



The Use of a Mid-IR Fiber Optic Probe in Sugar Analysis

by Aline Dimitri;

McGill University, Dept. of Food Science

The use of FT-IR spectroscopy for the determination of the sucrose content in sugar cane juice has been demonstrated (1). This important analysis is often used as a basis for cane sugar payment in the sugar cane industry. Quantitative analysis of aqueous solutions of sugars by IR spectroscopy is generally based on the bands in the 1250-800 cm^{-1} region. The major bands in this region correspond to C-O-H bending vibrations. Although individual mono- and disaccharides each have a characteristic set of absorption bands in this region, mixtures of these sugars have bands that are extensively overlapped. Individual sugars in aqueous solution can be readily quantitated by FT-IR spectroscopy with the use of the ATR analysis technique (2). Kemsley et al. (3) employed this technique to record the spectra of sugar mixtures (sucrose, glucose, and fructose) in water and used the K-matrix and P-matrix approaches to obtain calibrations for the individual sugars. While the results from the K-matrix approach did not prove satisfactory, the P-matrix calibrations yielded predictions for soft drink samples that were in reasonable agreement with high-pressure liquid-chromatographic (HPLC) data. The calibration standards employed were reconstituted sugar cane juices, prepared by adding sucrose, glucose, and fructose to water at the levels found in natural juices. Correlations between the FT-IR spectral data for these samples, obtained using the ATR technique, and their sucrose content were established by principal components regression (PCR).

An important quality control application that has been investigated is the determination of dextrose equivalent (DE) and dry substance (DS) measures, employed in corn syrup production, by FT-IR spectroscopy (4). DE is a particularly important measure of the sweetening power which develops as corn starch is converted to syrup, but the standard copper sulfate titration method for determination of DE is tedious and time consuming. PLS calibrations have been obtained for both DE and DS, and, with the use of a flow-through ATR accessory, the FT-IR technique could provide a means of continuous on-line process measurement. In another study (5), PLS regression was applied in the analysis of the three main components (glucose, maltose, and fructose) of the dry substance in glucose syrups. The samples were diluted in distilled water (1 g/2 mL), owing to the high level of dry matter in these syrups (~70% w/v), and their spectra were recorded using a horizontal ATR sampling accessory. The FT-IR predicted concentrations for glucose, maltose, and fructose were within 3-5% of the HPLC data for these samples, with the higher end of this error range corresponding to samples containing high levels of unquantified oligosaccharides.

On a more fundamental level, the utility of FT-IR spectroscopy as a technique for the quantitative measurement of the open-chain form of sugars has been demonstrated (6, 7). An absorption band in the spectrum of D-fructose in D₂O solution at 1728 cm^{-1} has been assigned to the open form (6), and this assignment was confirmed by isotopic substitution (7). FT-IR spectroscopy thus allows the quantitation of the open-chain tautomers of keto/aldehyde reducing sugars, which is important in determining their relative reactivities in the formation of Maillard reaction intermediates, and the monitoring of the concentration of the open-chain form as a function of temperature and pH. Basic quantitative investigations of this nature have important ramifications in terms of predicting color and flavor formation in products undergoing heating, especially for microwave applications (8).

In the present study, the fiber optic-ATR probe (FO/ATR) was used for the quantitative analysis of sugar solutions containing fructose, glucose, and sucrose.

Sugar solutions (70% wt/wt) were prepared by adding 1.5 ml of water to a total of 3.5 grams of the sugars (fructose, glucose and sucrose) in accordance with Table 1. The FO/ATR probe was thoroughly cleaned and a background spectrum of air was recorded. The FO/ATR probe was inserted into a tube containing water and the spectrum in water was recorded. The FO/ATR spectra of the different sugar solutions were recorded. Between each measurement the FO/ATR probe was thoroughly rinsed with water. The FT-IR spectra were recorded on a Nicolet 205 spectrometer purged with dry air. A total of 512 scans were co-added at a resolution of 4 cm^{-1} using a DTGS detector with a gain setting of 16 with one order of zero-filling. In a parallel study the FT-IR spectra of the same sugar solutions were recorded on a germanium (Ge) HATR (8 x 1 cm) (approximately 6 reflections). A total of 256 scans were co-added at a gain of 8.

Figure 1 shows the FT-IR spectrum of a 70% total sugar solution (40% sucrose, 12% fructose & 18% glucose: sample #3) recorded with the FO/ATR probe (Figure 1) and on a Ge HATR (Figure 2). The maximum absorbance at 1050 cm^{-1} is approximately 0.4 employing the fiber optic probe vs 1.05 on a Ge ATR. The single beam spectra of the same sugar solutions show that the cutoff of the FO/ATR probe is at approximately 950 cm^{-1} , while that of Ge ATR is at ~800 cm^{-1} . The noise at ~1000 cm^{-1} was approximately 2-3 higher for the FO/ATR probe compared to the Ge ATR. This comparison was based on the ratio of two

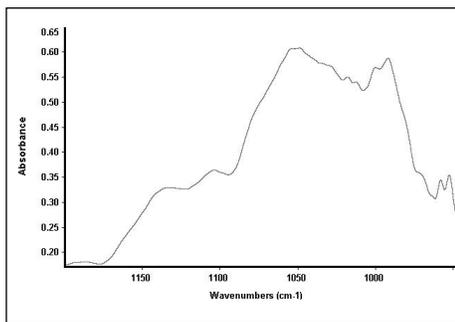


Figure 1: 70% Total Sugar Solution recorded with an FO/ATR probe - milk with water subtracted.

