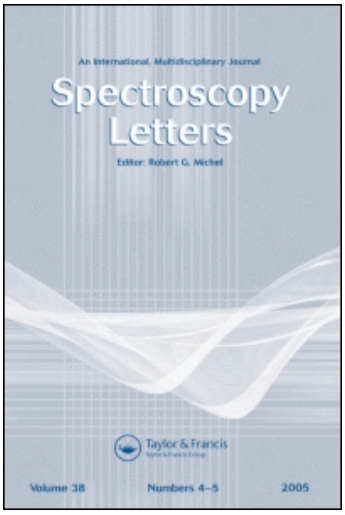

Spectroscopy Letters
Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713597299

Gram-Schmidt Orthogonalization and Elimination of the Effect of Unwanted Component Spectra Applied to a Biological Mid-infrared Spectra Collection

Frédéric Cadet[a]; Bernard Offmann[a]
[a] Laboratoire de Biochimie, Faculté des Sciences, Université de la Réunion, Réunion, France-Dom

To cite this Article Cadet, Frédéric and Offmann, Bernard(1996) 'Gram-Schmidt Orthogonalization and Elimination of the Effect of Unwanted Component Spectra Applied to a Biological Mid-infrared Spectra Collection', Spectroscopy Letters, 29: 5, 901 — 918
To link to this Article: DOI: 10.1080/00387019608001620
URL: http://dx.doi.org/10.1080/00387019608001620

SPECTROSCOPY LETTERS, 29(5), 901-918 (1996)

GRAM-SCHMIDT ORTHOGONALIZATION AND ELIMINATION OF THE EFFECT OF UNWANTED COMPONENT SPECTRA APPLIED TO A BIOLOGICAL MID-INFRARED SPECTRA COLLECTION.

Key words: elimination of interferences, Gram-Schmidt orthogonalization, Mid-FTIR, multivariate analysis.

Frédéric CADET* and Bernard OFFMANN

Laboratoire de Biochimie, Faculté des Sciences. Université de la Réunion, 15 avenue René Cassin. BP 7151, 97715 Saint-Denis Messag Cedex 9, Réunion, France-Dom. Fax: Intl (262) 93 81 66.

ABSTRACT.

Near Infraded spectroscopy (NIR) is the most widely used technique for the analysis of major biochemical constituants in food products. The Mid-infrared range spectroscopy is also being more and more studied in the field of food analysis. This range was primarily applied to qualitative analysis of food components, but with the advent of new techniques such as Attenuated Total Reflectance (ATR) together with the possibility of combination with powerful micro-computers, MIR is now more and more used for quantitative analysis.

In addition to baseline deformations, interference by unwanted compounds are major sources of problems that are encountered in analyses.

* to whom correspondence should be addressed.

We have previousy proposed (Cadet *et al.*, 1991) the use of multidimensional statistical analysis combined with Mid-FTIR spectroscopy for the prediction of sucrose concentrations in biological solutions containing three sugars : sucrose, fructose and glucose.

In this paper, a least-squares method has been used to assess the elimination of the component spectra associated to the fructose (the unwanted components were first orthogonalized and normalized by the Gram-Schmidt orthogonalization method). This procedure permitted the automatic subtraction of discriminant spectral patterns representative of fructose concentration before application of Principal Component Analysis (PCA) and Principal Component Regression (PCR). PCA was applied independently before and after spectral correction of the collections. It is found that when the factorial maps obtained before and after correction are compared, the elimination procedure improves significantly the classification of solutions according to their sucrose content. However the bias and standard deviation (SD) values that are associated with the sucrose content predicted values are not influenced by the correction method used : bias and SD are 3.62×10^{-2} and 3.097×10^{-1} before correction and after correction they were respectively 3.60×10^{-2} and 3.104×10^{-1}. This could be explained by the fact that the presence of fructose in solution does not interfere with caracteristic absorption bands of sucrose and that sucrose and fructose concentrations are strongly correlated. The absorbtions bands of the spectral representation of the principal component, which is identified to be that associated with sucrose, are identical before and after correction.

INTRODUCTION

Spectroscopic methods such as infrared spectroscopy, Raman spectroscopy, Nuclear Magnetic Resonance (NMR) presently, can be easily applied to analysis in food industry on a routine basis.

Near Infraded spectroscopy (NIR) is the most widely used technique for the analysis of major biochemical constituants in food products (Osborne *et al* Fearn, 1986). More and more products and their constituants are being analysed (Osborn., 1981, Williams *et al* Norris., 1987).

In only few cases have Mid-infrared spectroscopy been reported to be used for the analysis of food products since complex spectra are obtained and since incident rays only weakly penetrate the sample that is being analysed. With the recent developments in the field of computer and software engineering and the advent of new techniques such as ATR (Attenuated Total Reflectance) cells, the development and application of physical quantitative analysis methods such as Mid-infrared spectroscopy have considerably expanded (Depecker *et al.*, 1985; Crocombe *et al.*, 1987; Van de Voort and Ismail, 1991; Cadet *et al*, 1991). The interest of using Principal Component Analysis (PCA) for studies in infrared

spectroscopy has already been established (Antoon *et al.*, 1979, Gillete and Koenig, 1982; Bertrand *et al.*, 1984, Cowe and Nocol., 1985). We have previously reported (Cadet *et al.*, 1991) to use multidimensional statistical analysis combined with Mid-FTIR spectroscopy for the prediction of sucrose concentrations in clarified sugar cane juices. We have recently confirmed the interest of using such a method on completely opaque raw sugar cane juices that have not undergone preliminary clarification and filtration processes and thus that contained impurities and fibres (Cadet and Offmann, 1995a). However in the second case, the precision of the analysis can be improved. In addition to baseline deformations (Hruschka, 1987; Cadet and Offmann, 1995b), interference by unwanted compounds can influence the identification, the classification and the quantification of food products.

