Lamb muscle identification by near infrared spectroscopy

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Introduction

Meat and meat products made from the muscle tissue of beef, pork and lamb constitute a major staple food around the world. The food quality of meat is highly connected with biological and biochemistry functions in the animal body and loss of homeostasis in converting the muscle to meat.

When viewed as a whole, the biological image of muscle is one of a dynamic tissue that is highly specialised. There are, however, important differences among species and among muscles of the same animal, especially when muscle function is different.[1] For many years a considerable amount of research effort has been directed towards developing methods for determining the quality of the muscle "as meat". Conventional chemical procedures are too slow for many purposes and use hazardous and (or) expensive chemicals.[2] Near Infrared (NIR) Reflectance Spectroscopy can be used to rapidly analyse agricultural products with little or no sample preparation. The use of NIR to measure fat, moisture and protein content in homogenised beef and pork samples, either in transmittance or reflectance mode has been reported by several authors.[3–6] Since a cut of meat is a multicomponent system composed of blood, collagenous substances, fat and different muscles, it is important to know how individual muscles behave in the NIR region with the objective of setting up a method for studying and assessing the quality of muscles as "meat". Clearly, at present, both sample preparation and presentation (geometry) to the instrument are a bottleneck in the assessment of chemical parameters and optical properties of meat and meat products by NIR. This work reports the useful wavelengths in the visible and near infrared region to assess the quality of different lamb muscles. We also attempt to study the effects of presentation of the tissue (geometry) to the instrument as intact or minced in the development of calibration models for moisture, crude protein and intramuscular fat in different muscles.

Materials and methods

Samples of dissected and homogenised (Robotcoupe, R3, France) *longissimus dorsi*, *supraspinatis*, *infraspinatis, semitendinosus, semimembranosus* and *rectus femoris* lamb muscle (51 lamb × 6 muscles) were used. Samples were scanned intact and minced in the vis/NIR (400–2500) by diffuse reflectance (R mode) on a white ceramic in a monochromator (NIRSystems 6500, Silver Spring, MD, USA). Data were manipulated using ISI version 3.01 software (ISI, Port Matilda, PA, USA).[7]

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Results and discussion

Spectral characterisation of intact and minced samples

Figure 1 shows the mean and standard deviation of the mean spectrum of intact and minced lamb muscles. The mean spectrum of the intact samples shows absorption bands at 424 and 550 nm in the visible region related to the Soret and oxymyoglobin absorption bands, respectively[8–10] where myoglobin has an absorption band at 555 nm.[11] In the NIR region, the mean spectrum shows absorption bands at 762 nm related to the O–H third overtone[12] or an absorption band produced by the oxidation of the myoglobin (deoxymyoglobin).[13] The absorption band at 978 nm is related to the O–H second overtone, at 1200 nm to the C–H second overtone, at 1458 nm to the O–H first overtone, at 1730 nm and at 1766 nm to the C–H first overtone, at 1932 nm with water absorption and at 2308 nm with the C–H combination bands.[12,14,15] The mean spectrum of the minced sample shows the same absorption bands as that of the mean spectrum of the intact samples.

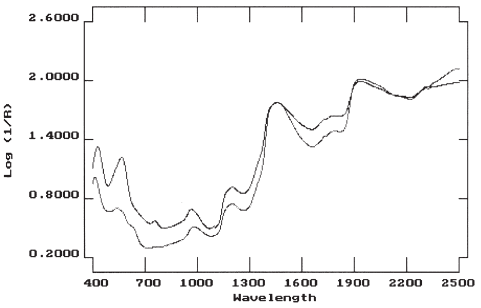

Figure 1. NIR mean spectrum of both minced lamb muscles (upper line) and intact lamb muscles (low line).

Chemical data of lamb muscles

Table 1 shows the chemical data for the dissected lamb muscles (6 muscles × 51 lambs) (*longissimus dorsi, infraspinatis, supraspinatis, semimembranosus, semitendinosus* and *rectus*

Table 1. Mean values for moisture, crude protein (CP) and intramuscular fat (IMF) in dissected lamb muscles g kg^{-1}(on a fresh weight basis).

