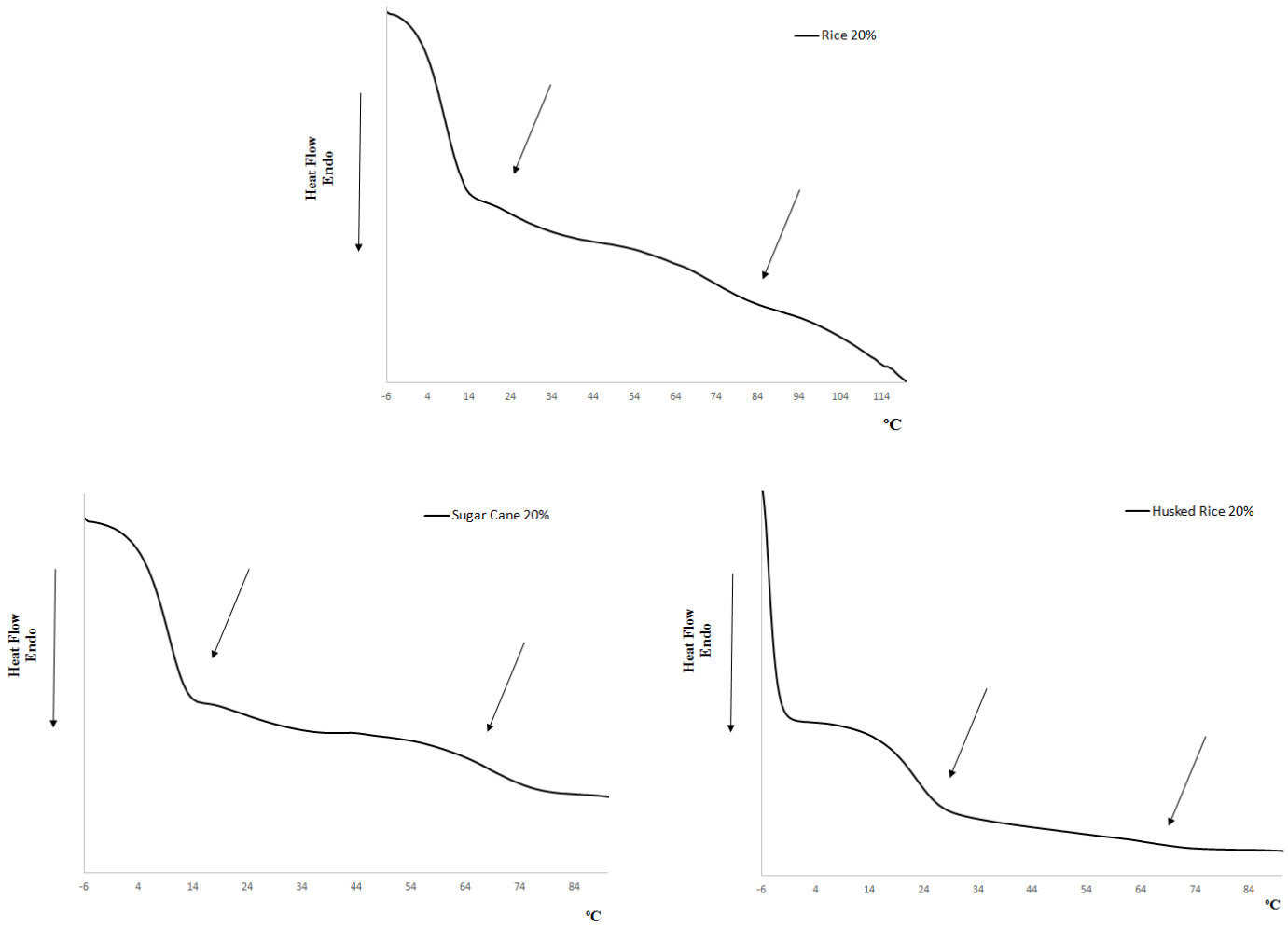
	H90:RIII10	57.7 (0.3)^{d,2}	67.8 (0.8)^{d,2}	41.40	38.53	0.00	2.72	0.255
	H95:RIII5	59.2 (1.5)^{d,3}	70.5 (1.9)^{d,3}	43.70	39.53	0.00	1.36	0.249
	H80:SC20	63 (3)^{g,1}	70.9 (1.5)^{f,1}	41.92	38.14	6.26	0.00	0.214
Sugar Cane (SC)	H90:SC10	66.75 (1.25)^{g,2}	76.6 (0.7)^{f,2}	43.96	39.34	3.12	0.00	0.228
	H95:SC5	68.4 (0.9)^{g,3}	77.5 (1.3)^{f,3}	44.97	39.94	1.56	0.00	0.235
	H80:B20	20 (6)^{a,1}	26 (5)^{a,1}	37.66	37.07	0.00	12.15	0.413
Barley (B)	H90:B10	53.2 (1.2)^{a,2}	62.5 (0.5)^{a,2}	41.83	38.81	0.00	6.07	0.328
	H95:B5	59.9 (1.3)^{a,3}	69 (4)^{a,3}	43.91	39.67	0.00	3.03	0.285
	H80:C20	72.9 (0.4)^{h,1}	80.4 (1.5)^{h,1}	38.50	40.69	0.00	6.66	0.200
Corn (C)	H90:C10	67.6 (0.9)^{h,2}	76 (2)^{h,2}	42.24	40.61	0.00	3.34	0.221
	H95:C5	68.2 (1.5)^{h,3}	78.5 (0.8)^{h,3}	44.11	40.57	0.00	1.67	0.232
ANOVA F-ratio (type of syrup)		209.03*	231.76*					
ANOVA F-ratio (adulteration level)		262.94***	375.08*					
Interaction (syrup x adulteration level)		135.58*	157.11*					

