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Differentiation between normal and white striped turkey breasts by visible/near-infrared spectroscopy and multivariate data analysis

Abstract

The appearance of white striations over breast meat is an emerging and growing problem. The main purpose of this study was to employ the reflectance of visible-near infrared (VIS/NIR) spectroscopy to differentiate between normal and white striped turkey breasts. Accordingly, 34 turkey breast fillets were selected representing a different level of white striping defects (normal, moderate and severe). The findings of VIS/NIR were analyzed by principal component analysis (PCA). It was found that the first principal component (PC1) for VIS, NIR and VIS/NIR region explained 98%, 97%, and 96% of the total variation, respectively. PCA showed high performance to differentiate normal meat from abnormal meat (moderate and severe white striping). In conclusion, the results of this research showed that VIS/NIR spectroscopy was satisfactory to differentiate normal from severe white striping turkey fillets by using several quality traits.

Keywords: VIS/NIR spectroscopy, white striping, quality, PCA.
Introduction

In the last few decades, tremendous improvements have been achieved in growth rate and breast yield of poultry birds, to meet the growing demand for poultry meat (Mudalal et al., 2015). Globally, the productivity of poultry meat has been enhanced by intentional genetic selection using traditional quantitative techniques (Zuidhof et al., 2014). The genetic selection was accompanied by histological and biochemical modifications in the muscular tissues of growing birds (Petracci and Cavani, 2012). It was found that genetically selected birds had low blood capillary vessels’ density which led to some disorders metabolism (Soglia et al., 2018a). Accordingly, this was accompanied by emergence of several muscle abnormalities such as Pale Soft exudative (PSE) (Petracci et al., 2017), Deep pectoralis myopathy (DPM). Recently, new muscle abnormalities have been observed such as white striping (WS) and hardening of the breast muscle known as ‘wooden breast’ (Kuttappan et al., 2017). Moreover, intramuscular connective tissue defects characterized by a loose structure of muscle fiber bundles called ‘spaghetti meat’, has been recently observed (Maiorano, 2017; Baldi et al., 2018). The previous poultry meat defects were attributed due to a consequence of substantial improvement towards increasing growth rate and breast yield (Petracci et al., 2015).

All previously mentioned defects in turkey and chicken meat (in particular breast meat) are considered as a serious problems to poultry industries because they affected adversely the quality traits of premium cuts (Soglia et al., 2018b). These defects impaired visual appearance as well as reduced technological properties such as water holding capacity, texture, and color. Accordingly, this was negatively reflected on consumer acceptance (Kuttappan et al., 2012b). The classification systems for affected meat by muscle
abnormalities are still based on aesthetic criteria (variations in the color of the meat, whether the meat is too pale or too red, and/or excessive fluid accumulation), couldn’t make an exact judgment to deal with meat quality issues (Barbut, 2009). The affected meat should be culled out from the processing line and transformed them for further processed meat (such as nuggets and sausages) while the rest of the carcass is suitable for human consumption (Brambila et al., 2017).

Differences in meat composition due to increase muscle abnormalities have imposed more pressure on the meat industries to guarantee good meat quality. Concerning production and meat evaluation, there is a need to look for rapid, non-destructive and non-expensive techniques.

Over the last years, the use of near-infrared spectroscopy (NIRS) as instrumental technique with spectrum wavelength (800-2500 nm) has increased enormously. Near-infrared spectroscopy (NIR) has the ability to estimate and predict different quality traits in food products by measuring the amount of NIR radiation that is reflected, absorbed, transmitted, and/or scattered at different wavelengths (Gardner, 2018).

NIR spectroscopy technique was employed to evaluate the chemical composition of meat and meat products (Van Kempen, 2001). It has unique advantages if compared with classical methods such as quick and frequent measurements, and the ease of sample preparation. Moreover, it is fit for on-line applications in the agriculture field (Abu-Khalaf, 2015; Beghi et al., 2018), pharmaceutical industries (Guillemain et al., 2017), as well as medical sectors (Monteyne et al., 2018) to assess different quality traits. On another hand, NIRS has still some limitations where there is a necessity for reference method, low sensitivity to minor constituents, as well as complexity in the calibration (Büning-Pfaue and Hans, 2003).
The ability of NIRS to predict several quality traits of meat such as chemical composition (protein, moisture, fat, and collagen), pH, water holding capacity, etc have been investigated (Brondum et al., 2000; Meulemans et al., 2003; Moran et al., 2018; Yang et al., 2018). Moreover, it was found that there was a possibility to classify meat based on feeding regimes (Cozzolino et al., 2002), strains (McDevitt et al., 2005), and tenderness (Yancey et al., 2010) by using NIR spectroscopy.