		M	CP	IMF	*n*
Longissimus dorsi	avg.	690.9^{a}	220.7^{a}	25.4^{a}	51
	SD	16.5	10.4	14.1	
Infra spinatus	avg.	716.8^{b}	204.6^{b}	18.6^{a}	51
	SD	21	10.8	12.9	
Supra spinatus	avg.	718.6^{b}	196.4^{b}	20.9^{a}	51
	SD	21.6	29.9	14.8^{b}	
Semimembranosus	avg.	710.1^{b}	224.6^{a}	12.8^{b}	51
	SD	21.6	10.3	8.6	
Semitendinosus	avg.	712.1^{b}	208.6^{b}	20.9^{a}	51
	SD	22.3	16.2	12.9	
Rectus femoris	avg.	701.0^{c}	213.01^{a}	23.6^{a}	51
	SD	20.8	11.5	15.4	

avg.: average
SD: standard deviation, n: number of samples
M: moisture
IMF: intramuscular fat
Different superscripts in the column indicates significant statistic differences at $P < 0.05$

femoris). Significant statistical differences ($P < 0.05$) were found among muscles in the content of moisture, crude protein and intramuscular fat. Table 2 shows the interrelations between chemical data.

Table 2. Intercorrelation between chemical parameters.

	M	CP	IMF
M	1	– 0.180	– 0.127
CP		1	– 0.070
IMF			1

M: moisture
CP: crude protein
IMF: intramuscular fat

NIR calibration and cross-validation statistics for intact, minced and individual lamb muscle samples

Table 3 shows the calibration statistics for crude protein (CP), moisture (M) and intramuscular fat (IMF) for intact samples. The R^2 and *SECV* for M, CP and IMF in the intact samples were 0.55 (*SECV*: 15.5), 0.71 (*SECV*: 8.8) and 0.34 (*SECV:* 8.2) respectively, in g kg^{-1} . Based in the optical and physico–chemical properties of the muscle, we have developed some hypotheses about why we obtained poor calibrations in the intact presentation. One explanation lies in the structure of the fibres in the muscle. These fibres in the muscles interfere with light and trap it in different ways. The presence of A (anisotropic) and I (isotropic) bands along the axis of the fibres in the muscle produce the so called birefringence effect in the cut of meat. Meat is anisotropic with a higher reflectance when the incident illumination is perpendicular to the muscle fibres.[16] The structure of the muscle interferes with the amount of light, which comes from the muscle to the detectors in the instrument. Some light is trapped and lost at the same time from the piece of meat. If it is not detected, some information escapes from the piece of meat and the instrument does not record it.

Table 4 shows the R^2 and *SECV* for the calibration statistics from the minced sample for M, CP and IMF. The results were 0.73 (*SECV*: 10.4), 0.83 (*SECV*: 5.5) and 0.76 (*SECV*: 4.7) for moisture, protein and fat, respectively in g kg^{-1}. The high *SEC* and *SEC*(*V*) values may occur for several reasons. One could be that the wavelengths in the equation and resulting regression coefficients do not reliably predict the reference methods. For the minced samples, either an inadequate calibration population or inaccurate laboratory values affects the calibration models. Reasons for this lie in the extreme variability

Table 3. NIR calibration and cross-validation statistics for moisture, crude protein and fat in intact lamb samples in R mode in g kg^{-1} (on a fresh weight basis).

	n	Mean	*SD*	*SEC*	R^2	*SECV*	1-*VR*	*T*
IMF	277	17.5	8.8	6.9	0.34	8.1	0.18	9
CP	283	213	12.4	6.6	0.71	8.8	0.49	9
M	289	709	19.4	12.9	0.55	15.5	0.36	4

n: number of samples
SD: standard deviation
SEC: standard error of calibration
R^2: coefficient of multidetermination in calibration
SECV: standard error of cross-validation
1-*VR*: coefficient of determination in cross-validation
CP: crude protein
M: moisture
IMF: intramuscular fat
T: number of factors used to perform the calibration models.

Table 4. NIR calibration and cross-validation statistics for moisture, crude protein and fat in minced lamb samples in R mode in g kg^{-1} (on a fresh weight basis).

	n	Mean	*SD*	*SEC*	R^2	*SECV*	1-*VR*	*T*
IMF	234	16.2	8.6	4.4	0.73	4.7	0.71	4
CP	271	213	12.0	5.0	0.83	5.5	0.79	9
M	278	710	19.5	9.4	0.76	10.3	0.72	4

n: number of samples
SD: standard deviation
SEC: standard error of calibration
R^2: coefficient of multidetermination in calibration
SECV: standard error of cross-validation
1-*VR*: coefficient of determination in cross-validation
CP: crude protein
M: moisture
IMF: intramuscular fat
T: number of factors used to perform the calibration models.

