Comparing dry extract spectroscopy by infrared reflection with direct measurements on liquid foods

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Introduction

Near infrared (NIR) technology is mostly used to determine constituents in solid or semi-solid products. Liquid food and other products, with a high content of water (> about 80%) are often considered as problematic. This is mainly because water is a strong absorber in the infrared (IR), including the NIR region. Water will strongly associate to ions, organic monomers and polymers by hydrogen bonds. The hydrogen bonds are relatively weak and, therefore, the light absorbance energy will be very sensitive to varying temperature, ion strength, gelatinisation, swelling, presence of other ions and molecules etc. However, a number of applications, using NIR to determine constituents in high moisture products, are reported.

Liquid products can be analysed directly by using cuvettes, fibre optic probes etc. Numerous successful studies have been reported, using dry extract spectroscopy by infrared reflection (DESIR) on liquid foods, such as fruit juices, beer, milk, wine and meat juices. Typical uses for DESIR are described by the following three steps:

1. apply a small volume (typically 0.5 mL) on a glass-fibre filter
2. dry the filter and
3. measure the NIR spectra.

A review paper of the use of DESIR was recently presented by Thyholt and Isaksson.[1] However, none of the 64 reviewed papers compared the prediction performance of the DESIR results with direct measurements of the liquid food products using, for example, transmittance or transflectance.

The present paper will present the comparison of prediction error results from DESIR and from direct measurements in 1 mm and 10 mm cuvettes of two liquid food product model systems.

Experimental

Sample sets

Two different sample sets, A and B, were designed (Figure 1) and produced. The design, of 61 samples, was chosen to get even distributions of all three analytes, simultaneously, with an added range from 0 to 1.6 weight-% carbohydrate. Sample set A was constructed to give no diffuse light scatter, by using water solutions and set B was designed to give diffuse light scatter, by using orange juice solutions.

The sample sets were made by gravimetrical adding designed aliquots of the carbohydrate stock-solutions. The stock-solutions were 25 weight-% solutions of sucrose (α-D-glucopyranosyl-β-D-fructofuranoside), glucose (α-D-glucopyranose) and fructose [β-D-fructofuranose (BDH Labo-

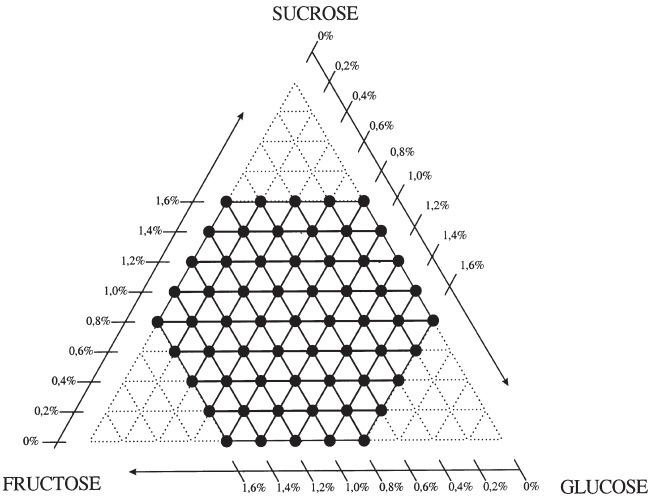

Figure 1. The reduced simplex design of 61 samples of the model systems. The carbohydrate concentration levels represent the actual amount in the water model system and the added amount in the orange juice system.

ratory Supplies, Poole, UK)]. Each sample had the same mass of carbohydrate stock-solutions added, so that the sum of all the three carbohydrate stock-solutions were about 10% of the final samples. The result is that all samples had equal dilution. The orange juice was delivered from a local dealer and mixed to get one stock-juice before mixing the samples. This stock-juice was analysed to contain 4.6% sucrose, 2.3% glucose and 3.2% fructose, by a "high pH anion exchange liquid chromatography" method (at AnalyCen AS, Moss, Norway). The final range in the juice samples were then 4.6–6.2% sucrose, 2.3–3.9% glucose and 3.2–4.8% fructose. All samples were mixed and measured in random order for each data set and for each measurement method (described below). The samples were measured within 48 hours of mixing.