One way to eliminate irrelevant spectral component is by subtracting the unwanted spectral component according to :

$$Sc = S - k*p$$

where S is the sample spectrum, p is the spectral component of the unwanted phenomenon, k is the subtracting coefficient and Sc is the corrected spectrum. This procedure requires the knowledge of p and the determination of k. It is common for the value of k to be graphically assessed by a skilled operator. Alternative methods principally based on least-squares procedures have been proposed for calculating the coefficient k. (Antoon *et al.*, 1977; Powell *et al.*, 1984; Gillette and Koenig, 1984).

Devaux *et al.* (1988) have proposed to use muldidimensional statistical analysis for the elimination of interference caused by unwanted compounds. A least-squares method has been used to assess the subtracting coefficients k_i, (i=1, 2, ...n), assuming that the undesired component spectra p_i (i=1,2, ...n) are known. With this method, several component spectra could be automatically subtracted .

In this paper, elimination, before PCA and PCR, of discriminant Mid-FTIR spectral patterns associated to fructose in an aqueous mixture containing sucrose, fructose and glucose has been tested. The influence of extracting MIR discriminant spectral patterns without any reference to the chemical values has been observed on the classification and prediction of sucrose concentration of the mixtures.

MATERIAL AND METHODS

Biological samples and fructose solutions.

Sampling of sugar from sugarcane by coring is used. The average of core is about 7000 g. After pulverization, a subsample of approximately 1000 g

is removed. A hydraulic press is used to extract juice from the samples obtained from the coring and from the disintegrator. The subample is pressed for two and a half minutes at 250 bars. The raw juices that are obtained and that contains impurities and fibres are filtered. Via a highly porous plastic filter, this filtration procedure is carried out instantenueously when ATR cells are filled.

Reference sucrose concentration values were determined by corrected polarometric measurements (Cadet *et al.* 1991).

Three pure solutions containing fructose at concentrations that covers the range observed in vivo, 0.5 %, 1 % et 2 % g/ml were used for the calculation of the discriminant spectral pattern

The calibration and verification sets were constituted of 20 and 18 raw sugar cane juices respectively.

Mathematical treatment.

Mathematical treatments were performed on a Compaq personal computer with software written in "C" language and developped in our laboratory. Multidimensional statistical analyses, such as principal component analyses (PCA), describe variation in multidimensional data by few synthetic variables. These synthetic variables are linear combination of all the original variables and have the advantage of having no correlation with each other. Simpler descriptions of data sets are thus obtained with minimal loss of information. These treatments were used for morphological analysis of spectra (le Nouvel., 1981) and for graphical representation of spectra similarity (Devaux *et al.*, 1988a).

PCA was applied to the spectra from 800 to 1250 cm^{-1} (with 235 data points used as principal variables). Spectra were centered prior to PCA according to :

$$X_{ij} = A_{ij} - A_j - A_i + A$$

where X_{ij} = centered data ; A_{ij} = spectral data (log 1/R) of spectrum i and wavelength j; A_j = mean value of spectral data at wavelength j for every spectrum; A_i = mean value of spectral of spectrum i for every wavelength; and A = average mean of all spectral data in the collection.

Principal component regression (PCR) was used to establish a prediction equation. PCR is basically a multilinear regression applied to scores assessed by PCA (Lefebvre., 1983, Lebart *et al.*, 1977). Interest in the introduction of scores according to their predictive ability had already been shown (Dagnelie, 1975, Bertrand *et al.*, 1987).

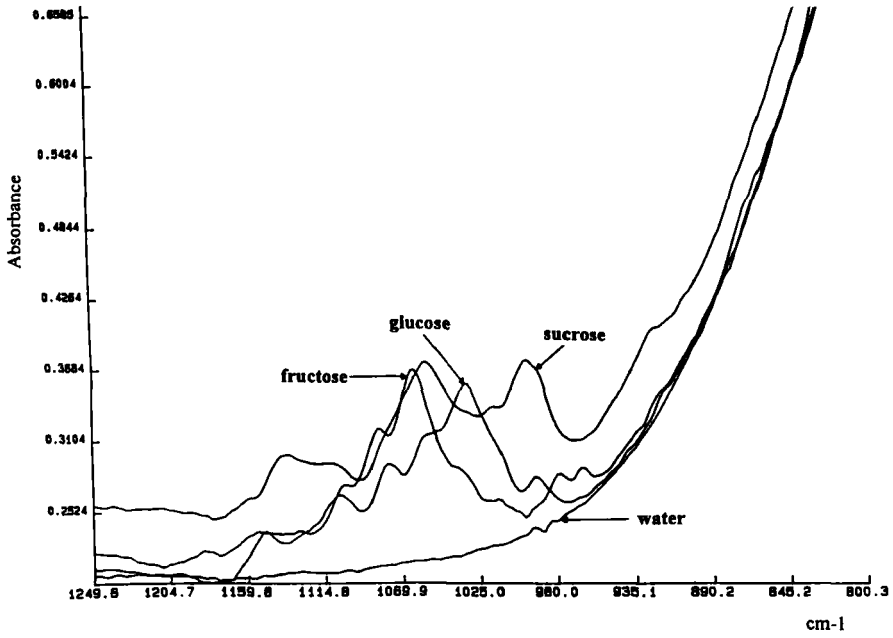

Figure 1. Mid-FTIR spectra of sucrose (15 %), fructose (10 %), glucose (10 %) and water.

