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# Rapid detection of frozen-then-thawed minced beef using multispectral imaging and Fourier transform infrared spectroscopy



MEAT SCIENCE

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#### ABSTRACT

In recent years, fraud detection has become a major priority for food authorities, as fraudulent practices can have various economic and safety consequences. This work explores ways of identifying frozen-then-thawed minced beef labeled as fresh in a rapid, large-scale and cost-effective way. For this reason, freshly-ground beef was purchased from seven separate shops at different times, divided in fifteen portions and placed in Petri dishes. Multi-spectral images and FTIR spectra of the first five were immediately acquired while the remaining were frozen (-20 °C) and stored for 7 and 32 days (5 samples for each time interval). Samples were thawed and subsequently subjected to similar data acquisition. In total, 105 multispectral images and FTIR spectra were collected which were further analyzed using partial least-squares discriminant analysis and support vector machines. Two meat batches (30 samples) were reserved for independent validation and the remaining five batches were divided in training and test set (75 samples). Results showed 100% overall correct classification for FTIR test set and external validation set respectively.

#### 1. Introduction

The safety of meat and meat products is currently very much in focus, owing to calamities with microbiological outbreaks, dioxin contamination and other threats to human health (EFSA (European Food Safety Authority), 2015, 2017). At a time when consumer awareness of nutrition and health is increasing, it is important that meat products should be safe for all consumers. This is the case particularly with beef, which is a large part of human diet and therefore its quality is of importance not only to consumers, but also to food authorities and the meat industry. Meat production encompasses slaughterhouses, packers, company- owned distribution and supply operations and importers. Wholesaling includes the nationwide network of meat and meat product wholesalers and related warehouse and transportation units. Retailing includes locations where meat is consumed "onpremise," such as restaurants. "Off-premise" retail outlets are supermarkets, butchers, warehouse stores, and similar locations. Therefore non-compliance to labels may lead to loss of consumer trust and subsequently loss of revenue for food retailers, as well as have other economic and safety consequences (Alamprese, Casale, Sinelli, Lanteri, & Casiraghi, 2013). Nevertheless, meat can be an attractive target for fraudulent and deceptive practices, such as selling

adulterated with cheaper ingredients meat and thawed meat as fresh due to its higher market price.

Various analytical methods have been proposed for the detection of frozen-then-thawed meat including enzymatic, DNA-based, microscopic and sensory techniques (Ballin & Lametsch, 2008). However, in order to ensure constant large-scale and effective monitoring, rapid, low-cost, non-invasive methods or methods requiring a small sample have to be employed. These rapid methods involve various sensors such as hyperspectral (HSI) and multispectral imaging (MSI), NIR and Raman spectroscopy. Such sensors have been previously used in tandem with various data analysis methods (Dai, Sun, Xiong, Cheng, & Zeng, 2014; Ropodi, Panagou, & Nychas, 2016) for spoilage estimation (Argyri et al., 2013; Argyri, Panagou, & Nychas, 2014; Dissing et al., 2013; Panagou, Papadopoulou, Carstensen, & Nychas, 2014; Wu & Sun, 2013), as well as fraud detection e.g. adulteration of raw meats, including beef or lamb with pork meat, beef with turkey, offal, fat trimmings and (Alamprese horsemeat et al., 2013; Kamruzzaman, Sun. ElMasry, & Allen, 2013; Morsy & Sun, 2013; Ropodi, 2017; Panagou, & Nychas, Ropodi, Pavlidis. Mohareb. Panagou, & Nychas, 2015). In terms of detection of frozen-then-thawed meat some work has been done with fish fillets and HSI (Cheng et al., 2015), minced chicken, pork and turkey and MIR spectroscopy (Al-

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Jowder, Kemsley, & Wilson, 1997), beef and NIR spectroscopy (Downey & Beauchêne, 1997), as well as pork and HSI (Barbin, Sun, & Su, 2013; Ma et al., 2015; Pu, Sun, Ma, & Cheng, 2015). The successful use of the sensors coupled with data modeling would provide a rapid, low-cost and -in the case of MSI- a non-invasive solution for large-scale screening purposes, providing both industry professionals and food safety authorities a way to conduct regular checks at all stages of the retail chain. To our knowledge, two or more sensors including multispectral imaging and vibrational spectroscopy have not been used previously either in combination or separately for frozen-then-thawed minced beef detection.

