Monitoring goat milk quality during pasteurisation and ohmic treatment using UV-VIS-SWNIR spectroscopy

Martins, R.C.[1,a], Borges, S.C., Lopes, V.V.[2], Pereira, R.N.[1] and Vicente A.A.[1]

[1] IBB - Institute for Biotechnology and BioEngineering, Universidade do Minho, Campus de Gualtar, 4710-057 Braga, Portugal
[2] Laseeb, Institute of Systems and Robotics, I.S.T. Technical University of Lisbon, Av. Rovisco Pais, P-1049-001 Lisbon, Portugal
[a]email: rui.martins@deb.uminho.pt

Written for presentation at the
2007 CIGR Section VI International Symposium on
FOOD AND AGRICULTURAL PRODUCTS: PROCESSING AND INNOVATIONS
Naples, Italy
24-26 September 2007

Abstract. This study evaluates the effectiveness of UV-VIS-SWNIR spectroscopy of goat milk quality degradation during pasteurisation and ohmic heating, being performed on: i) raw goat milk; ii) non-processed milk but passing though pumps, pasteurizer and ohmic heater; and iii) processed milk by pasteurisation and ohmic heating. Spectra were collected by a transmittance probe for UV-VIS and UV-NIR wavelengths. The samples temperature was recorded (18.0 ± 2.0^oC) and the probe was always checked for bubble formation or fat residues on lens/mirror system. The integration time was set to 25s and 4s for the collection of UV-VIS and VIS-NIR spectra respectively. Data analysis was performed on each product and for each spectral range independently. The spectra were normalized by its maximum intensity and the corrected for using a robust multiplicative scatter correction algorithm. A principal component analysis was performed to the pre-processed spectra. Results show that UV-VIS-SWNIR reflectance spectroscopy provides a quick and fast assessment of goat milk characteristics and thus it can be used as an indication of the overall product variability, allowing to develop monitoring and control models for both pasteurisation and ohmic heating of goat milk.

Keywords. Goat Milk, UV-VIS-SWNIR, Pasteurisation, Ohmic heating

1 Introduction

Goat milk is a complex emulsion of fat in watery solution, containing fat, proteins, lactose and minerals; being composed of 88.6% water and 11.4% solids; containing 3.28% fat and 8.13% non-fat. Non-fat solids are composed by 4.29% lactose, 3.20% proteins and 0.64% ash (calcium, phosphorous, magnesium and potassium). Milk composition is affected by the goat's bread, region and sanitary conditions (free pasture or captivity), feeding caracteristics, health conditions and normal seasonal lactation conditions. The highest variability is observed in the free fatty acids profile and protein concentration (caseins and whey proteins). Apparently, lactose and minor

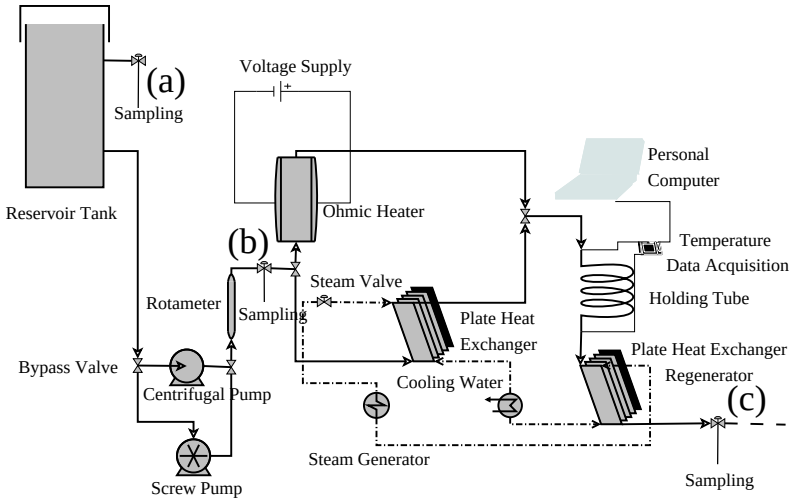

Figure 1: Goat milk pasteurisation and ohmic heating pilot plant schematic: (a) sampling site for raw milk; (b) sampling site for effects of centrifugal and screw pumps; and (c) pasteurization/ohmic heating/plate heat exchanger effects.

constituents are less subjected to biological and seasonal variance (Jandall 1996, Wong 1999, Gomes, Libera, Madureira & Araujo 2004).