Concentrations are predicted according to :

$$C_{n.l} = X_{n.k} \cdot V_{k.p} \cdot R_{p.l}$$

where C is the column vector of predicted concentrations, X is the centered matrix of spectral data, V is the matrix of latent vectors of PCA, and R is the column vector of the regression coefficients of the prediction equations. n, k, p are respectively the number of samples; the number of wavelengths; the number of significant principal components. The dot product V.R is a vector, the components of which may be interpreted in terms of absorption bands. Plotting the components against the corresponding wavelengths gives a spectral pattern. Peaks correspond to absorption bands which are characteristic of the measured chemical constituents. Hollows indicate that when the concentration increases, the corresponding absorption bands will decrease (Bertrand *et al.*, 1988).

Mid-FTIR ATR spectra.

Mid-Fourier Transform Infrared (Mid-FTIR) spectra were collected on a Michelson-100 Fourier transform spectrophotometer. Attenuated total

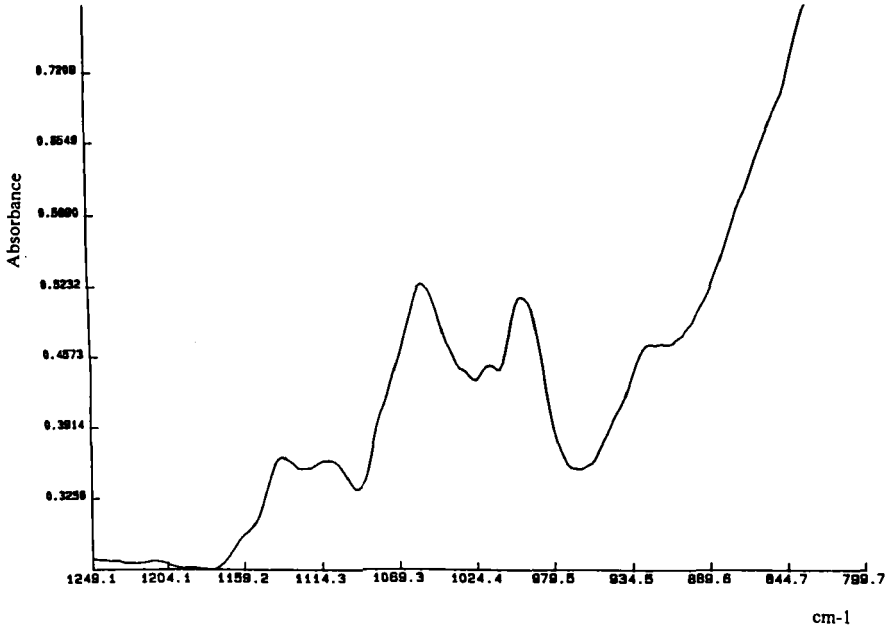

Figure 2a. Mid-FTIR (1250-800 cm-1) spectrum of a sample before correction.

reflectance spectra were obtained with a Specac Overhead ATR system. The crystal of the reflectance element is made from zinc selenide, a material that is quite inert to water; it is quite rapidly cleaned between samples by being sprayed with water and then dried with filter paper.

The data were recorded from 1250 to 800 cm^{-1} in 4 cm^{-1} increments at log(1/R), in which R is the ratio of the reflected intensity for the background to that of the sample. Although the ATR experiment does involve the reflection of the radiation within a crystal, the interaction of the radiation with the sample is the transmittance of radiation through the sample; this depth of penetration is wavelength dependent, but it is passing through a finite layer of the sample. For this reason, plots can read according to absorbance (or transmittance). The combination of four scans resulted in an average spectrum. The intensity the spectra was low; the highest peaks had log(1/R) values less than 0.6 on baseline spectra.

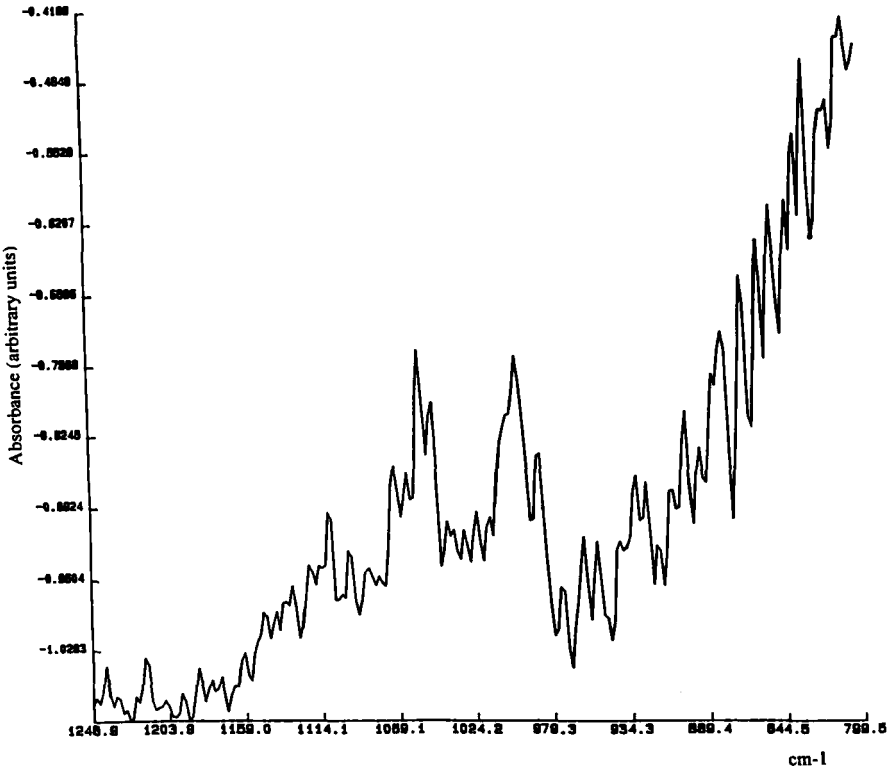

Figure 2b. Mid-FTIR (1250-800 cm-1) spectrum of the sample after elimination of unwanted component spectra assiciated to fructose.