DESIR measurements

Glass fibre filters (GF/A, 47 mm, Whatman) were dried at 70°C for 15 min in thin aluminium cups. Aliquots of 400 µL samples were pipetted on the centre of the filters. The filters were dried at 70°C for 15 min and stored for a minimum of 1 h in an desiccator, before NIR measurements. Each sample was applied on duplicate filters. Reflectance spectra were measured in triplicate (120° rotation between each triplicate) on each filter using a Foss NIRSystems 6500 Scanning Spectrophotometer, from 400 to 2500 nm, in 2 nm steps, at 20°C, using the 32 scan option. The internal ceramic plate was used to measure reference spectra. Average absorbance[(log(1/*R*)] spectra from the duplicate filters and triplicate measurements were used in the further data analyses.

NIR cuvette measurements

The samples were set at a temperature of 28°C for 45 min, before being filled in 1 mm and 10 mm quartz cuvettes (NIRSystems). The cuvettes were placed in the NIRSystems Temperature Controller Module, set to 28°C. Transmittance spectra from duplicate fillings were measured on the Foss NIRSystems 6500 Scanning Spectrophotometer, from 400 to 2500 nm, in 2 nm steps, using the 32

scan option. Reference spectra were measured without the cuvette. Average absorbance [log(1/*T*)] spectra was used in the data analyses.

Data analyses

All the six data sets were first reduced to the spectral NIR range of 780–2500 nm. Because of high absorbances of water at the 1800–2000 nm region, this region was deleted. Water also gave high absorbances from 1380 to 1500 nm. The carbohydrates gave clear bands at 2200–2360 nm. The detectors were a silicon detector up to 1100 nm and a leadsulfide detector above 1100 nm. For these reasons, regression models were estimated on the following four spectral regions; 1) 780–1100 nm, 2) 1100–1300 nm, 3) 1100–1800 nm and 4) 2200–2360 nm.

Partial least squares regresson (PLSR)[2] on centred multiple signal correction (MSC)[2] pre-prosessed data, were performed, using the MATLAB V. 4.2c.1 software package. As validation, full cross-validation was used.

The prediction error is expressed as root mean square error of prediction (*RMSECV*) and defined as:

