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Abstract

Aged beef is more tender and flavourful than beef that has not undergone the aging process. The characteristics of raw meats are influenced by a variety of factors, e.g. animal, environmental and processing. During the ageing, beef muscles undergo several changes that can affect their quality, as colour, tenderness, flavour and juiciness. Since the ageing process provides high added values that manifests in price, a quick, non-contact measurement method would be useful in industry to check ageing state.

The objective of this study was to estimate the ageing time of beef sirloin samples by their reflected NIR spectra and hyperspectral datacube as well. The hyperspectral signal-to-noise ratio had to be improved. Controlling software was developed to help proper calibration, data-acquisition and pre-processing. During 4-week ageing experiments, samples were measured by both spectral method. The ageing time and also the location of slices were registered, for making difference between the two influences. The suitability of two methods, the more accurate instrumental- and the non-contact hyperspectral measurment was proved.

Key words: spectral, hyperspectral, NIR, meat, beef.

1. Introduction

Aged beef is more tender and flavourful than beef that has not undergone the aging process. The characteristics of raw meats are influenced by a variety of factors (Yongliang et al., 2003), e.g. animal (breed, sex, age), environmental (feeding, transporting and slaughtering condition), and processing (storing time, temperature condition). During the ageing, beef muscles undergo several changes that can affect their quality. These changes are reflected in many characteristics such as colour, tenderness, flavour and juiciness.

Generally, beef is aged commercially with tightly controlled and monitored temperatures and humidity levels. Aged beef has increased tenderness because the muscle and collagen in the meat begins to change due to the work of certain enzymes. Maximum tenderness can be reached at around 11 days after the cow is slaughtered. Beef can either be dry aged or wet aged. Ageing is usually preferred because moisture levels are reduced to give the piece of meat a "beefier" taste and increased tenderness.

In the alternative, wet aged beef is aged in a vacuum packed and sealed bag. It is a quicker method of ageing and it keeps the moisture level of the meat high. Since the weight of the beef is retained, it is more cost effective than dry ageing (http://www.wisegeek.com/what-is-aged-beef.htm).

Since the ageing process provides high added values that manifests in its price, a quick, non-contact measurement method would be useful in industry to check ageing state.

Near infrared spectroscopy is a quick but contact way to assess the major components and other derived properties, like the chemical changes of aged beef. Over the years, this method has been developed and applied considerably in quality management of meat products. Usually fat, water and protein contents in industrial scale meat batches were determined on-line by NIR reflectance spectroscopy. E.g. the quantitative analysis of pork meat was developed by using partial least squares (PLS) regression (Tøgersen 1999). The correlation coefficients (R^2) of the calibration models of fat, protein and water contents based on integrating sphere diffuse reflectance were 0.902, 0.919 and 0.901, respectively. Several applications proved, that NIR is a promising technique for large-scale meat quality evaluation.

Prieto et al. (2009) concluded that NIR was much better at predicting meat composition (fat, protein and water) than meat quality (marbling grade, tenderness etc.). Other derived properties, like the chemical changes of aged beef might be also detected by spectral measurement. However, because of the non-homogeneous structure of beef, this method needs to grind, destruct the samples before measurement.

Conventional imaging (image processing and analysis) is a non-contact way to obtain the optical properties of non-homogeneous surfaces.

The marbling structure of meat were studied in several articles. Grayscale images were segmented by neural network (Shiranita et al, 2000), saturation value of coverted RGB images were classified by ANOVA (Li et al., 2001), image compressing algorithm was used to describe marbling structure (Taraichi et al, 2002), different unsupervised methods (statistical and neural network) were compared (Tan, 2004), formaldehyde curin were used to enhance pattern (Faucitano, 2005), thresholding and clustering were combined to improve segmentation (Jackman et al.,, 2009), edge detecting algorithm was applied to detect fiber structure of marbling (Hussein et al., 2011).

Hyperspectral imaging (HSI) offers a remote sensing way for measuring the whole spectra of each pixels of the surface. This method combines the advantages of spectroscopy and image processing. The marbling structure of meat was also studied using spectral information. Hyperspectral data was analized for segmentation in visible range (Quiao et al, 2007), statistical methods (MLR and PLSR) were built to predict marbling (Jackman & Sun, 2009), the significant wavelengthes were determined in NIR range for classification (Barbin et al., 2011) and for other properties, like water-holding capacity (Elmasry et al., 2011). Non-statistical segmentation algorithms were also developed for improving result (Liu, 2011).

