RAMAN SPECTROSCOPY FOR THE DIFFERENTIATION OF MUSCLES AND TISSUES IN MEAT USING CHICKEN AS A MODEL SYSTEM

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I. INTRODUCTION

Raman spectroscopy is a vibrational spectroscopy method that provides information on the composition and molecular structure of a sample [1].The technique has the advantage of being non-invasive and is fast in analysis [2]. Meat is usually sold commercially as different cuts of muscles with large variations in quality and financial value. It is important for consumers that high-value meat products are authentic and have not been adulterated with low quality animal meats or tissues [3]. Unscrupulous processors of comminuted meats such as minced meat and sausages can deliberately add varying amounts of undeclared animal products to maximise profit. It is important for the meat industry, and for food regulatory agencies and retailers, to have fast, robust and simple methods for detecting such frauds. In this study, Raman spectroscopy with chemometric analysis of the data was assessed for this purpose. Using a fresh broiler chicken as a model, ten muscle and four tissue types were analysed.

II. MATERIALS AND METHODS

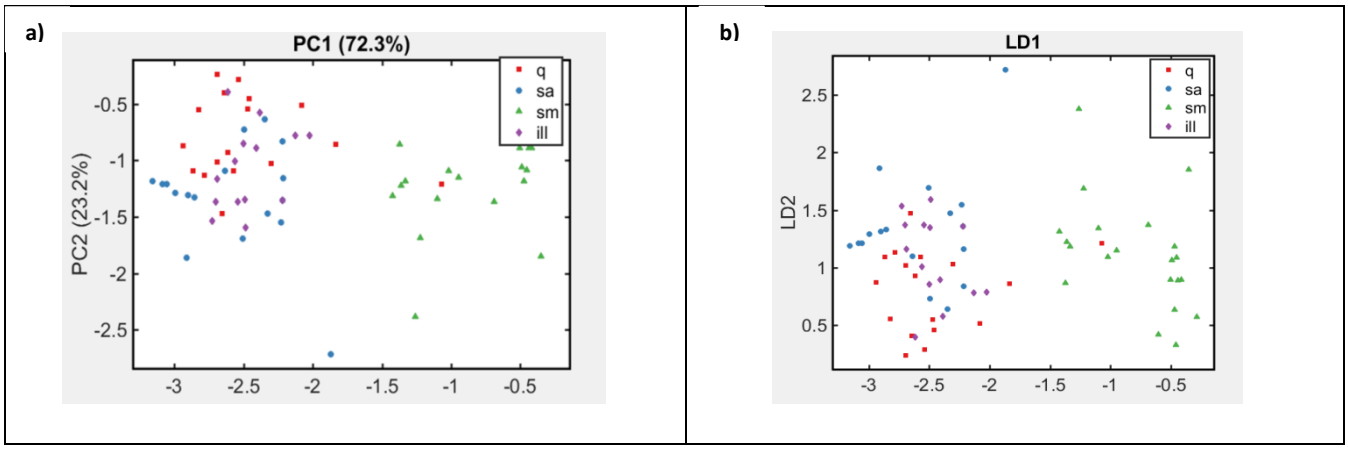
Muscles excised from different parts of the chicken carcass were as follows: thigh, *semimembranosus* (sm)*, illiotibialis* (ill)*, quadriceps femoris* (q) *and sartorious* (sa); leg, *peroneus longus* (pl)*, gastrocnemius* (g)*, tibialis anterior* (ta) and *semitendinosus* (st); breast, *pectoralis major* (pm) and *pectoralis minor* (pn). Samples of bone (b), cartilage (c), fat (f) and skin (sk) tissue were also tested. Three replicate samples from each muscle and tissue type were analysed.

Raman measurements were performed using a Perkin Elmer RamanStation 400F dispersive Raman spectrometer with a near-infrared 785 nm laser excitation source delivering 100mW at the sample and a CCD detector cooled to −50°C. Spectra were obtained at 20 s acquisition time, with 5 accumulations over a spectral range of 200 to 3200 cm^{-1}. After data pre-processing to remove cosmic ray effects, spectra were analysed by multivariate analysis (PCA and LDA) using Matlab R2017b with the IRootLab version 0.17.8.22-d toolbox (Mathworks). Three replicate samples were analysed for each muscle and tissue type with a total of 21 spectra collected for each sample replicate.

III. RESULTS AND DISCUSSION

Raman spectra vary in relation to meat sample composition (biochemical composition, lipid and collagen content) and protein secondary structure (α-helix and β-sheet) [2]. Principal Component Analysis (PCA) and Linear Discriminant Analysis (LDA) extract spectral components from large data sets, producing patterns to show similarities and differences between samples [4]. In this study, we applied these methods to fresh broiler chicken muscle and tissue samples.

Spectral peaks (not shown) were observed between 1645 -1685 $cm^{-}1$ for the thigh, leg and breast muscles. Presumably, these peaks were from the protein amide I α-helix vibrational modes, and from C=O stretching vibrations and peptide carbonyl (C-N) stretching [5]. PCA plots for thigh muscles (Fig. 1a) showed grouping of three muscles (*sartorius, illiotibialis* and *quadriceps femoris*) while *semimembranosus* (sm) grouped in another cluster. PC1 accounted for 72.3% and PC2 accounted for 23.2% of the variation. The LDA plots (Fig. 1b) similarly revealed differences in the *semimembranosus* muscle compared with the other thigh muscles.



a) PCA plot thigh muscles **b) LDA plot thigh muscles**

Figure 1. PCA (a) and LDA (b) plots of Raman spectroscopic data for thigh and leg muscles from fresh broiler chicken carcasses. Abbreviations for different muscles are described in Materials and Methods.

The two chicken breast muscles (*p. major* and *p. minor*) had similar spectra that could not be resolved by PCA and LDA plots (data not shown). The two white breast muscles are used in flight in the chicken and are found attached together, which may account for this high degree of Raman similarity. Overall, broiler chicken muscles are expected to have more subtle differences between them than those found in beef or lamb, since chickens are slaughtered at an early age and their growth rates are fast. The spectra for skin and fat (not shown) had sharp bands at 1660 and 1662 cm^{-1}, respectively and were grouped together into one distinct cluster in the LDA plot. These bands were likely derived from unsaturated fatty acid (C=C) bond stretching vibrations [6]. The LDA plot for all tissues showed cartilage and *semimembranosus* formed distinct clusters next to each other. Discrimination was based on the 866 and 870cm^{-1} bands for collagen.

IV. CONCLUSION

Raman spectroscopy with chemometric analysis can discriminate some but not all muscles and tissues in broiler chicken carcasses. This suggests Raman spectroscopy could be applied in the red meat processing industry to detect authenticity of meat cuts. If so, hand-held (portable) Raman spectrometers will be the way of the future.

ACKNOWLEDGEMENTS

We thank Dr Saeidah Ostovar pour and Mr Stuart Hombsch for helpful comments and technical assistance. This work was funded by the Australian Meat Processor Corporation.

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