Paper C1

Infrared spectroscopy (FTIR and NIR) possibilities to determine the tanning degree of the vegetable leather

Trini Canals, Josep Maria Morera, Felip Combalía and Esther Bartolí
Escola d'Enginyeria Igualada (EEI), Universitat Politècnica de Catalunya (UPC), Plaça del Rei 15, Igualada, España, Teléfono: 34-93-8035300, Fax: 34-93-8031589, e-mail: trini.canals@eei.upc.edu

1. Introduction

Physical tests, chemical analyses, and fastness tests serve the purpose of assessing the quality of leather. The IULTCS (International Union of Leather Technologists and Chemists) recognizes a number of methods of chemical analysis as official for leather assessment. These methods have the abbreviations IUF, IUC and IUP followed by a number and many of them have their equivalent in the ISO and EN Standards. Some of these methods determine the amount of metal (chromium, aluminium, etc) present in the leather. In vegetable-tanned leather, the equivalent to the amount of metal in leather is called tanning degree (TD), or also the combined tannin. It is known that the amount of tannin present in the leather affects some of its properties[1], which justifies our interest in such methods of analysis.

The traditional analyses to determine the combined tannins and subsequently calculate the tanning rate are very time-consuming and require considerable experience to get replicable[2] results. The combined tannins, expressed on the weight of the dried and degreased leather, are calculated by subtracting from 100 the sum of hide substance, inorganic non-washable ash and total washable materials (organic and inorganic). This is a process that involves several independent experiments. However, it has several drawbacks, as is quite slow and involves the use of pollutants. Also, the reproducibility of the results depends on the experience of the analysts performing the experiments. The results obtained from traditional methods of analysis are rather empirical.

In recent years, analytical techniques have advanced greatly. The synergy of different spectroscopic techniques with the powerful ability to perform mathematical calculations thanks to advances in computer technology have enabled new methods of analysis that are faster, more sensitive and cost-effective. These analyses enable to determine more parameters simultaneously and a great deal of samples[3].

Several research teams have studied the possibilities of these analytical techniques to determine the tannins present in different substances. Donkin[4] determined the raw material used to make vegetable extracts by means of near infrared spectroscopy (NIR). Cuadros and al.[5] began to study the application of NIR in the determination of tannin and non-tannin in vegetable extracts with good results. In this line, Gutterres[6] performed a study on the diffusion and degree of fixation of tannins in vegetable tanning by NIR. Nakagawa[7] studied the characterization and the molecular weight of vegetable tannins by using various spectroscopic techniques. Ozgunay and al.[8] investigated the molecular structure of valonea tannin by means of mid-infrared spectroscopy with Fourier Transform (FTIR).

In this current paper we intend to move forward in the research and study the possibility of determining directly the tanning degree of vegetable-tanned leather, without having to manipulate or destroy the sample. To this aim we have used the FTIR technique with a module of Attenuated Total Reflection Mode (ATR) and the NIR technique with a fibre-optic probe to record the spectra of the vegetable-tanned leather samples. We have calculated the parameters

with the help of multivariate calculation methods[9-10]. These test systems are suitable for systematic control of different batches of production due to the fact that the sample is never destroyed. In addition, these systems are fast and cost-effective, as they do not require expensive instrumental techniques. These methods can also be very useful for monitoring on-site the development of a vegetable-tanning process, especially in the penetration and fixation stages. By cutting a sample of leather, drying it, splitting it and analysing every layer the precise condition of the tanning can be monitored almost instantly.

2. Material and methods

2.1. Apparatus and software

The tests were carried out using 2.2 m-high (i.e. diameter) and 2.2 m-wide wood drums, Olcina brand.

FTIR spectra were recorded by on an IR Spectrum One (S/N 57458) spectrophotometer from PerkinElmer (Beaconsfield, UK) equipped with an ATR internal reflectance module (Universal Sampling Accessory, S/N P0DL01101418).

Near infrared spectra were recorded on a Foss NIRSystems 5000 instrument (Silver Spring, MD) equipped with a reflectance detector and an Ortiprobe fibre-optic probe.

Data were processed using PLS1 and PLS2 algorithm in Unscrambler v.7.5, from CAMO (Trondheim, Norway).