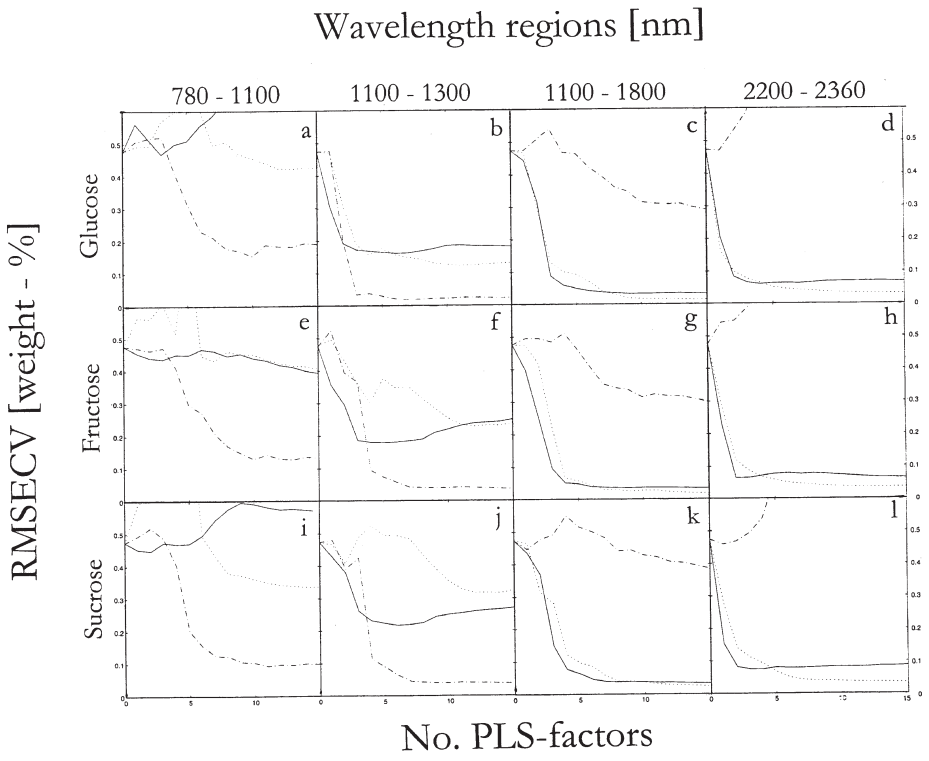

Figure 2. Prediction errors (*RMSECV*) as a function of the number of PLS factors in the calibration models for the water model system. Rows represent prediction of glucose, fructose and sucrose, respectively, and columns represent the different wavelength regions DESIR _____, 1 mm cuvette ..... . and 10 mm cuvette _._._._.

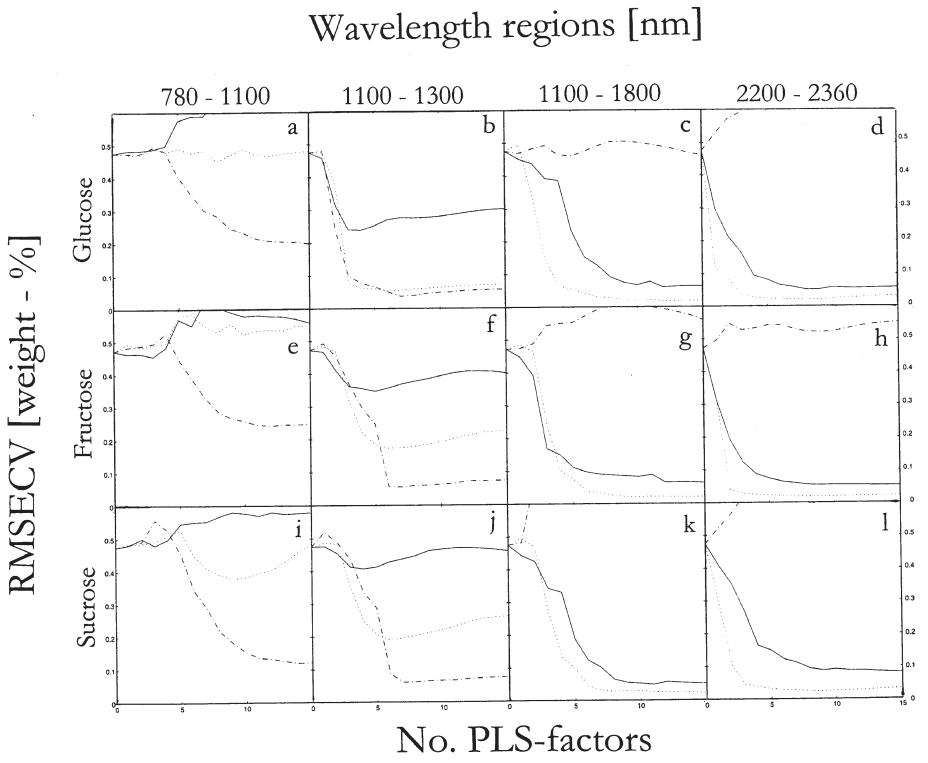

Figure 3. Prediction errors (*RMSECV*) as a function of the number of PLS factors in the calibration models for the orange juice model system. Rows represent prediction of glucose, fructose and sucrose, respectively, and columns represent the different wavelength regions DESIR _____, 1 mm cuvette ..... . and 10 mm cuvette _._._._.

$$RMSECV = \left[I^{-1} \sum_{i=1}^{1} (y_i \quad y_i)^2 \right]^{1/2}$$

where i = [1,2,.....i,.....I] is the number of samples, and y_i represent the predicted added concentration of each carbohydrate and the designed (calculated from the gravimetric measurements) concentration of each carbohydrate, respectively.

Results and discussion

Prediction error results, as a function of the number of PLS factors for glucose, fructose and sucrose for the different wavelength regions, are presented in Figures 2 and 3, for the water and the orange juice model systems, respectively.