Pasteurisation is known to denatured the whey proteins (lactoalbomins and lactoglobulases), which coagulate into tiny particles. Such denaturation affects the color of milk, increasing its ability to reflect all visible spectrum . Until today, fatty acids and proteins in milk interactions during thermal processing have not been fully described (Vasbinder 2002). Pasteurisation also increases Maillard reactions, and therefore the carbonyl compounds or α, β unsaturated aldehyde's resulting can form protein bounded carbonyl's. Also, free radicals produced by metal-catalysed oxidation may lead to the appearance of carbonylated aminoacids, being the carbonyl group and indication of the oxidation of milk proteins. Furthermore, there is still no information on how fat micelles are affected during pasteurisation and ohmic heating, and if these interact with caseins colloidal phase. As goat milk physico-chemical changes during pasteurisation and ohmic heating are highly complex, a multivariate strategy of monitoring and control should be applied. Under these circumstances, multivariate on-line or at-line instrumentation, such as spectrometers, may prove very valuable tools in processing control.

UV-VIS spectroscopy has not been widely applied to assess milk composition as NIR or MIR. However, the physical properties of the UV-VIS-SWNIR spectra of organic molecules provide a great potential for monitoring the described milk physical-chemical changes during processing and storage, as already explored in the NIR/Mir region, being in the food area, mostly developed for laboratory testing and not to be applied at processing lines. Therefore, this research aims at determining the effectiveness of UV-VIS-SWNIR as monitoring technique for pasteurisation control. Here in we record by spectroscopy the effects of: i) pumps; ii) heat exchanger; and iii) Ohmic heating treatment severity; and iv) storage under refrigeration.

2 Materials and Methods

2.1 Samples

Goat milk samples were collected at the day of delivery at a local dairy processing unit and processed in the pilot plant with the schematic presented in Figure 1. For each received batch, the goat milk was homogenised and thereafter different samples were collected in the pilot plant: i) without treatment (raw-milk/control at sampling place (a)); ii) pass through the centrifugal and screw pumps (sample to record pump effects at sampling place (b)); iii) pasteurization (15s, 75^oC)/plate heat exchanger and ohmic processing (15s, 75^oC)/plate heat exchanger. For each sampling site and processing run, five true replicates of samples were obtained for spectroscopy analysis.

2.2 Spectroscopy

Goat milk UV-VIS-SWNIR spectroscopy analysis was performed using a Ocean Optics high-resolution miniature fiber optics spectrometer HR4000, with a wavelength range from 200 to 1100 nm (3648 pixel). A 5 mm path transmission dip probe 1/4" UV-VIS and VIS-SWNIR probes, models RT-5MM-UV/VIS and RT-5MM-VIS/NIR and a deuterium-halogen light source Micropack DH2000-BAL was used for UV-VIS and VIR-SWNIR transmission measurements. The software Spectrasuite was used to control the spectrometer and data acquisition into a laptop PC. Spectra were obtained at the room temperature of 18 ± 2^oC. Transmission measurements were performed into the UV-VIS (deuterium) and VIS-NIR (tungsten) light sources, respectively by: (a) UV-VIS: the deuterium lamp was let to stabilize during 40 min; The spectra was recoded with a 25s integration time, averaged to 5 scans and a moving average of 5 data points, in three replicates; (b) VIS-NIR: the tungsten lamp lamp was let to stabilize during 20 min; The surface and spectra were recoded with a 4s integration time, averaged with 5 scans and a moving average of 5 data points, taken in three replicates. The dark spectra was recorded and measurements were taken with linear correction, for UV-VIS and VIS-NIR measurements, respectively. Both light spectra (deuterium and tungsten) were monitored by statistically assessing the reproducibility of the light source with measurements of ultrapure water transmittance during the several days of the experiment.

