Assessing and enhancing near infrared calibration robustness for soluble solids content in mandarin fruit

J.A. Guthrie[a] and K.B. Walsh[b]

[a]*Centre for Food Technology, Department of Primary Industries, Rockhampton, Qld 4702, Australia*

[b]*Non-Invasive Assessment Group, Plant Sciences, Central Queensland University, Qld 4702, Australia*

Introduction

Near infrared (NIR) spectroscopy has been applied to the sorting of intact fruit with a high moisture content for constituents such as soluble solids content (SSC) in cantaloupe fruit,[1] sugar content in intact peaches,[2] sugar content, acidity and hardness of intact plum fruit[3] and SCC of intact citrus (mandarin fruit). Commercial application to pack-house fruit sorting lines commenced in Japan in the mid 1990s, for the sorting of sweetness, ripeness and acidity of citrus fruit, apples, pears and peaches at three pieces per second per lane.[5] Commercial application within pack-houses of Western countries is nascent.

The application of NIR technology requires an appreciation of the distribution of the character of interest within the fruit and the absorption and scattering of light through the fruit, in order to design an appropriate optical configuration of light source, detector and fruit (for example References 6 and 7). The robustness of the NIR calibration model must be assessed across populations of fruit differing in, for example, temperature, variety and growing district. Unfortunately, these parameters are not well reported in the literature, with many NIR studies reporting the use of a standard optical design for spectral acquisition and the use of a single harvest population, divided into a calibration set and a validation set. Few studies have explored the issue of validation across populations varying in the locality of harvest, the time of harvest with a given season, or across years. A notable exception is that of Peiris *et al*.[8] who reported calibration validation across three seasons for peaches. A calibration developed in one year predicted poorly on other years, but a combined calibration performed well for validation groups drawn from those years.

In the current study we report on issues related to calibration robustness for intact mandarin fruit assessed for SSC.

Materials and methods

Plant material and SSC analysis

Imperial variety of mandarin were sourced from commercial orchards in Munduberra, Queensland. Fruit were sourced from three separate farms on one day, from three separate harvests over a five-day period from one tree and from one packhouse over three seasons. Fruit were halved, juiced and SSC determined by refractometry (Bellingham and Stanley RMF 320).

Near Infrared Spectroscopy: Proceedings of the 10th International Conference

Spectroscopy

Spectra were collected using an NIR enhanced Zeiss MMS1 spectrometer and a tungsten halogen light in the optical configuration reported by Greensill and Walsh.[7] Spectra were collected from one side of each fruit, on the equator of the fruit, equidistant from pedicel and stylar ends.

Chemometrics

The software package WinISI (ver.1.04a) was used for all chemometric analysis. Calibration performance was assessed in terms of coefficient of determination (R^2) standard error of prediction (*SEP*), variance ratio (1-*VR*), standard deviation ratio (*SDR*), slope and bias of the validation sets. Further, the criteria of Wortel *et al*.,[9] based on the Taguchi concepts as used in process control, were applied to evaluate model robustness. This approach involved calculation of an average *SEP* and a signal-to-noise statistic (s/n = 20 $\log_{10}$ [mean *SEP* / *SD SEP*]) for the performance of a given model across a range of validation sets.

Table 1.Calibration and validation statistics for a calibration on one population of mandarin SSC, used in prediction of three populations varying in (a) days of harvest, (b) location of harvest and (c) season of harvest.

Fruit population	*SD*	R^2	*SECV/SEP*	BIAS
Time				
Cal	0.95	0.90	0.35	
Val				
Day 1	0.73	0.68	0.48	0.173
Day 3	0.72	0.71	0.52	– 0.352
Day 5	0.68	0.55	0.52	0.209
s/n			**26.8**	
Av *SEP*			**0.51**	
Location				
Cal	0.85	0.87	0.353	
Val				
A	0.51	0.55	0.37	– 0.12
B	0.57	0.69	0.41	0.25
C	0.50	0.55	0.52	0.40
s/n			**14.93**	
Av *SEP*			**0.43**	
Seasons				
Cal	0.95	0.84	0.42	
Val				
Year 1	0.96	0.83	0.49	0.24
Year 2	1.05	0.31	2.45	0.40
Year 3	1.05	0.82	3.76	3.73
s/n *SEP*			**2.65**	
Av *SEP*			**2.23**	

Results and discussion

Calibration statistics and B coefficients

Typical MPLS calibration statistics for intact mandarin SSC were: R^2 0.87, *SECV* 0.35, using six principal components, on a population *SD* 0.85, n = 100 (Table 1).

The MPLS B coefficients for the mandarin SSC calibrations contain negative weightings on second derivative spectra around 910 and 850 nm and positive weightings around 880 nm (data not shown). Absorbance at *ca* 910 nm is ascribed to a third overtone stretching of CH bonds (Golic and Walsh, this volume). Absorbance at 880 nm may convey pathlength information. A calibration that does not contain spectroscopically 'relevant' information is likely to be over-fitted to the data and, thus, can be expected to perform poorly when applied to new validation populations.

