The *Journal of Cotton Science* 4:137-140 (2000)
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NOTES

Possible NIRS Screening Tool for Entomological Sugars on Raw Cotton

Donald E. Brushwood* and Young J. Han

INTERPRETIVE SUMMARY

In the processing of raw cottons, the presence of entomological sugars from insect contamination on surfaces of lint can affect quality. Sticky sugars and other carbohydrates collect on processing machinery, inhibiting speed and efficiency. Current methods used before processing to identify and screen potentially sticky cottons containing entomological sugars are generally effective, but also time-consuming, costly, and difficult to bring into routine classing systems. A rapid, reliable, and nondestructive test capable of detecting such sugars on cottons would be a welcome tool.

Near infrared (NIR) spectral analysis may offer the potential to identify and quantify such entomological sugars, and this study explores that possibility. Two sugars, unique to whitefly and aphid honeydew, were applied to a non-insect contaminated cotton at different concentrations. Subsequent correlations of actual levels on fiber surfaces and NIR spectral analysis within a fiber moisture range of 50 to 100 g kg^{-1} (5–10%) were studied. It was concluded that a good relationship between NIR spectral analysis and entomological sugar content can be developed if the fiber moisture content is known or can be measured.

ABSTRACT

Entomological sugars from insect contamination of raw cotton (*Gossypium hirsutum* L.) lint not only can affect quality, but these sticky sugars and other carbohydrates also collect on processing machinery, inhibiting its speed and efficiency. In the search for a nondestructive, reliable, and quick test to identify potentially sticky cottons that could be used as a screening tool in a fiber classing system, a single non-insect contaminated cotton was treated with different concentrations of the two honeydew-specific sugars trehalulose and melezitose. High performance liquid chromatography (HPLC) analyses identified and quantified individual carbohydrate concentrations, then near infrared (NIR) spectra scans characterized untreated and treated cottons, that subsequently were conditioned to four fiber moisture levels, ranging from 46 to 93 g kg^{-1} (4.6–9.3%). Statistical analysis of data from chemical analysis and NIR spectra resulted in the selection of the fiber moisture content and 12 wavelengths as independent variables in multiple regression equations to predict concentrations of entomological sugars on these cottons. Calculations for linear correlation coefficients of predictability were able to classify cotton samples with different entomological sugar contents with 89.2% success ratio.

Insect honeydew contamination from aphids (*Acyrthosiphon* spp.) and whiteflies (*Bemisia* spp.) on raw cotton has greatly influenced cotton production worldwide for years (Hector and Hodkinson, 1989). Heavily sticky lint deposits build up on processing machinery and frequent stops are required to remove these materials. Sticky honeydew materials have been isolated and identified as containing, in addition to the normal plant sugars, two sugars identified as unique to aphid and whitefly honeydew: trehalulose and melezitose (Byrne and Miller, 1990). Whitefly-honeydew-contaminated cotton sugar extracts generally contain larger amounts of trehalulose than melezitose (Brushwood, 1998). Aphid honeydew also has melezitose present, and smaller amounts of trehalulose.

Highly positive relationships between certain sugar levels and routine chemical sugar analysis and physical stickiness tests have been established (Brushwood and Perkins, 1993). These tests,

D.E. Brushwood, USDA-ARS-SAA Cotton Quality Res. Stn., P.O. Box 792, Clemson, SC 29633; and Y.J. Han, Dep. of Agric. and Biol. Engineering, Clemson Univ., Clemson, SC. Received 2 June 1999. *Corresponding author (DBrushw@clemson.edu).

although good predictors of stickiness, are difficult to integrate into a rapid classing protocol. A nondestructive, reliable, and quick test to identify potentially sticky cottons would be a valuable screening tool in a fiber classing system. One such test could be online NIR analysis, properly calibrated and corrected for micronaire and moisture differences.

This study was designed to produce a set of cotton samples with known concentrations of entomological sugars. By using NIR as a tool to measure concentrations of these sugars, a possible discriminant analysis procedure could be developed to classify honeydew-contaminated cottons. A potential outcome of this study could be the development of a simple device that would measure entomological sugar levels on cotton by using a limited number of optical filters at selected NIR wavelengths.

MATERIALS AND METHODS

Predetermined amounts of pure trehalulose and melezitose were randomly applied to the surface of the cotton. Triplicate samples were treated with water solutions of three different concentrations and 60/40 mixtures of trehalulose and melezitose. After treatment, these samples and untreated samples were stored for 2 wk at 21°C and 24% relative humidity (47 ± 0.1 g kg^{-1} [4.7 ± 0.01%] fiber moisture). Standard oven moisture contents for these samples were determined to be 47 ± 1 g kg^{-1} (4.7 ± 0.1%) before blending with a rotary laboratory blender and finally by hand. The samples were reconditioned at 24% relative humidity for 5 d before testing. Subsequent conditioning relative humidities for the three samples were 46, 70, and 81%. In each case, a conditioning period of at least 1 wk was allowed before testing.