RESULTS AND DISCUSSION

Reference values and spectra.

Sucrose content values for the calibration set ranges between 13.68 and 22.92 g/ml with a mean of 19.53 and a standard deviation (SD) value of 1.93. For the verification set, the sucrose content values range between 14.26 and 22.50 g/ml with a mean of 19.34 and a SD of 1.99.

The 1250-800 cm-1 spectral region is caracteristic of C-O, O-H and C-C saccharidic bands. However, the absorption bands observed in the 1250-800 cm-1 zone is the resultant of the three saccharides present in solution (figure 1).

Elimination of unwanted component spectra

Component spectra have been eliminated by a least-squares procedure according to Devaux et al. (1988b). All programs were written in "C" language in our laboratory and mathematical processing was performed on a Compaq 486 personnal computer.

When there are several component spectra, s can be written :

$$s = s_c + k_1\mathbf{p}_1 + k_2\mathbf{p}_2 + \ldots \qquad (1)$$

If PCA is performed on the collection of the corrected spectra s_C, s_C is decomposed according to:

$$s_c = a_1\mathbf{v}_1 + a_2\mathbf{v}_2 \qquad (2)$$

Where a_1, a_2... are the PC scores of corrected spectra s_C, and $\mathbf{v}_1$, $\mathbf{v}_2$...are the PCA eigenvectors. From Eqs. 1 and 2:

$$s = a_1\mathbf{v}_1 + a_2\mathbf{v}_2 + k_1\mathbf{p}_1 + k_2\mathbf{p}_2 + \ldots (3)$$

s is then split up into two parts : $s=s_C+k_1\mathbf{p}_1+k_2\mathbf{p}_2+\ldots\ldots$, corresponding to the unwanted part, and $a_1\mathbf{v}_1+a_2\mathbf{v}_2$, corresponding to the relevant part. The useful part of the spectum is obtained by vector subtraction of the discarded part from the initial spectrum. The coefficients k_i are in fact the regression coefficients given by the least-square method. However to be consistent with PCA, it is logical to give to $\mathbf{p}_1$, $\mathbf{p}_2$,... the same properties as those of the eigenvectors v_1, v_2,...: the pi vectors must be unit vectors and orthogonal to each other. The subtracting coefficients k_i are calculated similarly to the PC scores a_i, by projection of the spectra on orthogonal vectors. Generally, $\mathbf{p}_i$, $\mathbf{p}_2$,...are not orthogonal and not unit vectors. It is then necessary to orthogonalize them. This is done by the Gram-Schmidt orthogonalization method (Brown *et al.*, 1986; Nyden, 1986). The orthogonalization aids in conditioning the correlation matrix of $\mathbf{p}_i$ vectors for inversion.

The principle of the method is, for two-component spectra:

- a first component spectrum $\mathbf{p}_1$ is normalized according to:

$$\mathbf{g}_1 = \frac{\mathbf{p}_1}{\|\mathbf{p}_1\|} \qquad (4)$$

where $\|\mathbf{p}_1\|$ is the norm of $\mathbf{p}_1$. The projection of the second component $\mathbf{p}_2$ on $\mathbf{g}_1$ is assessed by the scalar product:

$$\mathbf{p}_{12} = \mathbf{p}_2\mathbf{g}_1^T \qquad (5)$$

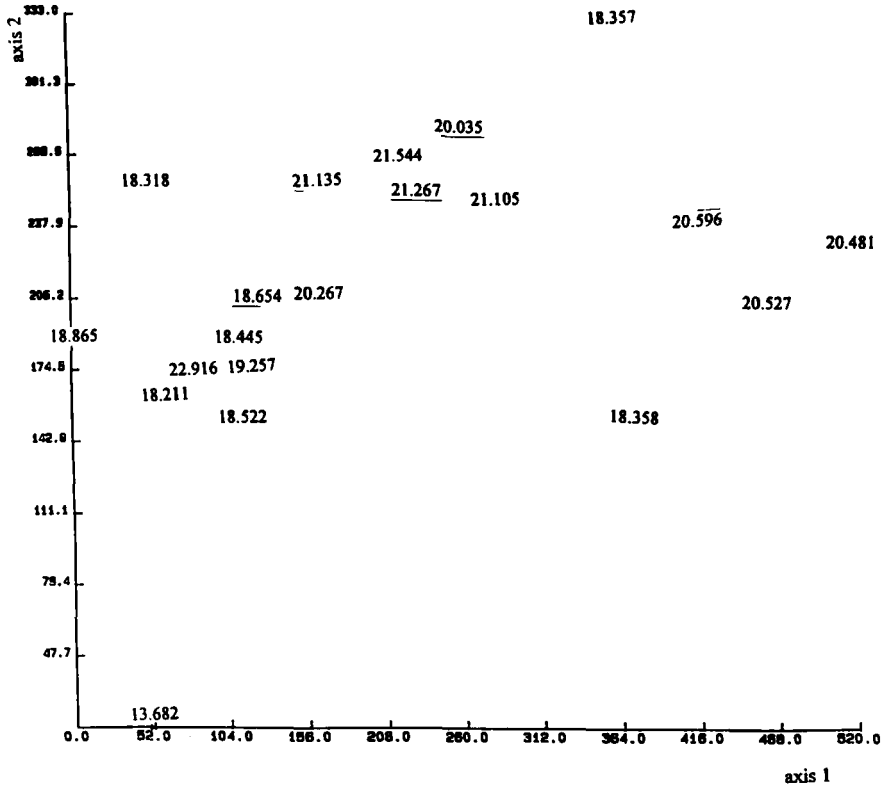

Figure 3. Factorial map as assessed by PCA before elimination of unwanted compounds.

