Application News

No. FTIR-1401

Spectrophotometric Analysis

Quantification of Natural Sugars in Baby Food Products by MID FTIR Spectroscopy

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■ Introduction

No other food products focus consumer attention as those that are prepared for consumption by children. Of current interest are the natural and added sugar contents of processed baby foods and juices. Identification and quantification of natural sugars was investigated in baby food products by mid-IR Fourier Transform Infrared (FTIR) spectroscopy. Using Horizontal Attenuated Total Reflectance (HATR), neat baby food samples were analyzed without need for extensive sample preparation. By use of the HATR technique it was demonstrated that high sensitivity could be easily achieved without significant effect from water content. Factor space chemo-metric analysis was used to establish a robust method that allowed the confident measurement of sugar concentrations in these food products. The method was developed using a training matrix of three naturally occurring sugars, fructose, glucose, and sucrose. The method was confirmed using a verification matrix and was found to be readily applicable to the evaluation of sugar quantities occurring in commercial baby food products. Several commercial products were analyzed with this method and quantities of fructose, glucose, and sucrose were determined.

■ Analytical Concept – FTIR Analysis

Baby food samples pose a unique challenge for FTIR analysis because of the strong IR absorption by water. Figure 1 shows the FTIR absorption spectra of water (red) and a 5% aqueous glucose solution (black) acquired using a Horizontal Attenuated Total Reflection (HATR) accessory with a trough liquid plate.[1, 2, 3] The strong absorption bands due to water can be seen between 3800 – 2800, 1700 – 1550, and below 900 cm^{-1}. However, absorption from the glucose can be seen in the fingerprint region between 1486 and 963 cm^{-1}. This water-free absorption region suggests that quantitative sugar analysis in aqueous solutions may be feasible.

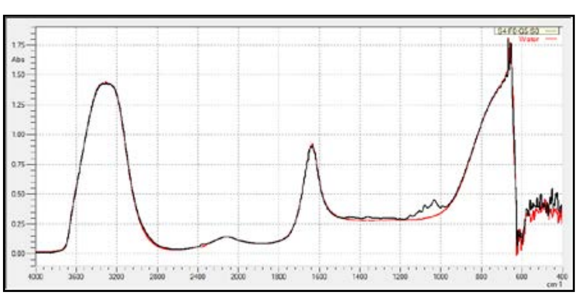

Figure 1: FTIR spectra for water and a 5% aqueous glucose

FTIR spectra of aqueous solutions of 5% fructose (blue), 5% glucose (green), and 5% sucrose (black) are shown in figure 2. The sugars have characteristic absorption bands that appear in the 1486 – 963 cm^{-1} range. The absorption bands overlap making quantitation by least squares or traditional multivariate analysis routines difficult.

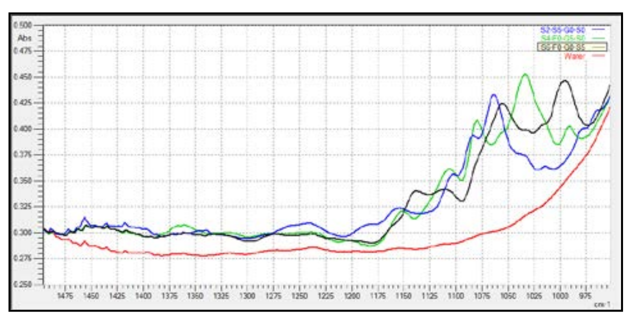

Figure 2: FTIR spectra of 5% aqueous solutions of water (red), fructose (blue), glucose (green), and sucrose (black)

■ Analytical Concept – Factor Space

Chemometric factor-space analysis was utilized to establish simultaneous calibration curves for the three-sugar aqueous mixtures. Partial Least Squares (PLS) was selected as the factor-space routine of choice. The use of a factor-space analysis routine increased the number of dimensions in the analysis.[4] This allowed for each sugar component to be assigned to a specific dimension or factor-space. In addition, noise in the spectra (e.g. water absorption) was also treated by the additional dimensions. By using the factor-based routines, the components that attributed to analytical noise (e.g. water absorptions) could readily be identified and separated out in the quantitation.