Individual sugar compositions of treated and untreated cottons (1 g each) were determined by HPLC (six replicates per sample) and moisture contents were determined by a routine oven test. NIR spectrophotometer (Pacific Scientific, Model 6500) scans of each sample from 1100 to 2500 nm were conducted at each conditioning relative humidity. A total of 12 measurements per treatment were conducted.

RESULTS AND DISCUSSION

Individual sugar concentrations

The concentrations of trehalulose on treated cottons averaged 0.8 g kg^{-1} (0.08%), 1.4 g kg^{-1} (0.14%), and 1.7 g kg^{-1} (0.17%) for the low, medium, and high treatments. Melezitose concentrations were determined to be 8 g kg^{-1} (0.08%), 1.5 g kg^{-1} (0.15%), and 1.8 g kg^{-1} (0.18%). In combination, the 60/40 induced mixture cottons yielded trehalulose and melezitose concentrations of 5/4 g kg^{-1} (0.05/0.04%), 1/7 g kg^{-1} (0.10/0.07%), and 120/8 g kg^{-1} (12/0.08%). In all of the above determinations, melezitose retention rates were slightly better than that of the trehalulose.

Moisture Measurements

The average fiber moisture contents for the four relative humidity treatments were: 24% relative humidity, 45.8 ± 0.7 g kg^{-1} (4.58 ± 0.07%) moisture; 46% relative humidity, 66.8 ± 0.7 g kg^{-1} (6.68 ± 0.07%); 70% relative humidity, 84.8 ± 0.6 g kg^{-1} (8.48 ± 0.06%); and 70% relative humidity, 93.3 ± 0.7 g kg^{-1} (9.33 ± 0.07%). Statistical analysis at a $P = 0.01$ confidence level did not indicate any significant difference in moisture content between untreated and treated samples at either level.

NIR Analysis

To predict the entomological sugar contents from the NIR absorbance spectrum, we selected the 12 most significant wavelengths in which the more negative peaks occurred in the second derivative of absorbance plots from the 700 available wavelengths between 1100 and 2500 nm. Second derivative plots of different amounts of entomological sugars at different moisture contents all show similar shapes, and their negative peaks occur within 2 nm of each other. Quantitative differences between sugar levels as determined by HPLC were only a fraction of the differences between moisture levels. Therefore, the moisture content of the sample was used as an independent variable in multiple regression analysis. The SAS procedure RSQUARE (SAS Inc., Cary, NC) was used to determine an optimum regression

Table 1. Values of the best regression equation with varying numbers of independent variables for four moisture content ranges tested.

Variables	Range of moisture content 40–50 g kg^{-1} (4–5%)	60–70 g kg^{-1} (6–7%)	80–90 g kg^{-1} (8–9%)	90–100 g kg^{-1} (9–10%)
No.			R^2	
3	0.3738	0.3655	0.1932	0.3785
5	0.5419	0.5023	0.4414	0.5893
7	0.6039	0.6145	0.4915	0.7181
9	0.6176	0.6413	0.5060	0.7318
11	0.6386	0.6461	0.5125	0.7415
13	0.6437	0.6471	0.5150	0.7422

Table 2. Discriminant classification result for total entomological sugar contents.

Sugar level†	Samples	Classified into High	Low
		no.	
High	76	74	2
Low	44	11	33
Total	120	85	35

† High ≥1 g kg^{-1} (0.1%); Low <1 g kg^{-1} (<0.1%).

Table 3. Discriminant classification result for trehalulose contents.

Sugar level†	Samples	Classified into High	Low
		no.	
High	44	41	3
Low	76	14	62
Total	120	55	65

† High ≥1 g kg^{-1} (0.1%); Low <1 g kg^{-1} (<0.1%).

Table 4. Discriminant classification result for melezitose contents.

Sugar level†	Samples	Classified into High	Low
		no.	
High	28	28	0
Low	92	12	80
Total	120	40	80

† High ≥1 g kg^{-1} (0.1%); Low <1 g kg^{-1} (<0.1%).

equation with a minimum number of wavelengths as independent variables.

