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Abstract. The ability to authenticate the method of production of the food products has become a major challenge for scientists and monitoring bodies. This study compared two methods based on the use of visible reflectance spectrum of fat tissues to discriminate pasture-fed (n=76), concentrate-fed (n=79) and concentrate-finished (for 28 days) pasture-fed (n=69) lamb carcasses. The reflectance spectrum of perirenal and subcutaneous caudal fat was measured at slaughter and at 24 h *post mortem*. The method 1 used an index calculated from the reflectance spectrum data at wavelengths between 450 and 510 nm, whereas the method 2 was performed at wavelengths between 400 and 700 nm. Method 2 yielded higher overall proportion of correctly classified lambs than method 1 (92.9% *vs.* 64.7%, P<0.0001), regardless of the measurement time and site. The proportion of lambs which were correctly classified using method 2 was 95.6% and 95.9% for measurements made on perirenal fat at slaughter and at 24 h *post mortem*. This study indicates the importance of the zones of light absorption by carotenoid and haeminic pigments in the discrimination between lambs from three different feeding systems.

Keywords. Authentication – pasture-feeding – concentrate-feeding – concentrate-finishing – spectroscopy – sheep – lamb

L'analyse du spectre de réflectance du tissu adipeux permet de discriminer trois modalités d'engraissement des agneaux : à l'herbe, en bergerie, ou avec une finition en bergerie après pâturage

***Résumé**. La possibilité d'authentifier le mode de production des animaux à partir de leurs produits est devenue un enjeu majeur pour les scientifiques et les organismes de contrôle. Cette étude a comparé deux méthodes basées sur l'analyse du spectre de réflectance dans le visible du tissu adipeux pour discriminer les agneaux d'herbe, les agneaux de bergerie et les agneaux d'herbe finis en bergerie pendant 28 jours. Les spectres de réflectance du gras périrénal et du gras sous-cutané caudal ont été mesurés à l'abattage et à 24 h* post mortem*. La méthode 1 a utilisé un indice calculé à partir de la zone du spectre de réflectance comprise entre 450 et 510 nm, alors que la méthode 2 a utilisé l'ensemble du spectre de réflectance entre 400 et 700 nm. La proportion d'agneaux correctement classés a été plus élevée avec la méthode 2 qu'avec la méthode 1 (92,9% vs. 64,7%, P<0.0001), indépendamment du moment et du site de la mesure. La proportion d'agneaux correctement classés par la méthode 2 était de 95,6% et 95,9% pour les mesures effectuées sur le gras périrénal à l'abattage et à 24 h* post mortem*. Cette étude montre l'importance des zones d'absorption de la lumière par les pigments caroténoïdes et héminiques dans la discrimination entre les trois types d'agneaux.*

***Mots-clés.** Authentification – agneau d'herbe – agneaux de bergerie – finition – spectroscopie – ovins – agneau*

I – Introduction

Consumers show growing interest in the method of production of their food and growing preference for food products of pasture-based systems of production (Prache and Thériez, 1999). It is therefore important to be able to authenticate food products from particular production systems. The measurement of the reflectance spectrum of the fat has proven to be of practical

interest since it is non-invasive, takes little time and can easily be implemented in the meat industry with a portable spectrophotometer. Prache and Thériez (1999) proposed a mathematical analysis of the reflectance spectrum of the fat at wavelengths between 450 and 510 nm (which corresponds to the zone of light absorption by carotenoids) to quantify the intensity of the signature of these pigments and discriminate pasture-fed from stall-fed lamb carcasses. Dian *et al.* (2007) increased the reliability of the discrimination between pasture-fed and stall-fed lambs by using the full data set of the reflectance spectrum at wavelengths between 400 and 700 nm. However, because of grass shortage during the summer period, pasture-fed lambs are often stall-finished with low-carotenoid diets. The purpose of this study was therefore to further test the reliability of these two spectral methods for discriminating lamb carcasses from less contrasted production systems (pasture-feeding, concentrate-feeding and concentrate-finishing after pasture-feeding).

II – Materials and methods

This study was carried out over 5 years (2008-2012) at two experimental farms of INRA (Unité Expérimentale des Monts d'Auvergne and Unité Expérimentale des Ruminants de Theix). A total of 224 Romane breed male lambs were used: 76 were fed pasture for at least 60 days (P), 79 were stall-fed concentrate and straw (S) and 69 were fed pasture for at least 60 days followed by an abrupt switch to concentrate and straw indoors (PS) for 28 days. The P and PS lambs grazed pastures that were maintained at a leafy stage and offered *ad libitum*. The P lambs were fed pasture until slaughter. The PS lambs were expected to gain on average 7.0 kg LW gain (LWG) during the finishing period indoors. Stall-fed lambs were fed *ad libitum* a commercial concentrate, containing no green vegetative matter, and straw until slaughter. These last feedstuffs were given also to PS lambs during the stall finishing period. The lambs were slaughtered when they had reached a target body condition score of 3 (on a scale of 0 to 5), which was manually assessed by a skilled technician.