of the set of samples used [six different muscles, different ages (body weights) of slaughter, sex] to perform the calibration. The presentation shows the minced sample gives higher R^2 for calibration and cross-validation models than those obtained with the intact set of samples. The homogenisation of the sample broke the structure of the muscle (fibres) and disarranged the cells in the tissue. This allows the instrument to read more information from the sample than in the intact geometry. But, also, after prolonged maceration in a blender muscle fibre fragments disintegrate and their myofibrils are released into suspension causing loss of some constituents, principally lipids and moisture, as well as soluble proteins. The use of homogenisation in some cases may alter the refractometry index of the tissue causing a denaturation of the tissue by increasing protein coagulation.[17,18] Another reason for the poorer calibration statistic lay in the time between the spectroscopic analysis and the chemical analysis. It is not new for the NIR methodology that we must do the chemical analysis at the time of collecting the optical data. The water is the most volatile component and variation in the moisture content also affects other parameters present in the sample (fat and protein). Consequently, the dissection of the sample affected the composition of the muscle, principally moisture and fat.

Individual muscles, intact and minced presentation

Table 5 shows the results for calibration and cross-validation for CP, IMF and M for each individual muscle in the minced presentation. Each muscle shows a different behaviour in the calibration models for the three parameters estimated. In the intact presentation, the *infraspinatis* and *supraspinatis* muscles had the best R^2 in the calibration for crude protein and moisture, respectively. The intact geometry presentation of the sample can not achieve good calibrations for IMF. One explanation for this is simply that the fat is either in the cells or in the intracellular spaces and the complex system of the muscle does not allow the penetration of light into the muscle structure. In the minced geometry presentation, each muscle behaves in a different way, according its function in the body and biochemistry. With these results we could suggest that a multicomponent and multivariate effect of each individual muscle in the pooled data is causing a distortion in the overall results either in calibration or cross-validation statistics. Such differences in muscle function affect composition, tissue architecture and, hence, optical properties. These confounding factors cause problems in NIR calibrations

Table 5. NIR calibration and cross-validation statistics for moisture (M), crude protein (CP) and intramuscular fat in individual minced muscle samples in g kg^{-1} (on a fresh weight basis).

	n	Mean	*SD*	*SEC*	R^2	*SECV*	1-*VR*	*T*
Longissimus dorsi								
M	44	687	17.5	9.2	0.724	12.6	0.501	4
CP	44	222	10.3	4.3	0.824	8.0	0.417	6
IMF	44	27	13.7	11.7	0.260	12.3	0.196	1
infra spinatis								
M	44	716	22.6	11.0	0.763	17.1	0.447	5
CP	43	206	9.4	3.7	0.839	7.8	0.323	6
IMF	41	17	8.6	4.2	0.766	7.9	0.587	4
supra spinatis								
M	41	721	20.9	5.9	0.920	11.8	0.689	7
CP	44	200	9.5	7.3	0.410	9.2	0.087	3
IMF	42	21	10.7	3.6	0.884	7.4	0.547	5
Semimembranosus								
M	37	711	13.3	2.7	0.958	8.7	0.606	7
CP	44	224	9.3	7.9	0.278	10.1	0.155	2
IMF	41	12	6.8	3.9	0.668	5.1	0.450	4
Semitendinosus								
M	40	715	19.5	10.6	0.703	13.3	0.561	2
CP	41	209	8.5	7.6	0.196	8.8	0.039	1
IMF	39	19	10.1	5.6	0.686	7.0	0.516	3
rectus femoris								
M	41	05	14.2	11.9	0.290	12.5	0.234	1
CP	43	213	10.4	6.4	0.610	7.8	0.457	3
IMF	39	21	9.5	5.5	0.688	7.2	0.494	3

n: number of samples
SD: standard deviation
SEC: standard error of calibration
R^2: coefficient of multidetermination in calibration
SECV: standard error of cross validation
1-*VR*: coefficient of determination in cross validation
T: number of factors used to perform the calibration models.

for general meat analysis but these same differences can also be used to advantage to discriminate between different muscle types.

Conclusions

We can draw many conclusions from these experiments. The most obvious is that the geometry of presentation affects the optical properties of the muscle that is intact or minced. The dissection of the muscle alters the chemical relationships in the sample. Losses of moisture and the difficulties related with the IMF chemical determination were the main factors that affect the performance of the calibrations.

The different types of muscle seem to be responsible for changes in the calibrations when we either put together all the samples or compare an individual calibration for each muscle.

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