

# Application News

Fourier Transform Infrared Spectrophotometer (FTIR)

# No. A629

### Quantitative Analysis of Sugars (Fructose, Glucose, and Sucrose) in Honey by FTIR

Honey has attracted attention as a health food which contains vitamins, minerals, and other nutrients, and is also used in foods and beverages. However, some products are susceptible to adulteration, for example, by intentionally adding cheap corn syrup, in order to reduce manufacturing costs. Although adulteration of honey with corn syrup does not cause any serious health problems, the resulting loss of consumer confidence has an adverse effect on market growth. Therefore, in quality control, the development of a simple analytical technique which makes it possible to determine whether substitutes have been added to honey has been demanded.

Infrared (IR) spectrophotometry is an effective technique for identifying the components contained in honey because organic compounds each display a different spectrum, and chemometrics (PLS: partial-least squares method) and multiple regression analysis of the IR spectra obtained by IR spectrophotometry enables quick quantitative analysis of multiple components.

In this article, a quantitative analysis of the sugars contained in several honey samples was conducted using a Shimadzu Fourier transform IR spectrophotometer.

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#### Analysis Method

Pure honey consists mainly of sugar. Virtually the entire sugar content is fructose and glucose, together with a small amount of sucrose. As percentage values, pure honey consists of 33-43 % fructose, 25-35 % glucose, and 0-2 % sucrose, and its fructose: glucose ratio is 1.2:1. On the other hand, adulterated products containing corn syrup consist mainly of glucose. Therefore, if a honey product displays an elevated level of glucose, it can be inferred that corn syrup has been added in order to reduce the cost of production.

In this experiment, nine types of commercial honey were prepared and diluted to 10 % w/w with pure water. The samples were then measured with a Shimadzu IRTracer™-100 Fourier transform IR spectrophotometer (FTIR) and a Quest™ single-bounce attenuated total reflectance (ATR) accessory (ZnSe prism), and their fructose, glucose, and sucrose contents were quantified by a chemometric analysis (PLS method).

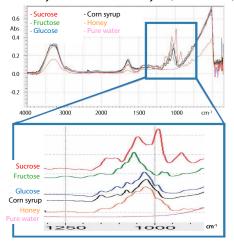


Fig. 1 IR Spectra of Sucrose, Fructose, Glucose, Corn Syrup, Honey (All 50 % Aqueous Solutions), and Pure Water

Table 1 shows the measurement conditions, Fig. 1 shows the IR spectra of sucrose, fructose, glucose, corn syrup, honey (in all cases, 50 % aqueous solutions), and pure water, and Fig. 2 shows the IR spectra of the nine commercial honey samples (10 % w/w).

#### **Table 1 Measurement Conditions**

Instruments

: IRTracer-100
Quest ATR accessory

Resolution
: 4 cm<sup>-1</sup>

Accumulation
: 32 times

Wavenumber range
: 4000 - 600 cm<sup>-1</sup>

Apodization function
: Happ-Genzel
Detector
: DLATGS

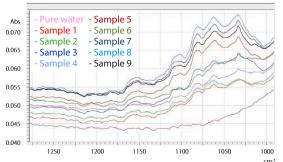


Fig. 2 IR Spectra of Commercial Honey Samples (10 % w/w)

#### Quantitative Analysis of Sugars in Honey

In order to prepare a calibration curve from a mixed aqueous solution of fructose, glucose, and sucrose, a 3-dimensional sample training matrix (model showing the mixing ratios of the 3 components in the standard sample) was prepared, as shown in Fig. 3, and the concentration of the standard sample necessary for quantitation of the sugars contained in the honey samples was studied. The percentages (0-15 %) of fructose, sucrose, and glucose are shown on the X, Y, and Z axes of the model in Fig. 3, and the concentration of the standard sample was decided so as to include the entire 3-dimensional space of the model. Chemometrics (PLS method) was used in the quantitative analysis. Table 2 shows a list of the standard samples. The PLS calibration curve was prepared using 25 of the 34 samples, and the calibration curve was verified using the remaining 9 samples.