2.2. Reagents

Quebracho extract Ato Unitan. 72% of tannins. pH (6.9 ºBé) = 4.3-4.8.
Mimosa extract Clarotan. 68% of tannins. pH (6.9 ºBé) = 4.0-4.5.
Sulphated oil. 75% active matter.
Raw oil. 100% active matter.
Sulphited oil. 70% active matter.
HCOOH (85%) P.A.

2.3. Hide processing

The analyses were performed on bovine hides vegetable tanned with the formulation shown in Table 1.

2.4. Leather samples

A total of 67 samples of vegetable leather tanned with different amounts of vegetable extract (mixed with the same amount of mimosa and quebracho) were prepared, to have a set with adequate variability in the tanning degree values. A sample of each leather was used to determine the tanning degree using the official method. Another sample from the same leather was used to record FTIR and NIR spectra.

2.5. Reference methods

All bovine leather samples were analyzed in triplicate using the official methods recognized by the IULTCS to compare results with those obtained using spectroscopic analysis. The chemical analyses and physical tests carried out, together with the methods followed, are detailed in Table 2. From the results of these analyses we calculated the values of the leather substance, combined tannins and tanning degree. These amounts are expressed in relation to dried and degreased leather weight and were calculated according to the formulas show in Table 2.

Table 1		
Formulation		
(on pre-tanned weight):		
Tanning	100% H_2O	
	20% Mimosa + 20% Quebracho	Mixed at 50% in three doses at 30 min. intervals
		r – 12 h.
		Check through tannage
		Overnight rest
Fatliquoring	100% H_2O	
	3% Sulphated oil	
	0.5% Raw oil	
	1.5% Sulphited oil	T = 40ºC
		r – 45 min.
	0.5% HCOOH (1:10)	r – 30 min.
		1 day rest
Toggling		

Table 2
Official methods
IUC 4. Determination of matter soluble in dichloromethane and free fatty acid content
IUC 5. Determination of volatile matter
IUC 6. Determination of water soluble matter, water soluble inorganic matter and water soluble organic matter
IUC 7. Determination of sulphated total ash and sulphated water insoluble ash
IUC 10. Determination of nitrogen and hide substance
Leather substance (%)=100 – Water soluble matter – Sulphated total ash
Combined tannins (%)=100 – Water soluble matter – Sulphated total ash – Hide substance
Tanning degree (%)=(Combined tannins / Hide substance) * 100

2.6. Recording of FTIR and NIR spectra

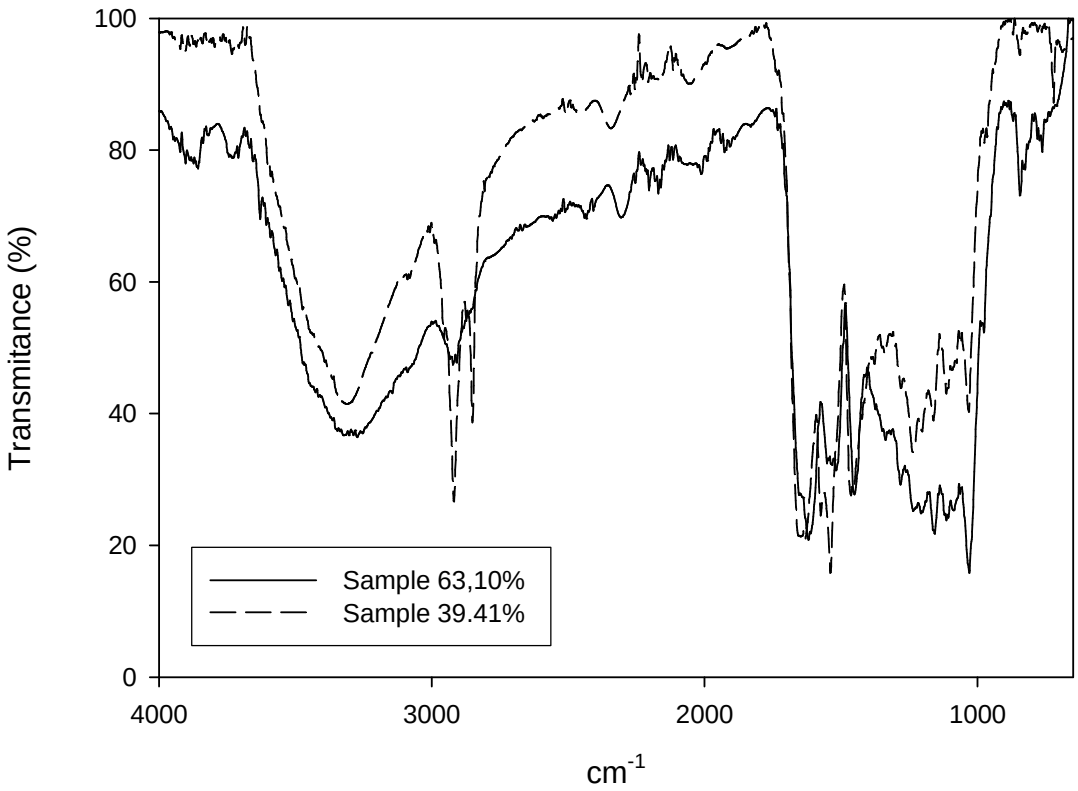
Spectral information about 67 leather samples was obtained by using the FTIR and 75 leather samples by using the NIR techniques, which feature disparate surface penetration abilities. FTIR spectra were recorded in the transmittance mode over the wavenumber range of 4000-650 cm^{-1}, using ATR module and averaging four scans. NIR reflectance spectra were recorded in the absorbance mode with a fibre-optic probe over the wavelength range of 1100-2500 nm, averaging 32 scans per spectrum.