2.3 Spectra pre-processing

The collected transmittance spectra were smoothed by using a Savisky-Golay filter prior to any exploratory data analysis procedure. Afterwards, the spectra was pre-processed using a modified multiplicative scatter correction algorithm. Each spectra is corrected by using the following equation:

$$\mathbf{x}_{corr} = \mathbf{x}b + a = \mathbf{x}_{ref} \tag{1}$$

The a and b are computed by minimizing the following error:

$$\mathbf{e}_j = b\mathbf{x}_j + a - \mathbf{x}_{ref} \tag{2}$$

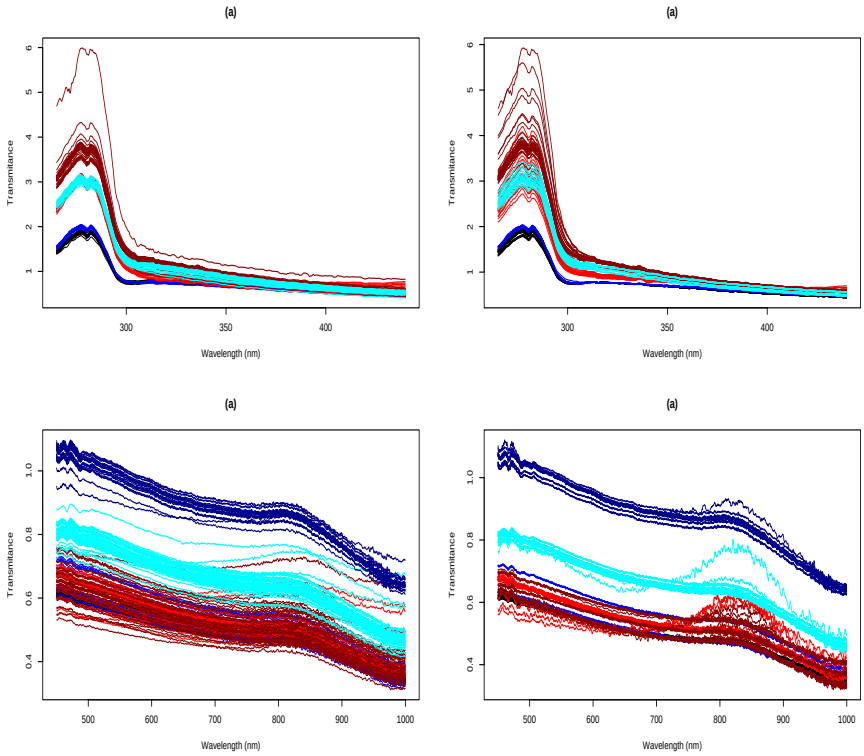

Figure 2: Goat milk UV-VIS-SWNIR spectra: (a) raw and (b) robust MSC spectra for UV-VIS spectra; and (c) raw and (d) robust MSC spectra for VIS-SWNIR spectra.

where the $\mathbf{x}_j$ is the j sample spectra and $\mathbf{x}_{ref}$ is a reference spectra.

This algorithm is based on the application of the robust least squares method to determine the a and b matrices ensuring that spectral areas that do not correspond to scattering artifacts are not taken into account. The robust least squares algorithm is implemented by the re-weighted least squares with the weights computed by using the Huber function. The algorithm high breakdown point (50%) means that existent outliers will not distort the model fitting 2 and thus, the a and b scatter correction parameters are determined using only consistent spectral areas. The iterative algorithm can be described, briefly as follow: 1) set the reference spectra ($\mathbf{x}_{ref}$) equal to the sample spectra closest to the median spectra; 2) correct the remaining sample spectra by applying the above described robust least squares procedure; 3) recompute the median spectra and iterate until convergence. The MSC spectra were thereafter subjected to principal component analysis (Gallager, Blake & Gassman 2005).

