

The use of an automated, high speed analyser of disintegrated sugarcane in a breeding programme

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ABSTRACT

The traditional method of whole cane analysis is both tedious and time consuming. This paper describes the use of FT-NIR based high speed analysis that allows up to 400 samples per day to be handled. This high-throughput system means that complete analysis can be achieved for samples from the very beginning of the variety selection programme. This was demonstrated in Barbados in 2007.

INTRODUCTION

The usual method for analysing whole cane samples for brix, pol and fibre content is labour intensive and time consuming. It requires that whole stick cane be disintegrated in a Jeffco cutter-grinder, a sample of the ground mass weighed and the juice expressed from the sample by hydraulic press. The sample size used at the Cane Breeding Station was 500g. The pressed cake was then oven dried for 48 hours to estimate the fibre content. The Brix of the juice was then measured with a hand held refractometer. About 200ml of the juice was then clarified and filtered before its pol was read with a polarimeter. This method allowed up to 40 samples to be analysed in any working day. The grinder had to be stopped and cleaned in between each sample. The cleaning time also allowed for the grinder to cool down, since it generated considerable heat during the disintegration.

Because of the time consuming nature of this procedure, variety selection programmes were unable to gather full analysis from early stages where large numbers of genotypes are still present in the system. Since fibre content has now become an important characteristic of any new variety, the analysis system was inadequate to provide the necessary accurate data for the selectors.

In 2006 the Cane Breeding Station began to investigate new methods of cane analysis based on NIR spectrometry. After looking at several options it was decided to purchase the Spectracane automated system. This paper describes its use during the 2007 crop season. The Spectracane was developed by collaboration between BSES of Australia (in their breeding department), Biolab Analytical Technologies of New Zealand and Bruker Optics of Germany. It is specifically designed for use in experimental systems where large numbers of small samples need to be analysed as is typical of breeding and selection programmes. The development process is described by Berding *et al* (2004).

The Basic Components of the Spectracane

1. The cane disintegrator is supplied by Dedini (Brazil). It is a D2500 model with a 10HP, 400v motor. It is capable of handling up to 400 samples per day without heating. It delivers the ground cane sample directly into the Spectracane.
2. The display panel and keyboard allows the operator of the disintegrator to control the delivery of samples and to enter identity data for each sample.
3. The delivery conveyor belt of the Spectracane receives the disintegrated sample and delivers it to the “fluffing chamber”.
4. The fluffing chamber contains two rotating spiked rollers that mix the sample to produce a homogenous mass of cane.
5. The presentation conveyor produces a stream of the cane sample about 50mm thick to pass in front of the NIR read head. This stream is compressed and made even by a roller placed above the belt.
6. The FT-NIR Spectrometer is a Bruker Matrix-F model. It is housed in its own compartment which is kept between 25 and 30 degrees Celsius by a small air conditioning unit.
7. The disposal conveyor carries the sample for dumping after it has been read by the spectrometer.
8. All motors, sensors and FT-NIR scans are controlled by an internal programmable logic controller (PLC).
9. A laptop computer collects the spectral data via a wireless network connection to the spectrometer.

Software

1. The automation of the system and supervision of the PLC is controlled by Windows™ based Canecon[®] software. It provides a graphical display of the whole system and shows the progress of each sample through the machine. This display is seen by the operator of the disintegrator and is also shown on the laptop from which the software runs.
2. Canecon[®] inter faces with Bruker OPUS™ software to initiate sample analysis, collect spectral data and name and save the necessary files.

Operation

The cane sample, typically five whole canes, is processed through the grinder until it is all deposited on the delivery conveyor. The operator then presses the “Go” button and the conveyor moves the ground cane forward and tips it into the fluffing chamber where the spiked rollers ensure complete mixing and distribute the mass of cane into a linear sample stream of uniform depth. This is then moved past the spectrometer lamps by the presentation conveyor. As the sample is being analysed the disposal conveyor creeps forward slowly collecting the entire sample. If the analytical results are within the calibration database range the disposal conveyor rapidly moves forward, dumping the sample into a disposal bin. If the analysed spectrum is outside of this known range the belt reverses and places the sample into a collection bin to be sent to the regular laboratory for analysis. These outliers are at a later date used with their

laboratory data to update the calibration database. The system also collects one in fifteen (this number is set by the operators) of the samples for routine check analysis to ensure that the spectrometer remains working within its normal parameters.