Water model system

The absolutely lowest prediction errors of all were obtained by using 1 mm cuvettes in the 1100–1800 nm region, giving the lowest error results of 0.015% (14 factors) for glucose, 0.019% (15 factors) for fructose and 0.025% (15 factors) for sucrose. The use of 1 mm cuvettes gave somewhat higher prediction error results in the 2200–2360 nm region. Data from 10 mm cuvettes gave similar, but somewhat poorer, prediction errors in the 1100–1300 nm wavelengths range, compared to the use of 1 mm cuvettes in the 2200–2360 nm region. Overall, 1 mm cuvettes gave the lowest prediction results, in the 1100–1800 and 2200–2360 nm wavelengths regions. DESIR gave relatively low errors in the 1100–1800 and 2200–2360 nm regions, but overall, not such low prediction errors as the direct cuvette measurements. However, data from 1 mm cuvettes needed more PLS-factors compared with DESIR.

The 780–1100 nm region gave poorer prediction results for all the three constituents, compared with the other wavelength regions studied.

Orange juice model system

The absolutely lowest prediction error results were obtained using 1 mm cuvettes in the 1100–1800 nm region for glucose (*RMSECV* = 0.018%, 12 factors) and sucrose (*RMSECV* = 0.023%, 15 factors) and 1 mm cuvettes in the 2200–2360 nm region for fructose (*RMSECV* = 0.018%, 7 factors). Using 10 mm cuvettes gave relatively good prediction errors in the 1100–1300 nm region. DESIR gave relatively low prediction errors but not as low as when using 1 mm cuvettes in the 1100–1800 nm and 2200–2360 nm regions. In contrast to the results from the water model system, the orange juice system gave smaller calibration models using 1 mm cuvettes compared with DESIR.

As for the water model system, the 780–1100 nm region gave poorer prediction error results compared to the other wavelength regions studied.

Both model systems

Overall, the use of the direct measurements in 1 mm cuvettes gave the lowest prediction error results. One reason may be that the measurements were performed on very carefully temperature-controlled samples. However, the variations in hydrogen bonding was small due to varying temperatures. Other reasons may be that the DESIR measurement procedures involved several steps which could add errors, such as variation between filters, pippeted volumes, drying etc.

The spectra from orange juice, measured in a 1 mm cuvette in the 1100–1800 nm region, gave a total absorbance variation of 1.3 with levels from 0.1 to 1.4 absorbance units, while the DESIR spectra gave a total absorbance variation of 0.016 with levels from –0.01 to 0.015 absorbance units. From theory,[3] the spectra from 1 mm is much closer to the optimal S/N (signal-to-noise) absorbance values of about 0.43 and should contain a higher S/N ratio compared with DESIR.

The eigenvalues, calculated from the singular value theorem, were 10–10,000 times higher for 1 mm cuvette measurements data compared with DESIR. This should indicate that 1 mm cuvette measurements had a higher spectral variability compared with DESIR. The only variation between samples is the designed concentration variation for the 1 mm measurements, while DESIR, in addition, also contain some other variations. This should give higher S/N ratio for the 1 mm cuvette measurements, compared with DESIR and, consequently, lower prediction errors for 1 mm cuvette measurements.

Also, taking into account that DESIR is more time-consuming and needs more skilled labour and laboratory equipment, 1 mm cuvette measurement is recommended over DESIR. This, of course, is only the case if a sufficient amount of liquid is available.

Conclusions

From this study, three main conclusions can be drawn:

1) The lowest prediction errors were obtained using 1 mm cuvettes in the 1100–1800 nm region. Using 1 mm cuvettes clearly out-performed DESIR for both model systems and, in particular, for the orange juice system.

2) The lowest prediction errors were obtained in the 1100–1800 nm region. Compared to the 1100–1800 nm region, the 2200–2360 nm region gave somewhat poorer prediction results for 1 mm cuvettes and DESIR, as did the 1100–1300 nm region using 10 mm cuvettes. The 780–1100 nm region gave the poorest results.

3) The light-scattering orange juice model system gave only very slightly higher prediction errors when using 1 mm cuvettes compared with the non-scattering water model system in the 1100–1800 nm region. In the 2200–2360 nm region the orange juice model system gave lower prediction errors compared with the water model system.

References

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3. H. Willard, L. Merritt, J. Dean and F. Settle (Eds), in *Instrumental Methods of Analysis,* 6th Edition. Wadsworth, p. 73 (1981).