The reflectance (R) spectrum of perirenal and subcutaneous caudal fat was measured on all lambs, using a MINOLTA CM-2002 spectrophotometer (D65 illuminant, observer angle 10°). For each tissue, 5 measurements were made at slaughter and at 24 h *post mortem*. In method 1, the fat reflectance spectrum data were used at wavelengths between 450 and 510 nm to calculate an index quantifying light absorption by carotenoid pigments stored in the fat. The R spectrum was translated to give a reflectance value at 510 nm of zero (TR). On the translated spectrum, the integral value ($I_{450\text{-}510}$) was calculated as follows: $I_{450\text{-}510}$ = [(TR 450/2) + TR 460 + TR 470 + TR 480 + TR 490 +TR 500 + (TR 510/2)] × 10

The integral value was averaged and then linear discriminant analysis was performed, followed by a cross-validation procedure, to classify the fat samples according to feeding treatment, using Minitab software v. 13 (Minitab Inc., Paris).

In method 2, the full R data set at wavelengths between 400 and 700 nm was used. The R data were averaged, then transformed (log (1/R)) and exported into Win ISI II version 1.6 software (Infrasoft International, Port Matilda, PA, USA) for multivariate analysis. The raw R spectra of each tissue representing the three feeding treatments were submitted to discriminant analysis using a partial least squares discriminant analysis (PLS-DA) approach. A principal component analysis (PCA) performed beforehand was used to rank the reflectance spectra from each feeding treatment according to the Mahalanobis distance (H) to the average R spectrum, in order to detect sample outliers (H>3). No outliers were found. The models were tested via a cross-validation procedure, in which 56 randomly chosen samples were temporally removed from the initial data set to be used for validation (i.e. a quarter of all data samples). The PLS-DA model was developed based on the other samples (calibration samples) and used to classify the validation samples. This procedure was repeated four times, i.e. until all data set samples had been used through the validation procedure. The cross-validation error of the models was calculated.

The proportion of correctly classified samples was analyzed using the CATMOD procedure of the SAS (1999) software package using a four-factor model (feeding treatment: P *vs.* S *vs.* PS; discrimination method used in the fat: method 1 *vs.* method 2; site of measurement: perirenal *vs.* subcutaneous caudal fat; and time of measurement: at slaughter *vs.* at 24 h *post mortem*).

III – Results and discussion

The proportion of correctly classified samples using linear discriminant analysis performed on $I_{450-510}$ (method 1) was 67.1%, 84.8% and 60.9% for P, S and PS lambs, respectively (70.9% on average), when the measurement was made on perirenal fat at slaughter, and 68.4%, 84.8% and 66.7% for P, S and PS lambs, respectively (73.3% on average), when the measurement was made on perirenal fat 24 h *post mortem* (Fig. 1). This method correctly classified 55.3%, 68.4% and 23.1% of the P, S and PS lambs, respectively (i48.9% on average), when the measurement was made on subcutaneous caudal fat at slaughter, and 76.3%, 69.6% and 50.7% of the P, S and PS lambs, respectively (65.5% on average), when the measurement was made on subcutaneous caudal fat 24 h *post mortem*.

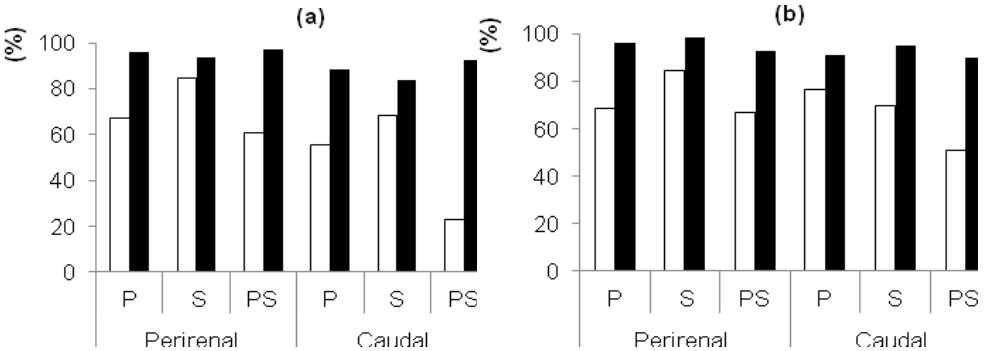

Fig. 1. Proportion of correctly classified lambs using method 1 (integral value $I_{450-510}$, white symbols) and method 2 (full reflectance spectrum data sets at wavelengths between 400 and 700 nm, black symbols) on perirenal and subcutaneous caudal fat measured at slaughter (a) and at 24 h *post mortem* (b). P: Pasture-fed lambs; S: Stall-fed lambs; PS: Stall-finished pasture-fed lambs