Thus, the objective of this study was to (a) evaluate the potential of multispectral imaging and FTIR spectroscopy in tandem with data analysis techniques to identify frozen-then-thawed minced beef, and (b) compare–and combine if possible– sensors and models.

#### 2. Materials and methods

#### 2.1. Experimental design/sample preparation

In this study, freshly-ground beef was purchased at seven separate dates from different butcher shops and supermarkets in Athens, Greece, with no prior knowledge as to the packaging and storage conditions before purchase. Each purchase corresponding to a unique meat batch was then transported to the laboratory within approximately 30 min (seven batches in total). Each batch of ground beef was divided in fifteen samples of 70–75 g each and placed in Petri dishes, while a small sample was retained for microbiological analyses for the purpose of acquiring background information. Then, multi-spectral images of the first five samples were acquired followed by Fourier transform infra red (FTIR) spectroscopy measurements using  $\sim 3$  g portions from each Petri dish.

The remaining samples were frozen at -20 °C and stored for 7 and 32 days (5 samples/batch). Samples were then thawed for close to 5 h at 4 °C so as there was no evidence of the sample having been frozen in the surface and subsequently subjected to similar image acquisition. FTIR measurements were then conducted within the next hour.

In total, 105 multispectral images and 105 FTIR spectra were collected (7 batches  $\times$  15 Petri dishes). In Fig. 1, a schematic presentation of the data acquisition process is presented.

#### 2.2. Image acquisition and segmentation

The procedure for image acquisition and subsequent image segmentation has been described extensively in previous work (Panagou et al., 2014; Ropodi et al., 2015, 2017). Briefly, multispectral images were captured in 18 non-uniformly distributed wavelengths ranging from405 to 970 nm, including the visible (VIS) and near-infrared (NIR) spectrum using the VideometerLab instrument commercialized by "Videometer A/S" (Carstensen & Hansen, 2003). Instrument calibration was performed with: (a) a light setup procedure called "autolight" which takes into account the type of object to be recorded and in this case, the first sample of the first meat batch, and (b) a geometrical and radiometrical calibration (Folm-Hansen, 1999).

The acquired images were then segmented so that redundant information (e.g. sample background, Petri dish) can be excluded. During this image-processing stage the respective routines of the VideometerLab software (version 2.12.39) which also controls the operation of the instrument were used and canonical discriminant analysis (CDA) was employed as a two-step supervised method to divide the images into regions of interest by using a simple threshold to separate between pixels of lean tissue and other pixels. This way, image background and the Petri dish was removed from the actual sample, as well as adipose from lean tissue was separated.

### 2.3. Fourier transform infrared (FTIR) spectrometry measurements and preprocessing

Small portions (approximately 3 g) of meat were placed on the surface of a ZnSe 45° HATR (Horizontal Attenuated Total Reflectance) crystal (PIKE Technologies, Madison, Wisconsin, United States) and using FT/IR 6200 JASCO spectrometer, FTIR measurements were performed. With the Spectra Manager software version 2 (Jasco Corp.), spectra were collected from 4000 to 400 cm<sup>-1</sup> (100 scans, resolution of 4 cm<sup>-1</sup>) within a period of 2 min. A reference spectrum was acquired at regular intervals using the crystal with no added meat and the crystal's surface was cleaned after each acquisition with detergent and distilled water and then with analytical grade acetone. Lastly, it was dried with lint-free tissue. Spectra ranging approximately from 1800 to 800 cm<sup>-1</sup> were exported.

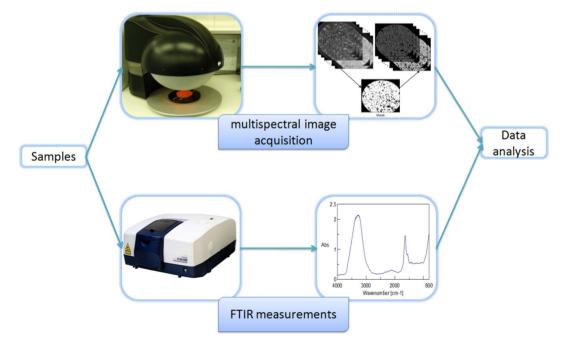


Fig. 1. Graphical presentation of the data acquisition process.

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