Calibration validation

A calibration developed from a single population of fruit (100 spectra) was applied to validation sets harvested on different days, different locations and different growing seasons (Table 1). The cause of the decrease in performance of a calibration when applied to a 'new' group presumably reflects change in the physical (optical) properties or the chemical properties (acid, water content) of the fruit. Temperature of the fruit was constant at scanning. Calibration performance across harvest day and location was comparable, as indicated by the mean *SEP* and s/n statistic, while performance was dramatically degraded across seasons. The cause of the dramatic decrease in performance of a calibration when applied to a new season of fruit is not clear and could reflect changes in the instrument used as well as change in the sample (fruit).

To improve calibration performance on a new validation set, a typical strategy involved addition of samples from the new set to the calibration group. The validation sets were divided into two equal groups. One group was retained as a validation set and the other group used for selection of samples for addition to the calibration set. Any validation sample with a GH > 3.0 (calculated on calibration set scores and loadings) was excluded from this process. Several approaches were used in the selection of samples from the validation group for addition to the calibration group, (1) random, (2) selection, on the basis of ascending GH (validation set ordered in ascending order of GH calculated on calibration set scores and loadings, and samples selected at equal GH intervals), (3) selection of the basis of spaced GH (calculated as per 2) and (4) selection on the basis of NH (increasing NH values calculated on calibration set scores and loadings to select increasing numbers of validation set samples, using the ISI 'Expand a Product File with New Spectra' feature). The performance of a calibration developed in one

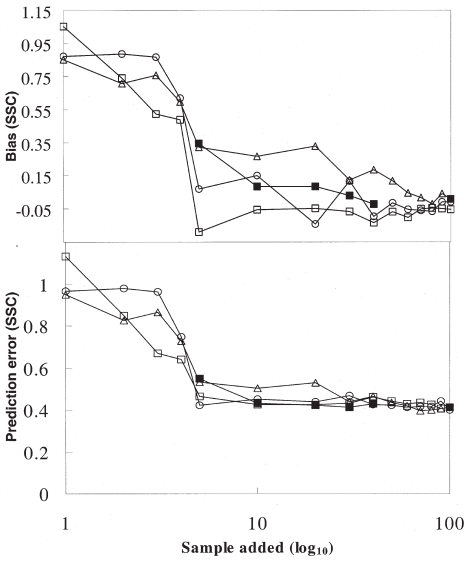

Figure 1. Prediction statistics for SSC of a mandarin validation population (different growing season to calibration population) using three treatments for sample selection from the new season group for addition to the calibration group. Open circle, random selection; open squares, central GH selection; closed squares, spaced GH selection; open triangle, NH selection.

growing season and applied to fruit of a subsequent season was improved in terms of *SEP* and bias as increasing numbers, up to *ca* 10, of 'validation set' samples were added to the calibration set, using any of the three selection approaches (Figure 1). It is surprising that so few fruit were representative of any physical or chemical change in the validation, relative to the calibration, set. In practical terms, we recommend it is sufficient to add data of *ca* 15 fruit to a calibration to update it for use across growing seasons.

Acknowledgements

This work was support by a Citrus Marketing and Development Grant, administered through Horticulture Australia Ltd. Supply of fruit from Gaypak packhouse and Steve Benham of Joey Citrus, Munduberra, is also gratefully acknowledged.

References

1. G. Dull, G. Birth, D. Smittle and R. Leffler, *J. Food Sci.* **54,** 393 (1989).
2. S. Kawano, H. Watanabe and M. Iwamoto, *J. Jpn Soc. Hort. Sci.* **61,** 445 (1991).
3. T. Onda, M. Tsuji and Y. Komiyama, *Journal of the Japanese Society for Food Science and Technology* **41,** 908 (1994).
4. S. Kawano, T. Fujiwara, and M. Iwamoto, *J. Jpn Soc. Hort. Sci.* **62,** 465 (1993).
5. S. Kawano, *NIR news* **5,** 10 (1994).
6. K.B. Walsh, J.A. Guthrie and J. Burney, *Aust. J. Plant Physiol.* **27,** 1175 (2000).
7. C.V. Greensill and K.B. Walsh, *Meas. Sci. Technol*. **11,** 1674 (2000).
8. K.H.S. Peiris, G.G. Dull, R.G. Leffler and S.J. Kays, *J. Am. Soc. Hort. Sci*. **123,** 898 (1998).
9. V.A.L. Wortel, W.G. Hansen and S.C.C. Wiedemann, in *Near Infrared Spectroscopy: Proceedings of 9th International Conference*, Ed by A.M.C. Davies and R. Giangiacomo. NIR Publications, Chichester, West Sussex, UK, p. 267 (2000).