A training set of samples was prepared to cover the full 3-dimensional quantitative space required for analysis of the baby foods. Since there were three sugar components of interest, fructose, glucose, and sucrose, a three dimensional training matrix was created (figures 3 and 4). Aqueous sugar samples were prepared to cover the eight corners of the matrix, the face centered positions of the matrix, and the matrix center.

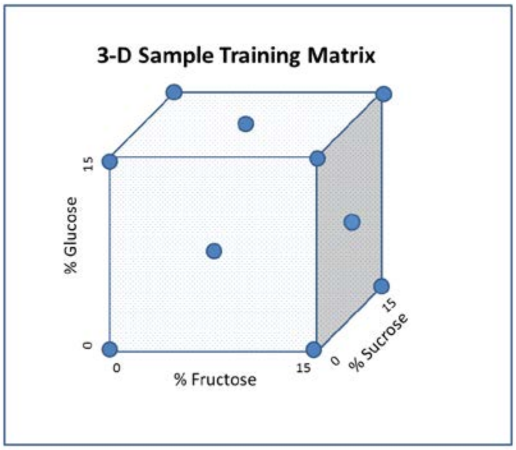

Figure 3: 3-Dimensional cube showing placement of training matrix calibration samples

In addition to the training matrix, a verification matrix (figure 5) of aqueous sugar samples was prepared using random concentrations of each of the three sugars, fructose, glucose, and sucrose. The verification matrix was used to evaluate the validity of the calibration method.

Sugar Concentration (Mass Percent)			
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	10.21	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	14.97	14.98	7.59
33	14.89	7.45	14.97
34	7.53	15.14	15.21

Sugar Concentration (Mass Percent)			
Sample	Fructose	Glucose	Sucrose
35	1.39	12.90	0.70
36	9.58	13.77	1.16
37	2.33	13.55	14.71
38	12.92	3.95	2.58
39	6.86	10.63	9.23
40	9.90	10.82	9.89

Figure 4 (top): Actual training matrix for aqueous sugar standards
Figure 5 (bottom): Verification matrix of random aqueous sugar standards

■ Calibration Results

FTIR Absorbance spectra were acquired of the training matrix standard samples using parameters of 4 cm^{-1} resolution, Happ-Genzel apodization, and the averaging of 32 scans. Figure 6 shows the spectra and demonstrates the complexity of the overlapping absorption bands.

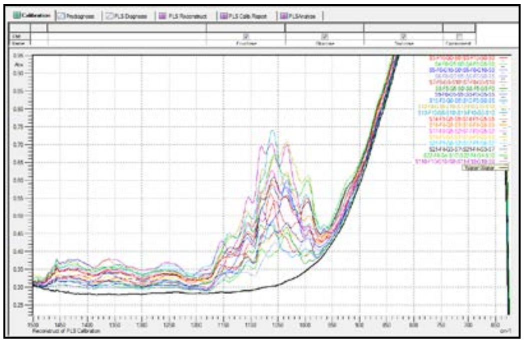

Figure 6: FTIR spectra of training matrix aqueous sugar standards

A Partial Least Squares (PLS) algorithm was utilized to establish a calibration curve for each of the three sugar components. It was found that good correlation could be achieved without the use of pre-processing methods such as smoothing, derivatives, or zero corrections. In addition, the use of five factors accounted for all of the noise in the spectra and provided good calibration curves with acceptable R^2 values (figure 7).