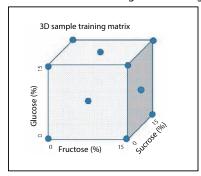


Fig. 3 Three-Dimensional Sample Training Matrix (Model Showing Mixing Ratios of 3 Components in the Standard Sample)

**Table 2 List of Standard Samples** 

Concentration of sugars (% w/w)						
Sample	Fructose	Glucose	Sucrose			
1	0.00	0.00	0.00			
2	5.08	0.00	0.00			
3	10.06	0.00	0.00			
4	0.00	5.00	0.00			
5	0.00	10.22	0.00			
6	0.00	0.00	4.97			
7	0.00	0.00	9.94			
8	4.92	4.91	0.00			
9	0.00	5.10	4.95			
10	4.95	0.00	4.91			
11	10.10	10.55	0.00			
12	0.00	9.83	9.88			
13	0.43	0.00	10.03			
14	5.10	5.06	5.04			
15	10.11	9.84	9.94			
16	3.84	7.83	2.68			
17	7.94	5.03	1.75			
18	1.83	4.67	0.73			
19	0.48	2.94	3.07			
20	4.95	6.39	1.47			
21	3.99	2.66	7.21			
22	3.56	3.53	9.63			
23	4.97	4.96	9.95			
24	10.13	5.05	5.05			
25	4.92	9.84	4.94			
26	14.95	0.00	0.00			
27	0.00	14.99	0.00			
28	0.00	0.00	14.72			
29	15.14	15.26	0.00			
30	15.31	0.00	15.24			
31	0.00	15.15 15.16				
32	0.65	14.98 7.59				
33	14.89	7.45 14.97				
34	7.53	15.14 15.21				

#### **Table 3 PLS Calibration Report**

	<u> </u>				
Algorithm	PLS I				
Number of components	3				
Number of standard samples	25				
Wavenumber range (cm <sup>-1</sup> )	963 - 1486				
Component	Fructose	Glucose	Sucrose		
Number of factors	5	5	5		
Correlation coefficient	0.9990	0.9987	0.9986		
Square of correlation coefficient	0.9980	0.9973	0.9973		
MSEP	0.0019	0.0026	0.0026		
SEP	0.0441	0.0506 0.0513			

Table 3 shows the PLS calibration report. The correlation coefficients of all sugars were satisfactory, at 0.99 or more, and the values of MSEP (mean square error of prediction) and SEP (standard error of prediction) were also small.

Table 4 shows the results of the quantitative analysis of the sugars contained in the nine commercial honey samples. Samples 1 to 5, which were labelled "100 % pure honey," had high ratios of glucose to fructose, suggesting addition of corn syrup. Sample 6 was labelled "7 % pure honey," but because it contained a high percentage of sucrose, it is considered possible that refined sugar was used in the preparation of that product.

#### Conclusion

A simple quantitative analysis of the sugars contained in honey was possible by FTIR measurement and analysis by chemometrics. The results of a quantitative analysis of commercial honeys showed component compositions different from the label information, suggesting addition of low-cost substitutes. The FTIR analysis method can be used as an efficient technique for analysis of sugars in quality control of food products.

#### <References>

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#### Table 4 Results of Quantitative Analysis of Sugar in Commercial Honey Samples

Sample	Label information	Concentration of sugars (% w/w)			Ratio
	Laberinionnation	Fructose	Glucose	Sucrose	Fructose / Glucose
1	100 % pure honey	0.081	0.242	0.00	0.33
2	100 % pure honey	0.119	0.180	0.00	0.66
3	100 % pure honey	0.186	0.367	0.00	0.51
4	100 % pure clover honey	0.031	0.236	0.00	0.13
5	100 % pure honey	0.279	0.404	0.00	0.69
6	Made with 7 % pure honey	0.00	0.299	0.089	0.00
7	None	0.462	0.428	0.00	1.08
8	None	0.536	0.464	0.00	1.16
9	Grade A	0.379	0.363	0.00	1.04

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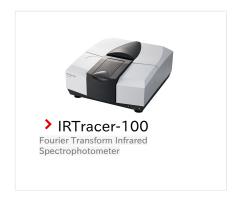
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