Table 1 shows the R^2 values of the best regression equation with varying numbers of independent variables when the samples were divided into four moisture content ranges. The R^2 values did not increase significantly beyond seven independent variables. However, different sets of wavelengths contributed at different moisture levels. Because all 12 wavelengths had to be measured anyway, the 13 variable models were determined as optimum multiple regression equations in this study.

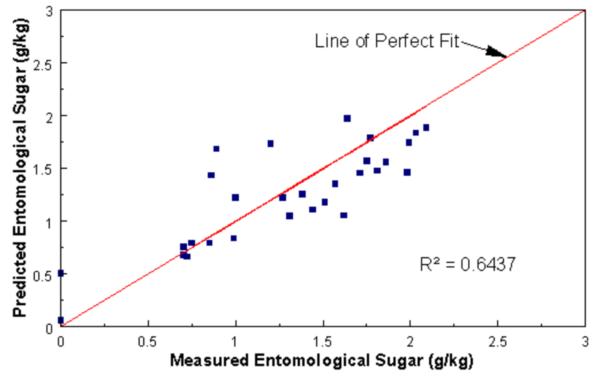

Fig. 1. Measured and predicted entomological sugar contents when moisture contents range between 40 and 50 g kg^{-1} (4 and 5%).

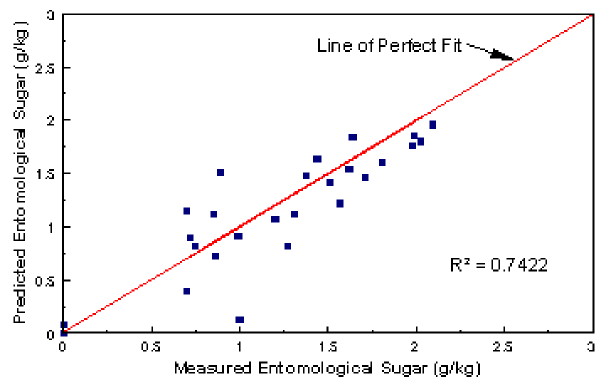

Fig. 2. Measured and predicted entomological sugar contents when moisture contents range between 90 and 100 g kg^{-1} (9 and 10%).

Figures 1 and 2 show the measured and predicted entomological sugar contents by a regression equation for moisture between 40 and 50 g kg^{-1} (4 and 5%) and 90 and 100 g kg^{-1} (9 and 10%) moisture, respectively. Individual sugar contents of trehalulose and melezitose also were predicted from the NIR spectrum and the moisture content. The R^2 values for trehalulose ranged from 0.5557 to 0.7829 and from 0.442 to 0.8038 for melezitose for different moisture content ranges.

When entomological sugar levels were classified into two classes - “high” for ≥1 g kg^{-1} (0.1%) and “low” for anything below that. The model was determined by a measure of generalized squared distance based on within-group covariance matrices. Table 2 shows the number of observations classified into two sugar levels using the this model. Among the 120 observations, 107 samples were correctly classified for the success ratio of 89.2%.

The result was obtained by a single discriminant model for all moisture content ranges. When four different discriminant models were developed and applied, one for each moisture range, all 120 samples were correctly classified for 100% success ratio. The same procedure was applied to classify the levels of trehalulose and melezitose individually. Tables 3 and 4 show the classification results. For trehalulose, 103 samples out of 120 were correctly classified for 85.8% success ratio. For melezitose, 108 samples were correctly identified for 90% success ratio.

SUMMARY AND CONCLUSIONS

We used NIR spectroscopic analysis to study entomological sugars on cotton identified as combinations of trehalulose and melezitose. Batches of non-insect contaminated cotton samples were treated with these two honeydew sugars separately and in 60/40 combinations. Treated cottons conditioned at four different relative humidities were characterized by HPLC for total and individual sugar contents, routine oven moisture content, and NIR spectral analysis.

Our NIR analysis showed that the effect of moisture content was greater than that of sugar levels. Therefore, 12 wavelengths, chosen from a second derivative of the absorbance plot, and the fiber moisture content were used as independent variables in multiple regression analysis to predict the total entomological sugar contents. Poor predictability was obtained when a single equation was applied to all tested moisture ranges.

Predictability increased significantly when the samples were divided into four ranges of moisture content. When analysis of the treated cotton samples was divided into “high” and “low” levels of entomological sugar content, 107 of 120 observations (success ratio of 89.2%) were identified. This model, when applied for each moisture range, correctly classified all 120 samples. The same procedure classified the sugars melezitose and trehalulose at success levels of 90 and 85.8%, respectively. This preliminary NIR analysis work may serve as a model to further refinement and eventual development of a rapid online screening test device to detect the presence and level of insect honeydew on cotton lint.

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