The tenderness of 14-day aged, cooked beef was also predicted using a visible hyperspectral imaging system (Naganathan et al., 2007) in the range of 400–1000nm) with a diffuse-flood lighting system. The prediction of three beef tenderness categories (tender, intermediate and tough) succeeded with a 96.4% accuracy.

2. Objective of the study

The objective of this study was to estimate the ageing time of beef sirloin samples by their reflected NIR spectra and hyperspectral datacube as well.

First the hyperspectral signal-to-noise ratio had to be improved. Software was developed to control proper AD-, spectral- and spatial calibration, data-acquisition and pre-processing.

The reproducibility of meat's spectral measurement was checked by multiple measuring of fresh-cut surfaces, and the outer surfaces of samples stored in different conditions (O_2- and vacuum-packed). The effects of open air and temperature were examined.

For predicting the ageing time, during 4-week ageing experiments, samples were measured by both spectral method. The ageing time and also the location of slices were registered, for making difference between the two influences.

Hyperspectral images were segmented by ENVI algorithm retrieving the average spectra of pure meat. Finally statistical models were calibrated on the instrumental and also the hyperspectral dataset to predict ageing time and location.

The suitability of two methods, the more accurate instrumental- and the non-contact hyperspectral measurement, had to be proved.

3. Materials and methods

The spectra of the samples were taken by use of **MetriNIR spectrometer**. This instrument works in the range of 740-1700 nm with 2 nm spectral resolution.

For hyperspectral data acquisition a **HeadWall Hyperspectral Imaging System** was used. This system works in the range of 900-1700 nm with 5 nm spectral resolution. The Indium-Gallium-Arsenide (InGaAs) sensor matrix had 256 px in spectral and 320 px in spatial and 14-bit dynamical resolution. The altitude of sensor and the lens resulted in 0.46 mm spatial resolution.

Software environment (ARGUS, 2011) of push-broom hyperspectral system was developed to control sensors and Y-table stepping motors. It supports the setting of optimal AD parameters, two-point spectral calibration, defining spectral- and spatial ROI, controls the table and acquires frames as to avoid spatial distortion in the hypercube. The hypercube retrieved is saved in ENVI file format for later image processing. A screenshot shows a form of software (Fig. 1a)

- The first panel of the controlling environment helps to set optimal AD parameters of the sensor. In case of proper parameters, the histogram of signal shows a wide distribution within the range.
- The middle one helps spectral and spatial calibration, setting the spectral and spatial region of interest. The pure signal and the relative or absolute reflectance can be displayed or saved. After setting the Y length of object, a measurement can be started moving the table and grabbing the frames synchronously.
- On the third panel, a hypercube is displayed. A channel or a pseudo-image of three channels or the linear combination of three channels can be shown. As the sample of a leaf shows, the linear combination of bands (e.g. 1250nm - 1500nm) can be very useful to enhance a property (Fig. 1b).