PLS Calibration Report			
Calibration Table:	Quant_1to25_a.irqc		
Algorithm:	PLS I		
Number of components:	3		
Number of references:	25		
Range[1]:	963.00 - 1486.00		
Preprocess:			
Scale:	None		
Component:	Fructose	Glucose	Sucrose
Number of factors:	5	5	5
Correlation coeff.:	0.999	0.9987	0.9986
Square of correlation coeff.:	0.998	0.9973	0.9973
MSEP:	0.0019	0.0026	0.0026
SEP:	0.0441	0.0506	0.0513
X Leverage warnings:	3	3	3
Y Residual warnings:	1	0	2

Figure 7: PLS Calibration Report showing R^2 values for each sugar calibration curve

To further demonstrate that the use of 5 factors was appropriate, the P Loadings for each sugar component were examined. As seen from the graphs (figure 8), the P Loading of the 5th factor resembles a random noise spectrum, suggesting that all of the spectral noise had been accounted for.

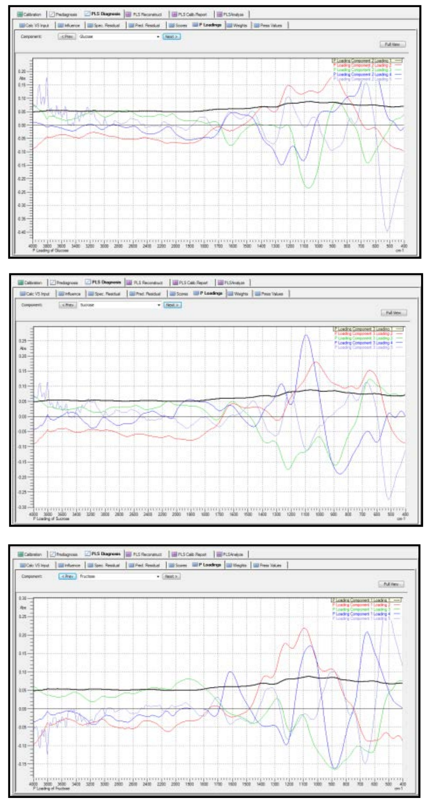

Figure 8: P Loading graphs for each sugar supporting the selection of five factors

■ Calibration Validation

Once the calibration curve for each sugar component were established, FTIR spectra were acquired of the verification matrix of samples using the same acquisition parameters as was used for the training matrix (figure 9).

Results of the verification matrix showed average residuals for each sugar of 0.004% and established that the calibration method was valid.

Spectrum	Fructose	*Resid. 1*	Glucose	*Resid. 2*	Sucrose	*Resid. 3*
S35-F1-G13-F1	1.638	*0.005*	13.088	*0.005*	1.076	*0.005*
S36-F10-G14-S1	9.41	*0.003*	14.036	*0.003*	1.762	*0.003*
S37-F2-G14-S15	1.589	*0.005*	13.591	*0.005*	15.156	*0.005*
S38-F13-G4-F3	12.189	*0.006*	3.411	*0.006*	2.642	*0.006*
S39-F7-G11-S9	6.636	*0.002*	10.813	*0.002*	9.655	*0.002*
S40-F10-G11-S10	9.213	*0.003*	11.151	*0.003*	10.175	*0.003*
Average		*0.004*		*0.004*		*0.004*

Figure 9: Verification Matrix results demonstrating the validation of the PLS calibration method

■ Sugar Component Analysis of Baby Foods

Commercial baby food samples from three major manufactures were acquired for fructose, glucose, and sucrose analysis. The baby foods selected consisted of pureed fruits and vegetables and fruit juices.

FTIR spectra for each baby food were acquired neat without any pretreatment using the HATR accessory and the spectral acquisition parameters noted previously (figure 10). The fructose, glucose, and sucrose sugar contents were calculated using the calibration established for each sugar from the factor-spaced analysis.