There are no available studies that used VIS/NIR spectroscopy to predict the quality traits of turkey breast meat affected by different levels of white striping. Therefore, the main objective of this research is to employ VIS/NIR spectroscopy to differentiate different levels of white striping defects.

Materials and Methods

Samples Selection and Preparation

From a local Palestinian slaughterhouse near Tulkarm city (Palestine), more than 60 the pectoralis major muscles of 20-wk old tom turkey birds were randomly selected based on the appearance of white striations. The evaluation of the presence of white striping was performed on the processing line at 1-2 h of post-mortem after the breast-deboning area. Out of 60 pectoralis major muscles, 34 muscles were classified into three groups: normal (12 samples) (free of white striations), moderate (12 samples) (when white striations thickness <1mm), and severe (10 samples) (when white striations thickness >1 mm) (Soglia et al., 2018 b). Samples were subjectively pre-classified into categories, packed on ice, and transported to Palestine Technical University- Kadoori laboratory for VIS/NIR
measurements then to An-Najah National University laboratories for other quality traits analysis. The *pectoralis major* muscles were excised from the whole breast muscle. Excessive fat, connective tissue, cartilage and bone fragments were avoided to minimize sampling errors.

**VIS/NIR Spectroscopy Measurements**

In each turkey breast meat sample (*n* = 34), three spectra were collected (at a room with a temperature of 23± 2°C and relative humidity of 60%) directly on the skin side, radial section, and tangential section. A USB2000+ miniature fiber optic spectrometer (Ocean Optics, USA) with a vivo light source and 50 µm fiber optics probe was used for spectra acquisition. The spectra were obtained at scans with a resolution of 0.35 nm full width at half maximum (FWHM), and spectra range 550-1100 nm. It also has a 2048-element CCD-array detector, 2-MHz analog-to-digital (A/D) converter, in addition to a high-speed USB 2.0 port. The USB2000+ can be controlled by Spectra Suite software. This device is equipped with an active fan cooling to overcome the risk of sample overheating. The 4 halogen tungsten light sources make the vivo a high-powered VIS/NIR source, which allows a shorter integration time than conventional methods (Ocean Optics, USA). The integration time used in this investigation was 1340 µs. A total of 102 spectra were obtained for turkey samples, and then the average spectra were taken. The VIS/NIR analyses were performed in the diffuse reflectance mode and then recorded as absorbance (log 1/R). To ensure the stability of the measurements, a diffuse reflectance standard WS-1 (Ocean Optics, USA) was used as the optical reference standard for the system every 5 minutes during the experiment. The dark reference was done once at the beginning of each experiment, by closing the entrance of incoming light from probe to
the USB2000+ miniature fiber optic spectroscopy using a plastic cap. At the end of all spectral measurements, the acquired data were well stored for later analysis. 34 samples were used for building PLS calibration models using their VIS/NIR spectra.

**Statistical Analysis**

The Unscrambler program (version 9.7, CAMO Software AS, Oslo, Norway) was used for both PCA and PLS multivariate data analysis (Abu-Khalaf, 2015). In PCA, VIS/NIR spectra represented a bilinear model of the data matrix X. PCs represent in a pattern of observations in plots.

To investigate the possible differences in three types of turkey breast meat (normal, moderate and severe) at three ranges, i.e. VIS (550-700 nm), NIR (700-1100 nm) and VIS/NIR (550-1100) wavelengths, a PCA model was carried out.

**Results and Discussion**

Typical mean spectral curves representing the three levels of white striping fillets in the wavelength range 550–1100 nm are shown in Fig. 1. The depressions and peaks in spectra showed the strong and weak absorbance characteristics of the samples, within the range of study. The spectra of normal, moderate (WS) and severe (WS) breast fillets showed similar absorption bands, which were in agreement with previous studies (Barbut, 1996; Fumiere et al., 2000).

NIR spectra often contain undesired scattering variation due to heterogeneous content and sample surface amongst others. The scattering effect in NIR consists of a multiplicative effect and an additive effect. The additive effect is reflected as the baseline offset. The
multiplicative effect is reflected as a slope that scales the entire spectrum. Data pre-treatment was employed to minimize these complex baseline variations and scattering effects. NIR spectra of the samples set were pre-processed using standard normal variate (SNV) to delete slope variation and to correct for scattering effects (Fig. 2). Light scattering in fresh meat samples does not always travel the same distance before it is detected. As a longer light traveling path corresponds to a lower relative reflectance value while more light is absorbed (Jens et al., 2019). This causes a parallel translation of the spectra. For that reason, multiplicative scatter correction (MSC) was used to eliminate these effects (Li and He, 2006) (Fig. 3). Savitzky–Golay first derivatives ($1^{st}$ D) was done to delete baseline flung in meat spectral data and small spectral differences were strengthened, this followed by Savitzky–Golay smoothing (Cunha et al., 2010) to prevent increasing the noise, which came from the derivative (Fig. 4) (Li and He, 2006). The relative values of spectra may vary from sample to sample, which might be due to changes in surface texture and moisture content of three types of fillets (Soglia et al., 2018 a,b; Mudalal, 2019)