The orthogonal part of **p**2 is given by:

$$\mathbf{p}_2 - \mathbf{p}_{12}\mathbf{g}_1 . \qquad (6)$$

g2 is then calculated by the normalization of **p**2 - **p**12**g**1.

When there are more than two component spectra, the process is iterated : the spectrum **p**3 is orthogonalized withg **g**1, similarly to Eqs. 5 and 6, and the result is orthogonalized with **g**2 in a same manner and finally normalized, giving **g**3. The procedure can be applied with any number of component spectra **p**1.

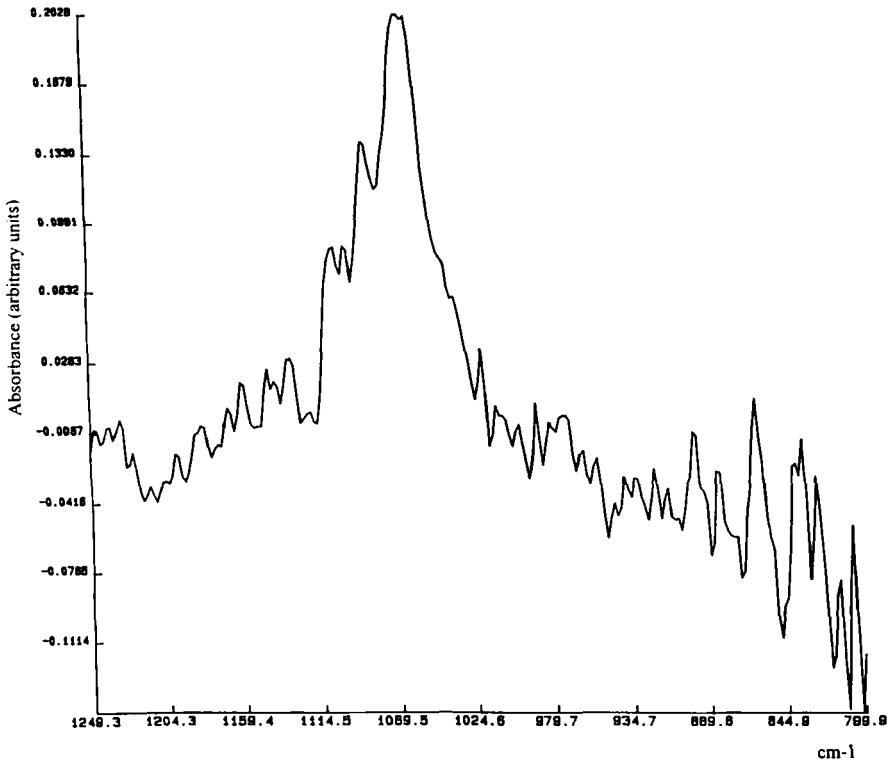

Figure 4a. Spectral representation of axis 1 associated to fructose.

The subtraction coefficients are calculated by orthogonal projection of the spectra on the **g** vectors according to:

$$\mathbf{K}=\mathbf{S}\mathbf{G}^{\mathbf{T}} \qquad (7)$$

where **K** is the n x m matrix of the subtraction coefficients, **S** is the n x v spectral data matrix, $\mathbf{G}^{\mathbf{T}}$ is the v x m transposed matrix of the **g** vectors, m is the number of undesired component spectra, n is the number of spectra, and v is the number of wavelengths.
The corrected spertral data matrix **S**c, is given by:

$$\mathbf{S}_C=\mathbf{S}-\mathbf{K}\mathbf{G} \qquad (8)$$

PCA is then performed on the matrix of the corrected spectra **S**c.

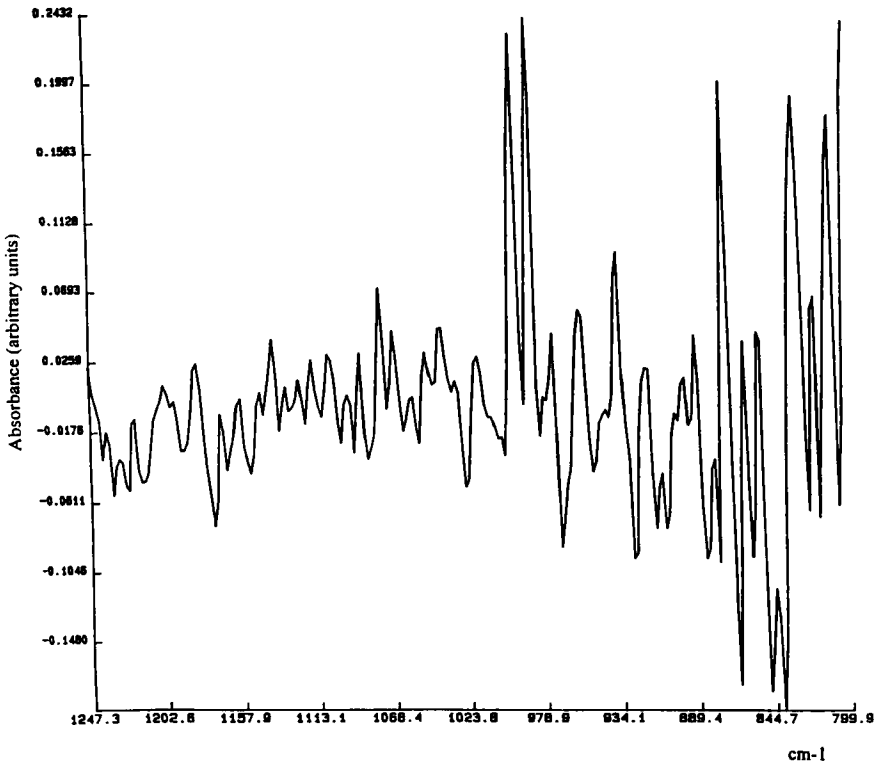

Figure 4b. Spectral representation of axis 2 associated to fructose.