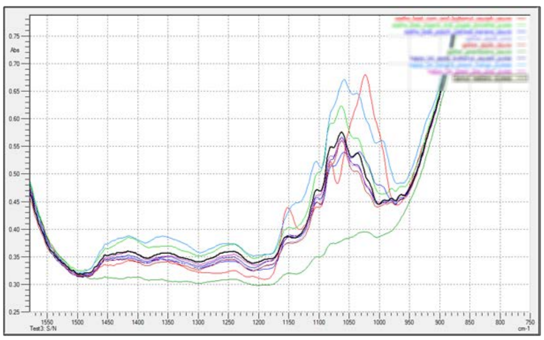

Figure 10: FTIR acquired for the neat commercial baby food samples

Spectrum	Fructose	Resid. 1	Glucose	Resid. 2	Sucrose	Resid. 3
corn_and_butternut_squash_sauce	-3.25	0.05	10.55	0.05	4.52	0.05
organic_fruit_yogart_smoothie_puree	8.80	0.02	4.43	0.02	0.93	0.02
peach_oatmeal_banana_sauce	3.51	0.01	5.08	0.01	2.31	0.01
apple_sauce	7.01	0.01	2.79	0.01	1.45	0.01
greenbeans_sauce	0.68	0.01	2.34	0.01	0.36	0.01
banana_peach_mango_pureee	5.59	0.01	5.78	0.01	5.32	0.01
rasberry_pureee	7.12	0.01	4.29	0.01	0.66	0.01
apple_butternut_squash_puree	7.08	0.01	3.11	0.01	1.45	0.01
banana-peach-coconut_prune	6.90	0.01	7.61	0.01	3.38	0.01
green_pea_pear_puree	7.14	0.01	3.16	0.01	1.17	0.01
apple_juice	6.59	0.01	3.54	0.01	1.42	0.01

Figure 11: Measured sugar concentration results for the commercial baby food samples

Examination of the calculated residuals from the calibration suggested very good fits with the various sugar calibration curves (figure 11). Baby foods that were more fruit based appeared to give better residual values, whereas baby foods that were more vegetable based generally gave higher residuals.

A total sugar concentration for each baby food was calculated from the sugar content provided on the nutrition label of each package. The calculated total sugar value from the nutritional label was compared to the total sugar value calculated from the FTIR quantitative spectral analysis. The measured total sugar values showed a high bias when compared to the reported total sugar values for all samples (figure 12).

Baby Food Analyzed	Measured % Fructose	Measured % Glucose	Measured % Sucrose	Total Measured Sugars (%)	Reported Total Sugar Conc. (%)
corn_and_butternut_squash_sauce	-3.25	10.55	4.52	11.82	2.65
organic_fruit_yogart_smoothie_puree	8.80	4.43	0.93	14.15	10.00
peach_oatmeal_banana_sauce	3.51	5.08	2.31	10.90	4.42
apple_sauce	7.01	2.79	1.45	11.24	9.73
greenbeans_sauce	0.68	2.34	0.36	3.37	2.65
banana_peach_mango_pureee	5.59	5.78	5.32	16.69	9.17
rasberry_pureee	7.12	4.29	0.66	12.06	9.17
apple_butternut_squash_puree	7.08	3.11	1.45	11.64	7.50
banana-peach-coconut_prune	6.90	7.61	3.38	17.88	10.00
green_pea_pear_puree	7.14	3.16	1.17	11.47	7.50
apple_juice	6.59	3.54	1.42	11.55	11.02

Figure 12: Total measured sugar comparison to that listed on the nutritional labels for each commercial baby food sample

■ Conclusion

FTIR Analysis, using a horizontal attenuated total reflection accessory, was demonstrated to be a suitable method to acquire FTIR spectra of commercial baby foods without sample pretreatment or concern for IR water absorption.

Chemometric Partial Least Squares (PLS) routines were used to establish and validate calibration curves for fructose, glucose, and sucrose concentrations in aqueous solutions.

Commercial baby food samples were analyzed for fructose, glucose, and sucrose sugar content. Residual data from the calibration suggested that the baby food samples were within the calibration algorithm's area of analysis. A comparison was made of the total sugars measured to those reported on the nutritional labels of the baby food packages.

This data demonstrates that FTIR analysis of baby foods offers a quick and efficient means of sugar analysis for QA/QC applications.

■ References

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