Different letters in the same column indicate differences (*p <0.001) due to the type of syrup (a-h) or adulteration level (1-3).



362

363 **Figure 1**



364

365 **Figure 2**

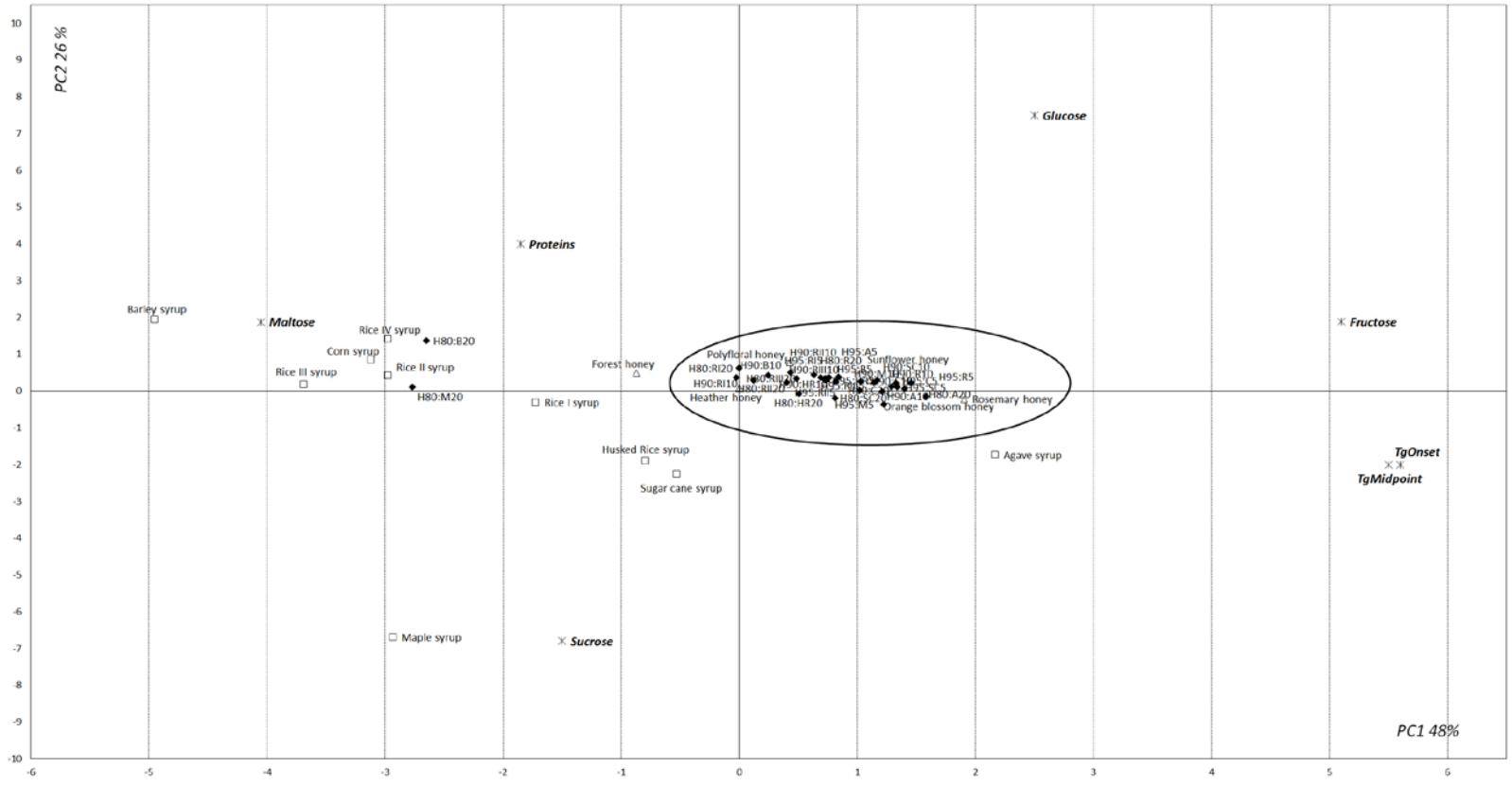
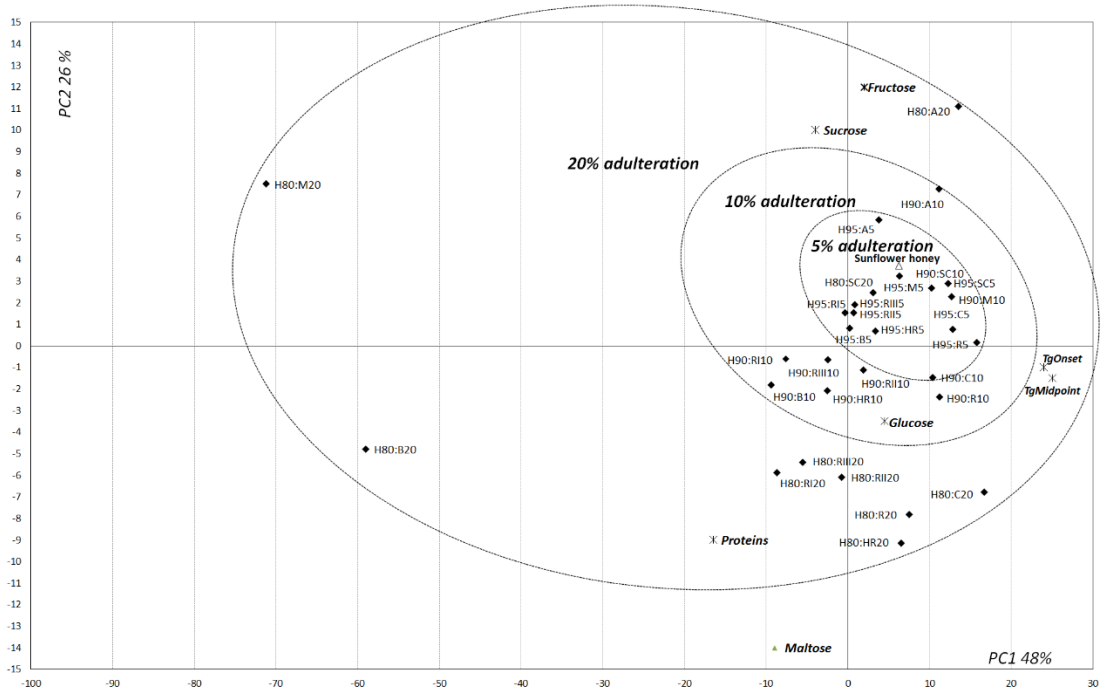

PC2 26 %
PC1 48%
Glucose
Proteins
Fructose
Maltose
Sucrose
TgOnset
TgMidpoint
Barley syrup
Rice IV syrup
H80:B20
Corn syrup
Rice III syrup
Rice II syrup
H80:M20
Rice I syrup
Forest honey
Polyfloral honey
H90:RI10
H95:A5
H80:RI20
H90:B10
H95:RI5
H80:R20
Sunflower honey
H90:RI10
H95:R5
Heather honey
H80:HR20
H95:M5
Orange blossom honey
Rosemary honey
Agave syrup
Husked Rice syrup
Sugar cane syrup
Maple syrup

Figure 3



368

369 **Figure 4**

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