Application

Generally, prediction equations are established by multilinear regression between spectral absorbances and known biochemical values of samples. If this method is precise in the case of pure solutions of macromolecules, in the case of complex mixtures, the precision of this method can be improved. It has been shown that PCA splits up a spectrum into a sum of orthogonal spectral patterns (Le Nouvel, 1981; Cowe and Mc Nichols, 1985; Robert *et al.*, 1987; Bertrand *et al.*, 1985, Renard *et al.*, 1987, Devaux *et al.*, 1988a). Prediction are assessed by a prediction equation established by Principal Component Regression. We have previously shown the interest of using such a method for the prediction of monosaccharides and oligosaccharides contents (Cadet *et al.*, 1991, Cadet and Offmann, 1995b).

orthogonal spectral patterns (Le Nouvel, 1981; Cowe and Mc Nichols, 1985; Robert *et al.*, 1987; Bertrand *et al.*, 1985, Renard *et al.*, 1987, Devaux *et al.*, 1988a). Prediction are assessed by a prediction equation established by Principal Component Regression. We have previously shown the interest of using such a method for the prediction of monosaccharides and oligosaccharides contents (Cadet *et al.*, 1991, Cadet and Offmann, 1995b).

PCA and PCR was applied independently before and after spectral correction of the collections;

PCA and PCR before correction

An example of the spectrum of a sample before correction is given in figure 2a.

Figure 3 shows the factorial maps obtained with the first two axes from PCA. The inertia percentage explained by these two axes are 88.32 % and 8.84 % respectively. The correlation coefficients between these two axes and the sucrose chemical data are 0.955 and 0.019.

PCA and PCR after correction

The spectral patterns associated with fructose are shown in figures 4a and 4b. The two spectral patterns were used as the unwanted component spectra. The peaks that are centered at 1063, 1087 and 1103 cm^{-1} in the first spectrum (figure 4a) are characteristic of fructose. On the other hand it is difficult to find an interpretation of the spectral pattern associated to axis 2 (figure 4b) which probably corresponds to the informations relative to water and spectral deformations.

Figure 2b illustrates an example of a spectrum obtained after elimination.

The factorial maps of the first two axes assessed by PCA are given in figure 5. When the factorial maps obtained before and after elimination are compared, it can be noticed that the elimination procedure improves significantly the classification of the solutions according to their sucrose content. Hence, the positioning of the various mixtures on the factotial map is influenced by the presence of fructose in the solution.

The inertia percentage for the first two axes are 87.46 % and 9.80 % respectively. The correlation coefficients found between these two axes and the corresponding chemical data are 0.949 and 0.022 respectively.

From the data shown in table 1, the reference sucrose content values and the predicted values before and after correction can be compared. Before

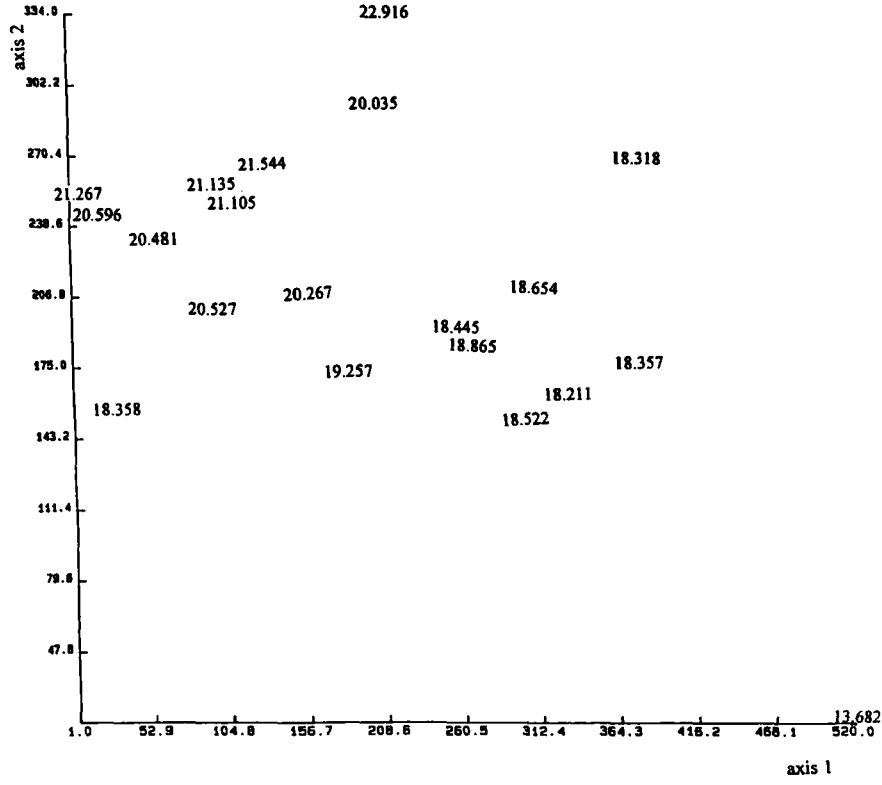

Figure 5. Factorial map as assessed by PCA after elimination of unwanted compounds.

correction, the bias and standard deviation (SD) values are respectively 3.62×10^{-2} and 3.097×10^{-1} and after correction the bias is equal to 3.60×10^{-2} while the SD found is equal to 3.104×10^{-1} after correction.