2.4 Data Analysis

The principal components analysis (PCA) was used to classify and differentiate the factor combinations that affect the goat milk quality. Principal components (PC) were calculated using

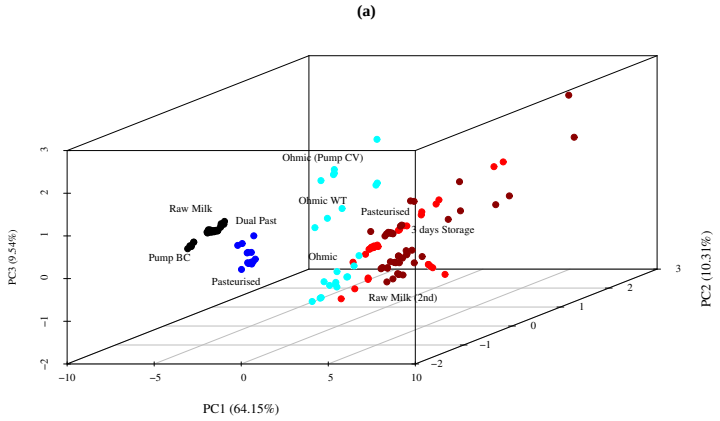

Figure 3: PCA of MSC UV-VIS spectra: (a) Triplot: PC1 (64.15%), PC2 (10.31%), PC3 (9.54%).

the singular value decomposition (SVD) after data normalisation (mean centering and variance scaling). The number of significant PC's were obtained by randomisation (comparing with the null distribution of singular values) and analysed for their singular values and component coefficients significance (Krzanowski 1998).

3 Results and Discussion

3.1 Pre-processing and descriptive assessment

Figure 2 presents the transmittance and the RMSC transmittance of goat milk spectra. The the robust MSC was capable of removing scattering artifacts from the UV-VIS spectra effectively. It has also shown that this spectra has higher variance in the 260-320nm wavelengths. This region is given by n-π electron transitions (carbonyl groups) which reflect major differences not only in milk composition in terms of free fatty acids as well as proteins, holding significant and valuable information for monitoring the goat milk quality during processing and storage. Furthermore, spectra show that the different goat milk batches present significantly different total fatty acids content due to the transmittance variance in the observed region, as goat milk was obtained from September to December 2007, being the fat content is higher during the fall season (Gomes et al. 2004, Maia, Branco, Mouro, Coneglian, Santos, Minella & Guimarães 2004). A direct qualitative analysis of the spectra does not reveals if milk proteins in the UV region suffer any red or blue shift, which could be a trace of maillard reactions or hydrolysis during processing, respectively.

The robust MSC was also effective in removing the scattering effect in the vis-swnir wavelengths. In this region, high variance was observed all over the spectra from 450 to 1000nm.. In this region, both chromatophores and carbonyl's are recorded in this region, as well in the SWNIR, the N-H, C-H, and O-H overtones, proving to record goat milk variability upon the several processing conditions. Furthermore, was directly observable in the spectra that after processing that

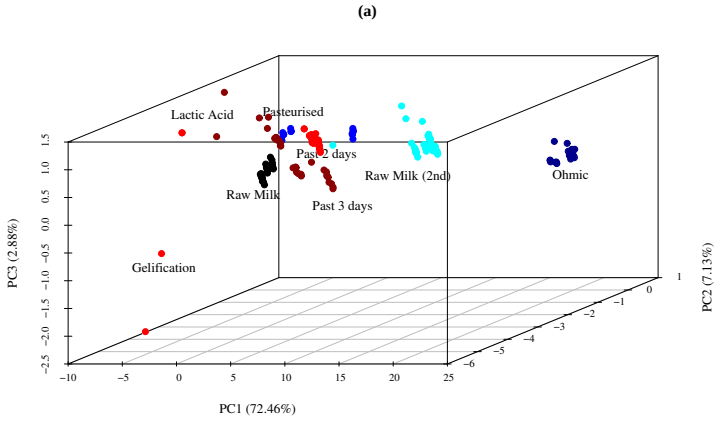

Figure 4: PCA of MSC VIS-SWNIR spectra: (a) Triplot: PC1(72.46%), PC2(7.13%), PC3(2.88%).

transmittance is higher. Such is attributed to the coagulation of the whey protein which do not precipitate under pasteurisation or ohmic heating. These are known to denatured the whey proteins, which coagulate into tiny particles, which do not precipitate, where is know that the Thiol group of α and β lactoglobulins react with caseins upon coagulation, by forming di-sulfite links with other thiol groups of caseins or dissulfite bridges with α-lactoalbomin and β-lactoglobulase (Vasbinder 2002). Moreover, a small red-shift was observed in the processed ohmic-heating milk samples. Perhaps the effect of temperature may triggered part of Maillard reactions. To better understand the differences and potential of spectroscopy in capturing the main changes of goat milk during processing, a principal component analysis was performed.