The method 2 allowed for the correct classification of 96.1%, 93.7% and 97.1% of the P, S and PS lambs, respectively (95.6% on average), when the measurement was made on perirenal fat at slaughter, and 96.1%, 98.7% and 92.8% of the P, S and PS lambs, respectively (95.9% on average), when the measurement was made on perirenal fat 24 h *post mortem* (Fig. 1). This method correctly classified 88.2%, 83.5% and 92.8% of the P, S and PS lambs, respectively (88.2% on average), when the measurement was made on subcutaneous caudal fat at slaughter, and 90.8%, 94.9% and 89.9% of the P, S and PS lambs, respectively (91.9% on average), when the measurement was made on subcutaneous caudal fat 24 h *post mortem*.

By using all the reflectance spectrum data at wavelengths between 400 to 700 nm, method 2 showed a higher performance ($P<0.001$) than method 1(Fig. 1), which only used the reflectance spectrum data at wavelengths from 450 to 510 nm to calculate $I_{450-510}$. Taking both sites, both measurement times and all feeding treatments together, the overall proportion of correctly classified lambs was 64.7% for method 1 and 92.9% for method 2. The proportion of correctly classified lambs also differed between feeding treatments (79.8%, 84.8% and 71.8% for P, S and PS lambs, $P<0.0001$), measurement sites (83.9% for perirenal and 73.6% for subcutaneous caudal fat, $P<0.0001$) and measurement times (75.9% at slaughter and 81.6% at 24 h *post mortem*, $P<0.001$). Method 2 reliably discriminated the 3 types of lambs when the measurement

was made on perirenal fat at slaughter (the proportion of correctly classified lambs ranging between 93.7% and 97.1%) and at 24 h *post mortem* (the proportion of correctly classified lambs ranging between 92.8% and 98.7%).

The PCA loadings of the two PC axes for perirenal fat measured at 24 h *post mortem* are shown in Figure 2. PC1 and PC2 axes explained 60% and 24% of variability. The highest loading was situated around 460 to 480 nm for PC1, indicating the importance of carotenoid pigments in the discrimination between the lambs from three different feeding systems. The PC1 and PC2 axes loading values at wavelengths around 420 nm and the PC2 axis loading value at wavelengths around 550-560 nm indicated that haeminic pigments (Prache *et al.,* 1990) may also be involved in the discrimination between the lambs from three different feeding systems, as already suggested by Dian *et al.* (2007).

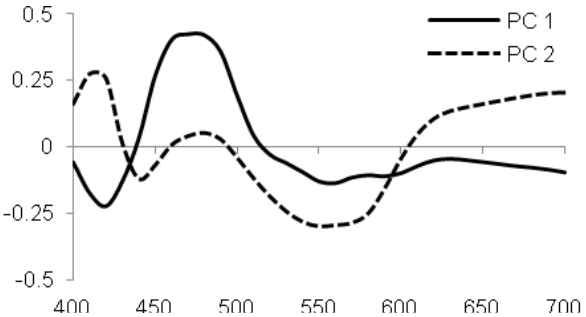

Fig. 2. Loadings for the two principal components axes (PC1 and PC2) for perirenal fat samples measured 24 h *post mortem*

IV – Conclusions

Using the full set of the reflectance data of the carcass fat in the visible range (at wavelengths between 400 nm to 700 nm) enabled to reliably discriminate lamb carcasses from 3 production systems (pasture-feeding, concentrate-feeding and stall-finishing after pasture). The reliability of the discrimination at best with measurements made on perirenal fat at 24 h *post-mortem*. The proportion of lambs which were correctly classified using this method was 95.6% and 95.9% for measurements made on perirenal fat at slaughter and at 24 h *post mortem*. This method is of obvious practical interest for the meat industry, since it is non-invasive, rapidly and easily on-line implemented with a portable spectrophotometer.

References

Dian P.H.M., Andueza D., Barbosa C.M.P., Amoureux S., Jestin M., Carvalho P.C.F., Prado I.N. and Prache S., 2007. Methodological developments in the use of visible reflectance spectroscopy for discriminating pasture-fed from concentrate-fed lamb carcasses. In: *Animal,* 1(8), p. 1198-1208.

Prache S., Aurousseau B., Thériez M., & Renerre M., 1990. Les défauts de couleur du tissu adipeux sous-cutané des carcasses d'ovins. In: *INRA Productions Animales,* 3, p. 275-285.

Prache S. and Thériez M., 1999. Traceability of lamb production systems: carotenoids in plasma and adipose tissue. In: *Animal Science,* 69, p. 29-36.