Six bands (peaks at 550, 574, 580, 600, 630 and 643 nm) in the visible region (550- 700 nm) and eight bands in the NIR region have been observed (Fig. 4). Several researchers found similar bands and spectral features (Barlocco et al., 2006; Andrés et al., 2008; De Marchi et al., 2012). Absorption bands at 550 to 580 nm were associated to the Soret band attributed to the traces of erythrocytes of myoglobin with both haemoglobin and oxyhaemoglobin absorption (Stryer and Latchman, 1995) as well as to oxymyoglobin (Liu and Chen, 2000).

Our findings showed that severe and moderate white striping fillets had higher absorption at 550, 574 and 580 nm than normal fillets. This result may be attributed due to discoloration over the surface of white striped fillets (Ellekjaer and Isaksson, 1992). The mean spectrum in
the NIR region has absorption bands at 980 nm and it could be related to the second overtone of the OH- vibrational mode of water (Bowker et al., 2014). The absorption at 760 and 908 nm corresponds to the deoxhaemoglobin (Hollo et al., 1986) and the third overtones of C-H bonds (Weyer and Workman, 2007), respectively. The identified band at 552 nm related to myoglobin (Cozzolino et al., 1996). Absorption band at 574 nm was associated with oxyhemoglobin (Mitsumoto et al., 1991). The absorption bands at 540 and 580 nm were associated with both myoglobin and oxymyoglobin, respectively (Cozzolino and Murray 2004).

PCA has been carried for VIS/NIR regions spectrum considering the three levels of muscle abnormalities (normal, moderate and severe). PCA showed an ability to distinguish the three groups (normal, moderate WS and severe WS) from each other (Fig. 5-7).

In this analysis, 2PCs for VIS, NIR and VIS/NIR region explained 99%, 100% and 98% of the variance, respectively. Our findings showed that PCA had high performance in separating normal turkey breast meat from abnormal meat (moderate and severe). These results were in agreement with previous studies where VIS/NIR spectroscopy with PCA was used to separate poultry and meat products into different categories (Wold et al., 2017).

In conclusion, visible-near infrared (VIS/NIR) spectroscopy is considered a quick, safe and nondestructive technique which is very suitable for online control for meat defects. The findings of this study showed that VIS/NIR spectroscopy was satisfactory to differentiate normal from severe WS turkey fillets. Moreover, the results open a wide door for using a portable VIS/NIR technique in the turkey industry. Further studies with a high number for samples are recommended to confirm the ability of VIS/NIR combined MVDA techniques to differentiate normal turkey breast meat samples from defect WS.
Conflicts of Interest

The authors declare that there is no conflict of interest regarding the publication of this paper.

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Fig. 1. A typical VIS/NIR (500-1100 nm) spectral curve obtained from turkey fillets. Normal fillets (red), moderate WS fillets (blue) and severe WS (green) fillets, without pre-processing.

Fig. 2. The effect of Standard Normal Variate (SNV) preprocessing on the spectra obtained from turkey fillets. Normal fillets (red), moderate WS fillets (blue) and severe WS (green) fillets.
**Fig. 3.** Effect of Multiplicative Scatter Correction (MSC) preprocessing on the spectra that obtained from turkey fillets. Normal fillets (red), moderate WS fillets (blue) and severe WS (green) fillets.

**Fig. 4.** The effect of the first derivative and smoothing preprocessing on the spectra obtained from turkey fillets with different absorption bands. Normal fillets (red), moderate WS fillets (blue) and severe WS (green) fillets.
Fig. 5. Score plot of PCA model based on VIS (550-700 nm) spectra of turkey fillets. Normal fillets (N), moderate WS fillets (M) and severe WS (S) fillets. Two PCs explained 99% of the data variation.

Fig. 6. Score plot of PCA model based on NIR (700-1100 nm) spectra of turkey fillets. Normal fillets (N), moderate WS fillets (M) and severe WS (S) fillets. Two PCs explained 100% of the data variation.
Fig. 7. Score plot of PCA model based on VIS/NIR (550-1100 nm) spectra of turkey fillets. Normal fillets (N), moderate WS fillets (M) and severe WS (S) fillets. Two PCs explained 98% of the data variation.