3.2 Principal component analysis

Most of the variance in the UV-VIS spectra is captured in the 1st 3PC's (PC1 (64.15%), PC2 (10.31%) and PC3 (9.54%)), accounting for 84.00% of variance in spectra. The triplot in Figure 3 shows that is possible to distinguish both raw and processed goat milks using the UV-VIS region of the spectra. This PCA analysis presents clear differences between the raw milk, pass throught the different pumps, single or dual pasteurisations, or even subjected to ohmic heating leads to a different spectra from pasteurisation. Furthermore, significant differences are also observable in the spectra suffers significant changes during storage at +5^oC for 3 days, when compared to the raw milk, ensuring that perhaps both chemical, biochemical or microbiological activity occured during storage. Most variance in the VIS-SWNIR region was captured in the first 3PC's (PC1 (72.46%), PC2 (7.13%) and PC3 (2.88%)), totalising 82.47% of observed variance. In these wavelengths is possible to differentiate between the different raw milks (e.g. fat content), processing types (pasteurisation and ohmic heating), gelification of milk proteins and increases in lactic acid during storage (see Figure 3).

4 Conclusions

This preliminary lead us to the following conclusions: i) there is a clear advantage to perform scatter artifact effect estimation by the proposed robust multiplicative scatter correction method since spectral areas containing high sample variance (due to physical/chemical composition changes) do not after the estimate. The proposed MSC modification has an high breakdown point, allowing for at least 50% possible outliers in the scatter effect estimation; ii) results shows that UV-VIS (230-420nm) and VIS-SWNIR (450-1000nm) spectroscopy can be used to characterise the different goat samples and record relevant changes of goat milk composition and physical properties from scattering effects; and iii) this preliminary study is now subjected to further statistical analysis in order to develop models that allow monitoring and control of goat milk quality during pasteurisation and ohmic heating.

Acknowledgements

The authors R.C. Martins (SFRH/BPD/26133/2005), V.V. Lopes (SFRH/ BPD/20735-/2004) and Pereira, R.N. (SFRH/BD/31635/2006) acknowledge their grants to Fundação para a Ciência e Tecnologia (FCT) and partial support (IBB/CEB and ISR/IST plurianual funds) through the POS-Conhecimento Program (FEDER funds); and Borges, S.C. acknowledge her studenship grant to the PRIME program.

References

Gallager, N. B., Blake, T. & Gassman, P. 2005. Application of extended inverse scattering correction to mid-infrared reflectance of soil. *Journal of Chemometrics* **19**: 271–281.

Gomes, V., Libera, A. M., Madureira, K. M. & Araujo, W. P. 2004. Influence of lactation stage on goat (*Capra hircus*) milk composition. *Brasilian Journal of Veterinary Research and Animal Science* **4**: 339–342.

Jandall, J. M. 1996. Comparative aspects of goat and sheep milk. *Small Ruminant Research* **22**: 177–185.

Krzanowski, W. J. 1998. *Principles of Multivariate Analysis*, Oxford University Press, Oxford.

Maia, F. J., Branco, A. F., Mouro, G. F., Coneglian, S. M., Santos, G., Minella, T. & Guimarães, K. C. 2004. Inclusão de fontes de óleo na dieta de cabras em lactação: produção, composição e perfil dos àcidos graxos no leite. *Revista Brasileira de Zootecnia* **35**, 1504–1513.

Vasbinder, A. J. 2002. Casein-whey protein interactions in heated milk. PhD thesis, Universiteit Utrecht, The Netherlands.

Wong, N. P. 1999. *Fundamentals of Dairy Chemistry*. Aspen Publishers, Gaithersburg.