Spectral data are collected and analysed for the whole length of the sample as it passes in front of the spectrometer read head. The final output is the result of the average of the many hundreds of spectra read during this process.

At the end of the working day, the machine is washed down with a hose to clean it. All components are stainless steel and electrical parts sealed from water damage. This makes cleaning very quick and easy.

Numbers of Samples Analysed

The Spectracane is advertised as having the capability to handle 400 samples in an eight hour working day with a minimum of two people to operate it. We usually had three people, one to grind the cane, one collecting the saved samples and reading the polarimeter for saved samples and one operating the hydraulic press and collecting juice from the saved samples. We were able to manage between 320 and 360 samples a day for most of the operating period.

A total of 4375 samples were analysed. These comprised all Stage 2, Stage 3 and Stage 4 clones from the Barbados variety selection programme, Stage 1 family samples from the high fibre programme, various small trials with high fibre varieties and about 700 samples from the Cane Breeding Station. The average time per sample during a full day of operation was one minute thirty seconds from grinding to analysis. It is possible to calculate this because each analytic entry contains a time stamp.

Local Calibrations

All data collected from the regular press method (saved samples) were matched with the output from the Spectracane and the spectra from the Matrix-F and sent back to Biolab in New Zealand. There the chemometrics technician updated the calibration database and sent it back to us to replace the old one. In this way the calibrations become fine tuned each season of operation. So far in 2008 we have run 735 samples with thirteen outliers to be analysed.

Table 1 shows a sample of the data output from the Spectracane. Files are saved in Excel readable format.

Table 1: Sample of data output from the Spectracane

Time	Variety¹	% Brix In Juice	% Brix In Cane	Fibre %	Moisture %	Pol Reading	% Pol In Juice	% Pol In Cane	Purity	MD Index²	Spec Outlier³
09:30:22.255 (GMT+4)	DB87138.0	20.7	17.1	17.2	68.5	46.9	10.6	8.8	51.5	26.878119	YES
09:27:32.000 (GMT+4)	WI8253.0	15.3	13.2	13.9	73.0	45.0	10.5	9.0	68.3	0.4954802	NO
09:25:09.957 (GMT+4)	WI82705.0	19.0	16.3	14.3	69.3	69.9	16.0	13.7	84.2	0.314463	NO
09:26:46.026 (GMT+4)	WI8620.0	15.7	12.5	20.3	67.9	45.6	10.6	8.4	67.6	0.5254879	NO
09:28:09.170 (GMT+4)	WI8704.0	17.3	14.4	16.4	69.3	57.0	13.1	11.0	76.2	0.2851562	NO
09:29:36.883 (GMT+4)	WI8816.0	17.5	15.0	14.5	71.0	55.3	12.7	10.9	72.8	0.2492351	NO
09:26:07.915 (GMT+4)	WI88702.0	20.5	17.0	17.1	66.4	72.5	16.5	13.7	80.4	0.1703828	NO
09:28:53.933 (GMT+4)	WI90601.0	17.8	13.9	22.1	64.1	59.3	13.7	10.6	76.7	0.5698333	NO

¹Variety name plus .0 indicating that it was a single reading for this sample; multiple readings of same sample would be .1.2 etc

²Mahanalobis Distance Index - a multivariate distance index to indicate if the readings were within threshold values for known calibrations

³Spectral Outlier YES or NO, indicates whether the readings were accepted as part of the known calibrations based on the MDI

REFERENCES

Nils Berding, David Marston, W. Fred McClure, Maarten van Eerten and Brian Prescott. (2004). FT-NIR spectrometry and automated presentation for high-speed at-line analysis of disintegrated sugarcane. Proc. INCIS 11: 81-87