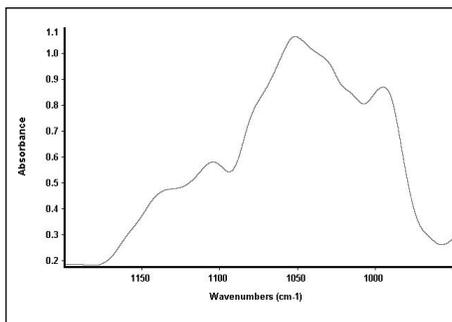


Figure 2: 70% Total Sugar Solution recorded on Ge HATR.

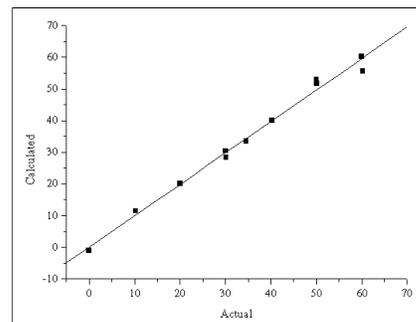


Figure 3: Calculated vs. actual plot for sucrose obtained with the Fiber Optic/ATR probe.

consecutive single-beam spectra of water recorded by employing the FO/ATR probe (512 scans at a gain of 16) and compared to the ratio of two water single-beam spectra recorded on a Ge ATR (256 scans, gain of 8).

Individual PLS calibration models with FO/ATR probe for sucrose, fructose, and glucose were developed using the 12 solutions in *Table 1* as calibration standards. A plot of calculated vs actual %sucrose from "leave-one-put" cross validation is shown in *Figure 3* ($R = 0.984$). Comparable performance was obtained from the PLS model for sucrose based on spectra recorded with the Ge HATR accessory.

The above results indicate that a PLS calibration for the quantitative analysis of the three sugars can be effectively carried out employing either a Ge HATR or the FO/ATR probe. The FO/ATR probe is much easier to use and more amenable to automation. The loss of spectral information (below 1000 cm^{-1}) does not appear to significantly deteriorate the performance of the calibration. Thus, the FO/ATR probe can be employed in sugar analysis.

Future work is directed at determining the limit of detection using the FO/ATR probe and developing calibration models designed for the analysis of carbohydrates in beverages, syrups, and juice concentrates. In each step a comparison of the performance of the FO/ATR probe to the Ge HATR accessory will be carried out. Other work will also be undertaken to determine the usefulness of the FO/ATR probe to monitor sugar hydrolysis in real time. Hydrolysis is commonly used in the food industry to increase and stabilize the sweetness of the product. For example, sucrose in syrups (~ 70% wt/v) is hydrolyzed to glucose and fructose to enhance the sweetness of the syrup (fructose is 1.2 times sweeter than sucrose). These processes are currently monitored by taking an aliquot of the sample every few hours and checking the refractive

index of the solution to estimate the extent of the hydrolysis or inversion reaction. This study indicates that a FO/ATR system could be used for at-line or on-line analysis of sugars in the soft drink and syrup industries.

Table 1: Table of Prepared Solutions

Sample #	Sucrose (g)	Fructose (g)	Glucose (g)	% Total Sugar	% Sucrose	% Fructose	% Glucose
1	3.0104	0.2497	0.2517	70.236	60.208	4.994	5.034
2	2.5061	0.4011	0.5993	70.13	50.122	8.022	11.986
3	2.0114	0.5994	0.9107	70.43	40.228	11.988	18.214
4	1.5072	1.2043	0.812	70.47	30.144	24.086	16.24
5	1.0016	1.5101	1.0005	70.244	20.020	30.202	20.01
6	0.5107	1.5989	1.4017	70.226	10.214	31.978	28.034
7	0	1.7261	1.7301	69.124	0	34.522	34.602
8	1.7268	0.7769	0.7593	65.26	34.536	15.538	15.186
9	3.0015	0	0	60.03	60.03	0	0
10	2.5021	0.5109	0	60.26	50.042	10.218	0
11	2.0102	0	1.0006	60.216	40.204	0	20.012
12	1.5014	0.7601	0.751	60.25	30.028	15.202	15.02

References

1. F. Cadet, D. Bertrand, P. Robert, J. Maillot, J. Dieudonné, and C. Rouch, *Appl. Spectrosc.* 45, 166-172 (1991).
2. J. Hopkins and J. Newberry, *FT-IR Spectral Lines* 7, 2 (1986).
3. E. K. Kemsley, Li Zhuo, M. K. Hammouri, and R. H. Wilson, *Food Chem.* 44, 299-304 (1992).
4. M. P. Fuller, M. C. Garry, and Z. Stanek, *Am. Lab.* 22, 58-69 (1990).
5. F. de Lène Mirouze, J. C. Boulou, N. Dupuy, M. Meurens, J. P. Huvenne, and P. Legrand, *Appl. Spectrosc.* 47, 1187-1191 (1993).
6. V. A. Yaylayan and A. A. Ismail, *J. Carbohydr. Chem.* 11, 149-158 (1992).
7. V. A. Yaylayan, A. A. Ismail, and S. Mandeville, *Carbohydr. Res.*, in press.
8. V. Yaylayan, A. A. Ismail, and A. Huyghues-Despointes, in *Maillard Reaction: Relevance to Food, Health and Disease* (T. Labuza and G. Reineccius, eds.), Birkhauser Verlag, Basel, pp. 69-74 (1995).

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