To obtain representative spectra for each sample the possible variability of the distribution of these vegetable extracts has been taken into consideration. Therefore, measurements on each side of the leather sample (grain and flesh) were performed. When assessing the degree of penetration and fixation of vegetable extracts, spectra of the leather samples were recorded at

different horizontal slices. Both FTIR and NIR spectra were acquired at room temperature (25±1ºC).

Obtaining the spectrum takes a very short time (seconds), which emphasizes the overall simplicity and the high speed of the analysis. Besides, the concentration of an individual analyte or of several analytes simultaneously can be obtained from two spectra of each of the samples. Figures 1 and 2 show the FTIR and NIR spectra directly recorded on the surface of two leather simples. Spectral differences between them, with calculated tanning degree values of 39.40 and 63.10 respectively, can be observed.

Fig 1. FTIR spectra for samples 3a11 and 21a2 as recorded directly on the leather surface



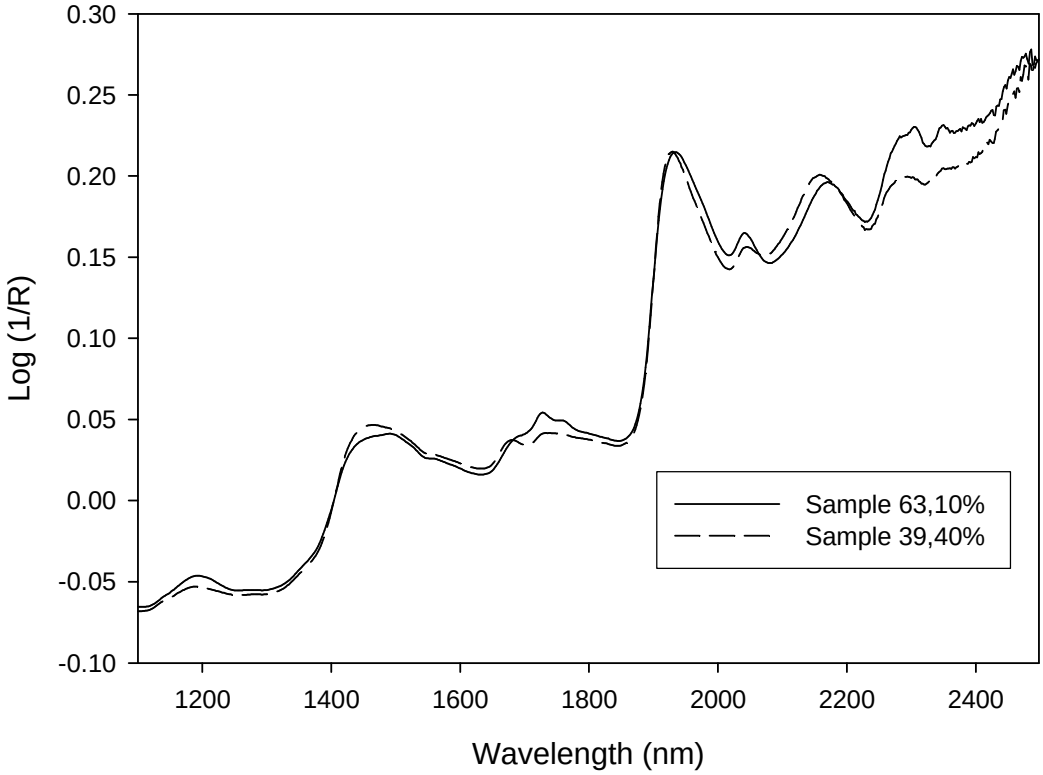
2.7. Processing of data

In this work, we studied leather samples after tanning process. The determination of tanning degree, combined tannins, hide substance and leather substance, was based on the use of partial least-squares regression (PLSR) for calibration. With the aim of developing a robust analytical method to determine, simultaneously and without destroying the sample, the four parameters indicated, the predictive ability of final model PLS2 with individual PLS1 models for each property was compared.

The values provided by the official methods were used as references. Spectra were recorded over the range 1100-2500 nm in NIR and over the ranger 4000-650 cm^{-1}in FTIR. Both absorbance, first-derivative and second-derivative data were examined. Each data matrix was split into a calibration set and external prediction set. The data used were centred and unscaled. Models were constructed by cross-validation, using many segments as samples were included in the calibration set to calculate the mean square error of cross validation (MSECV). The number of PLS factors used in each calibration was chosen in terms of the variation of MSECV in the