These results show that the method used, in this case, does not improve the precision of the predicted values. This can be explained by the fact that sucrose and fructose concentrations are strongly correlated and that the fructose molecules in solution do not interfere with the characteristic absorbtion bands of sucrose.

Table I. Reference values and predicted values of sucrose content in the verification set before and after correction

		initial spectra		corrected spectra	
samples number	reference	predicted values	deviation	predicted values	deviation
1	19.56	19.365	-0.195	19.363	-0.197
2	20.421	20.05	-0.371	20.047	-0.374
3	14.26	14.133	-0.127	14.136	-0.124
4	16.926	16.53	-0.396	16.531	-0.395
5	19.534	19.537	0.003	19.544	0.01
6	18.791	18.759	-0.032	18.757	-0.034
7	19.793	20.1	0.307	20.096	0.303
8	21.084	21.038	-0.046	21.032	-0.052
9	21.14	21.158	0.018	21.15	0.01
10	20.22	20.053	-0.167	20.053	-0.167
11	18.693	18.738	0.045	18.739	0.046
12	22.501	22.487	-0.014	22.478	-0.023
13	19.883	20.83	0.947	20.832	0.949
14	20.821	21.267	0.446	21.269	0.448
15	15.852	16.067	0.215	16.072	0.22
16	19.068	18.921	-0.147	18.925	-0.143
17	19.023	19.108	0.085	19.112	0.089
18	20.629	20.71	0.081	20.711	0.082
		Mean	0.0362		0.0360
		Standard Deviation	0.310		0.310

The absorption peaks observed from the spectral representation of the principal component, axis 1, before correction (926, 996, 1016, 1053 and 1135 cm^{-1}) indicates that this spectral pattern corresponds to that of sucrose. This spectral pattern remains unchanged after correction (Figure 6).

Since no shift is observed in the absorption bands of sucrose, this indicates that fructose does not interact with the sucrose molecules in aqueous solutions. The absorbance values of fructose are only added to those of sucrose on a linear model.

Conclusion

In conclusion, unwanted component spectra were eliminated by a least-squares procedure. They were first orthogonalized and normalized by the Gram-Schmidt orthogonalization method. The subtraction coefficient was then assessed, similarly to principal component scores, by projection of the MIR spectra on the orthogonalized component spectra. PCA and PCR

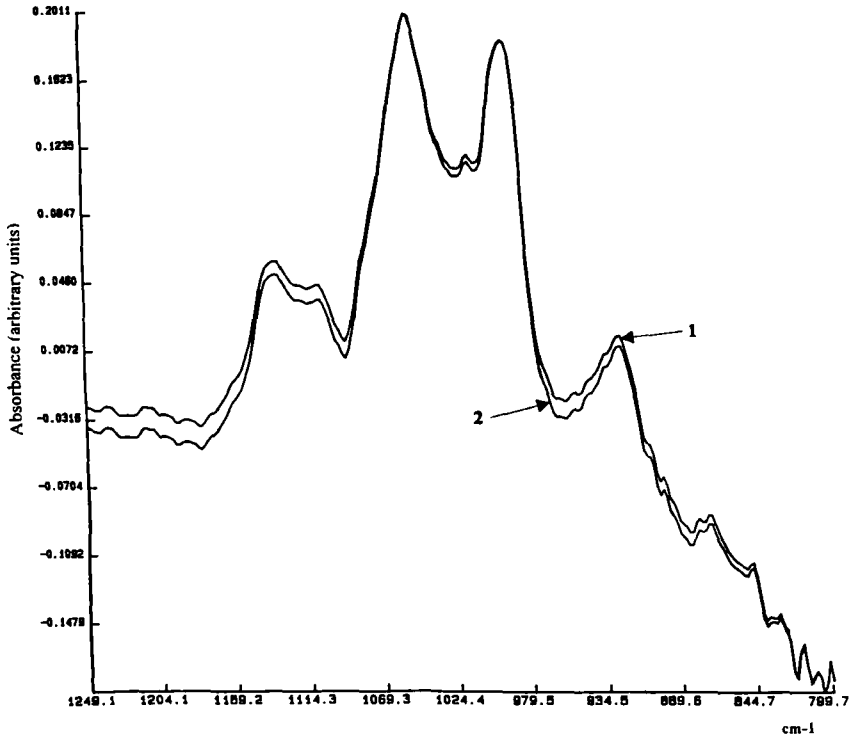

Figure 6. Comparaison between the spectral patterns representative of sucrose before and after correction of the biological samples.

were performed on the corrected spectra. While correction by elimination of interference from unwanted compounds significantly improves classification of samples on factorial maps, such a correction does not improve quantitative predictions. Besides the fact that absorbance values of fructose simply add up to that of sucrose according to a linear model, fructose does not interfere with the absorption bands of sucrose when they are together in an aqueous solution. In order to improve the precision of the predicted quantitative values, other methods need to be developed.

Acknowledgements

This work was supported by a grant from the Ministère de la Recherche et de la Technologie and the Conseil Général de la Réunion.

REFERENCES

- Antoon M.K., Koenig J.H., Koenig J.L., (1977), "Least square curve-fitting of Fourier transform infrared spectra with application to polymer system," *Appl.Spectrosc.*, 31, 512-517.

- Antoon M.K., D'Esposito L., Koenig J.L., (1979), "Factor analysis applied to Fourier transform infrared spectra", *Appl. Spectrosc.*, **33**, 4, 351-357.

- Bertrand D., Robert P., Tran V., (1984), "Traitements mathématiques des spectres NIR de mélanges," 11ème Congrès de L'Association Internationale de Chimie Céréalière, Vienna, Austria, 6th june.

- Bertrand D., Robert P., Loisel W., (1985), "Identification of some wheat varieties by near infrared reflectance spectroscopy," *J. Sci. Food Agric.*, **36**, 1120-1124.

- Bertrand D., Lila M., Furtoss V., Robert P., and Downey G., (1987), "Application of principal component analysis to the prediction of lucerne forage protein content and in vitro dry matter digestibility by NIR spectroscopy," *J. Sci. Food Agric.*, **41**,299-307.

- Bertrand D., Robert P., Devaux M.F., Abecassis J., (1988), "Assignment of Near Infrared Absorption Bands by Muldimensional Analyses of Spectral Data,' in Analytical Applications of Spectroscopy," C.S. Creaser and A.M.C Davies, Eds (Royal Society of Chemistry, London), pp 450-455.

- Brown C.W., Obremski R.J., Anderson P., (1986), "Infrared quantitative analysis in the Fourier domain : processing vector representation," *Appl. Spectrosc.* **40**, 734-739.

- Cadet F., Bertrand D., Robert P., Maillot J., Dieudonné J.and Rouch C., (1991) "Quantitative determination of sugar cane sucrose by multidimensional statistical analysis of their Mid-Infrared attenuated total reflectance spectra," *Appli. Spectros.*, **45**, 2, 166-172.

- Cadet F. and Offmann B. (1995a) "Direct spectroscopic sucrose determination of raw sugarcane juices", *J. Agric. Food Chem.*, Submitted for publication.

- Cadet F., Offman B., (1995b), "Extraction of characteristic bands of sugars by multidimensional analysis of their infrared spectra", *Spectros. Lett.*, **29**, 3,.27-43. Under press.

- Cowe I.A. and Mc Nichol J.W., (1985), "The use of principal component in the analysis of near infrared spectra," *Appli. Spectrosc.*, **39**, 2, 257-262.

- Crocombe R.A., Olson N.L., Hills S.L., (1987), "Quantitative fourier transform infrared methods for real complex samples.", American Society for Testing and Materials, 95-130.

- Dagnelie P., (1975), "Analyse Statistique à Plusieurs Variables," Les Presses Agronomiques de Gembloux, Belgium, 185-190.

- Depecker C., Legrand P., Merlin J.C., Sombret B., (1985), "Contribution de la reflexion diffuse à l'étude de composés biologiques", *Spectrosc. Biol. Mol.*, 69-75.

- Devaux M.F., Bertrand D., Robert P., Qannari M., (1988a) "Application of multidimensional analyses to the extration of discriminant spectral patterns from Near Spectra," *Appl. Spectrosc.*, **42**, 6, 1015-1019.

- Devaux M.F., Bertrand D., Robert P., Quannari M., (1988b) "Application of principal component analysis on NIR spectral collection after elimination of interferences by a least squares procedure," *Appl. Spectrosc.*, **42**, 6, 1020- 1023.

- Gillette P.C., Koenig J.L., (1982), "Noise reduction via factor analysis in FT-IR spectra," *Appl. Spectrosc.*, **36**, 5, 535-539.

- Gillette P.C. and Koenig J.L., (1984), "Objective criteria for absorbance subtraction," *Appl. Spectrosc.*, **38**, 3, 334-339.

- Hruschka W.R., (1987), "Data analysis : wavelength selection methods." In: "Near Infrared Technology in the agricultural and food industry," Eds Williams P.C. and Norris K.H., AACC, Saint Paul, MN (USA), 35-46.

- Lebart L. , Morineau A., and Tabard N., (1977), "Techniques de la Description Stastitique" (Dunod, Paris), 7-46, .

- Lefebvre J. , (1983), "Introduction aux Analyses Statistiques Multidimensionnelles," (Masson, Paris), 3rd ed., 137-148.

- Le Nouvel J., (1981), "Etude d'une famille de courbes par des méthodes d'analyse des données. Application à l'analyse morphologique de courbes provenant de données médicales," These de 3ème cycle, Université de Rennes I, France.

- Nyden M.R., (1986), "Computer assisted spectroscopic analysis using orthonormalized reference spectra. Part I : application to mixtures", *Appl. Spectrosc.*, **40**, 6, 868-872.

- Osborne B.G. (1981), "Principles and practice of near infrared (NIR) reflectance analysis", *J. Food Techn.*, **16**, 13-19.

- Osborne B.G. and Fearn T, (1986), "Near Infrared spectroscopy in food analysis," Longman Scientific and Technical, Wiley and Sons, NewYork.

- Powell J.R, Wasacz F.M., Jacobsen R.S., (1984), "An algorithm of the reproductible spectral subtraction of water from FT-IR spectra of proteins in dilute solutions and adsorbed monolayers," *Appl. Spectrosc.*, **40**, 3,339-345.

- Renard C., Robert P., Bertrand D., Devaux M.F., and Abecassis J., (1987), "Qualitative characterisation of the purity of milled durum wheat products by multidimensional statistical analysis of thier Mid-Infrared diffuse reflectance spectra," *Cereal Chem.*, **64**, 3, 177-181.

- Robert P., Bertrand D., Devaux M.F., and Grappin R., (1987), "Multivariate analysis applied to near infrared spectra of milk," *Anal. Chem.*, **59**, 17, 2187-2191.

- Van de voort F.R, Ismail A. A, (1991), "A rapid FTIR quality control method for fat and moisture determination in butter," *Trends Food Sci. Technol.*, 13-17.

- Williams P, Norris K., (1987), " Near infrared technology in the agricultural and food industry," American Association of Cereal Chemists Ed, Saint Paul, MN, USA, 330 p.

RECEIVED: January 8, 1996
ACCEPTED: February 15, 1996