region around the minimum in order to avoid overfiting. Then, the tanning degree and the others parameters provided by the PLSR method, were plotted against those obtained with the official method, and data points fitted to a straight line. Once the regression line was obtained, the slope and the intercept test were used to check for the absence of statistically significant differences between the responses of both methods as a significance level $\rho < 0.05$.

Fig. 2. NIR spectra for samples 3a11 and 21a2 as recordes direrectly on the leather surface



3. Results and discussion

Table 3 shows the results obtained by the official methods.

TABLE 3

Reference methods: Analysis results

Number of samples = 67

Parameter	Average	Range
Hide substance	**60.82**	**56.78-66.23**
Leather substance	**92.11**	**88.3-95.80**
Combined tannins	**31.32**	**24.37-34.89**
Tanning Degree	**51.01**	**39.40-63.10**

The vegetable tanned leather simples were registered by the spectroscopic techniques studied (FTIR and NIR). Appropriately processing the spectral information obtained from them with a view to their accurate quantification required the use of effective chemometric method.

We tested carefully various types of calibration models and number of PLS components, wavelength ranges, spectral modes and choices of samples for quantifying external vegetable samples in the prediction set. The calibration models used were chosen in terms of predictive ability as reflected in their calibration and prediction errors, % RSE.

3.1. Quantification of vegetable leather samples from FTIR spectra
The set of available samples is divided into two parts, the calibration set, with 49 samples, and the prediction set, with 18 samples.

Table 4 shows the values of relative mean square error of calibration (RSEPC) and prediction (RSEPP) obtained in a PLS1 model calculated in absorbance mode with 7 principal components (PC) for the different analytes, which were lower than the suggested by Unscrambler software as optimal to ensure the absence of overfiting in the models. Table 4 also shows the results obtained in a PLS2 model in first-derivative mode and with 9 PC. PLS1 presented the best predictive abilities for IC parameter, but PLS2 needs only one model calculated and give quite the same results.

Despite the complex nature of leather as a substrate, the multivariate treatment applied to the FTIR spectra provided good results in all cases. The difference between the results obtained by spectroscopic techniques and those obtained by official method are acceptable for the tanning industry.

Table 4

RSEPC and RSEPP values

	RSEPC (%)		RSEPP (%)	
Parameter	**PLS1 (7PC)**	**PLS2 (9PC)**	**PLS1**	**PLS2**
Hide Substance	0.93	1.50	2.19	1.84
Leather Substance	1.74	2.11	3.63	3.23
Combined tannins	2.24	2.26	6.58	6.78
Tanning Degree	3.83	3.54	6.95	8.83

3.2. Quantification of vegetable leather samples from NIR spectra
In this study, the horizontal splits of some samples were included in the sample set. The set of available samples is divided into two parts, the calibration set, with 57 samples, and the prediction set, with 18 samples.

Table 5 shows the values of relative mean square error of calibration (RSEPC) and prediction (RSEPP) obtained in a PLS1 model calculated in first-derivative mode with 7 principal components (PC) for the different analytes, which were lower than the suggested by Unscrambler software as optimal to ensure the absence of overfiting in the models. Table 5 also shows the results obtained in a PLS2 model in first-derivative mode and with 9 PC.

NIR light penetrate more deeply into the leather than does MIR light, thus the quantification errors obtained from PLS treatment applied to the NIR spectra are quite better. The tanning degree spectroscopic analyses of the splited leather samples demostrated the homogeneous distribution of tannins in the full thickness leather

Table 5

RSEPC and RSEPP values

Parameter	RSEPC (%)		RSEPP (%)	
	PLS1 (7PC)	PLS2 (9PC)	PLS1	PLS2
Hide Substance	0.96	1.39	1.55	1.60
Leather Substance	1.49	2.09	2.69	2.31
Combined tannins	3.21	2.74	5.38	4.55
Tanning Degree	4.18	2.60	7.31	6.77

4. Conclusions

Tanning industry requires fast analytical control methods without the use of polluting reagents. In this presentation, infrared spectroscopy (FTIR and NIR) in conjunction with chemometric processing of the information possibility the determination simultaneously of different analytes (Tanning degree, combined tannins, hide substance and leather substance) directly on samples of vegetable-tanned leather with mimosa and quebracho extracts. Both, FTIR with ATR cuvette and NIR with fibre-optic probe allow register the spectrum of the samples in few seconds.

An appropriate model of PLS2 and the PLS1 models calculated for each analyte yielded good results in the samples studied in the prediction. Quantification errors were of an order of magnitude accepted in the tanning industry. It can therefore be stated that there are no significant differences in values obtained through both calculation methods.

The possibility of a fast quantification of analytes from a large number of samples, compared to reference methods that require much more time-consuming and very often empirical procedures, leads us to conclude that the proposed method is an advantageous alternative to the official method. This is specially the case regarding the systematic control of the evenness of production batches and the on-site control of the development (penetration and fixation) of vegetable tanning.

5. References

1. Fundamentals of leather manufacturing, 1st ed., E. Heidemann, Eduard Roether KG, pp. 290-293, 1993.
2. Análisis y ensayos en la industria del curtido, 1st ed., J. Font, EUETII-ESAI, pp. 21-111, 2002.
3. Estadística y quimiometría para química analítica, 4th ed., Miller J. and Miller J., Prentice Hall DL, p. 239, 2002.
4. Donkin, MJ. and Pierce J.; Tannin analysis by near infrared spectroscopy, *JSLTC* **79,** 8-11, 1995.
5. Cuadros, R., Lalueza, J., Morera JM. and Rius, A.; Determinación simultánea de taninos y no taninos en extractos vegetales mediante espectroscopía infrarrojo cercano (NIR) y calibración multivariante, *AQEIC* **53,** 74-77, 2002.
6. Gutterres, M.; Analysis of vegetable tannin absorption during the tannage of hide/hide power, *JALCA* **102**, 216-221, 2007
7. Nakagawa, K. and Sugita, M.; Spectroscopic characterisation and molecular weight of vegetable tannins, *JSLTC* **83**, 261-264, 2000.
8. Ozgunay, H., Sari, O. and Tozan, M.; Molecular investigation of valonea tannin, *JALCA* **102**, 154-157, 2007.
9. D.G.M. Vandeginste, D.L.Massart, L.M.C.Buydens, S. de Jung, P.J. Lewi and J.Smeyers-Verbeke, Handbook of Chemometrics, Part B. Elsevier. Amsterdam (1998) p.p. 349-370
10. Multivariate Calibration, 1st ed., H. Martens and T. Naes, John Wiley & Sons, 1989.