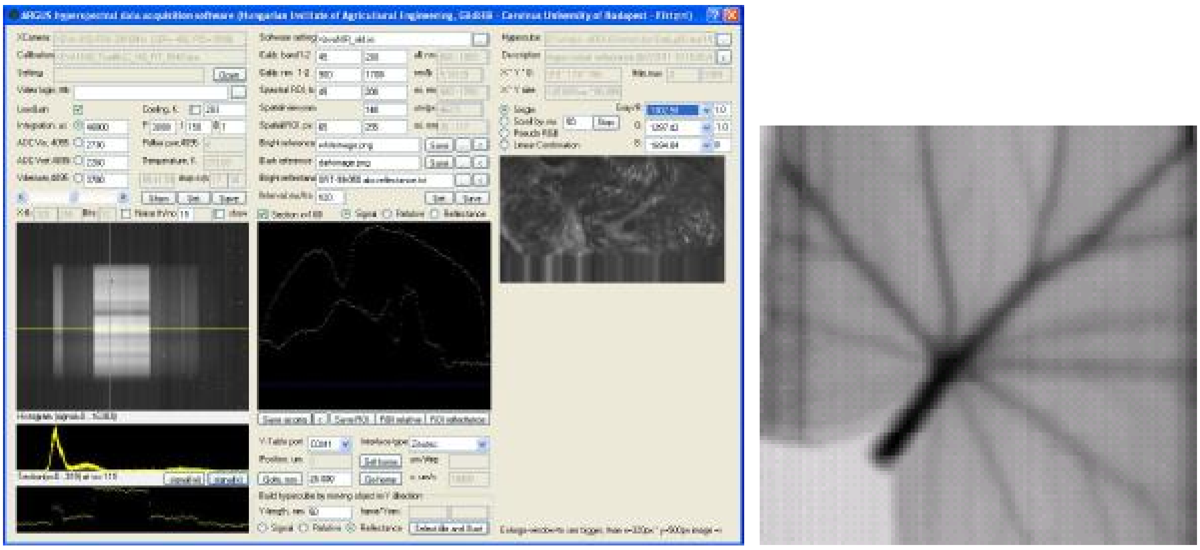

FIGURE 1: Screenshot of Argus software (a). Linear combination of bands for a leaf (b)

In case of the **stability experiment** 8-8 independent beef sirloin samples were measured by each following three measurements:

- **fresh-sliced surface** of vacuum-packed beef sirloin samples
- outer surface of **vacuum-packed** samples (3 days storing) after opening the package
- outer surface of **oxygen-packed** samples **(**3 days, 100% O_2**)** after opening the package

The samples were left on open air during the 7 times 10 minutes of the measurements, each samples were inspected in every 10 minutes. A smaller part of a slice was measured by spectrometer, and the other bigger one by hyperspectral system every time.

In case of the **ageing experiments**, whole beef sirloins (length approx. 60 cm) were sliced into 20 equal slices. These slices were aged vacuum-packed for 4 weeks at 5 °C in stabile air-conditioned storage room. Each week 4-4 slices were inspected from fore rib to the direction of the rump (no. 1, 6, 11, 16 at first, etc.), and NIR spectra and hyperspectral data were recorded (Fig. 2).

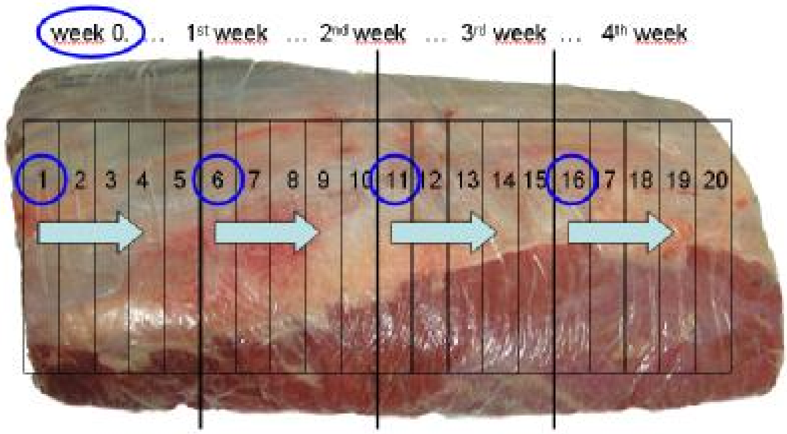

FIGURE 2: The location of the inspected beef sirloin samples during ageing

The reflected hyperspectral data were pre-processed by **ENVI** (Environment for Visualizing Images) algorithm. A supervised classification method (Spectral Angle Mapper, SAM) was used for segmentation of the areas of the meat and the fat tissues. SAM is a physically-based spectral classification that uses an n-dimensional angle to match pixels to reference spectra. The algorithm determines the spectral similarity between two spectra by calculating the angle between the spectra, treating them as vectors in a space with dimensionality equal to the number of bands. After segmentation, the average reflectance spectra of pure meat were transformed to absorption spectra (relative to gold standard and dark-current measurement).

Finally the recorded absorption spectra of the **stability** and the **ageing experiments** were processed by **R Project** software building **PLS** regression models. The spectrum (reflection on different wavelengths) as independent space was mapped (calibrated) into the ageing time or location as dependent variable's space. Cross-validation was used and the number of factors was determined on the base of the minimum of RMSEP (Root Mean Square Error of Prediction). The efficiency of PLS regression models can be characterized by their determinants.

4. Results

According to the **stability experiment**:

- In case of fresh cut surface all absorption spectrum represented the same slowly decreasing (brightening) characteristic by the 7 times 10-minute measurements. The two significant wavelengths (at 1190 and 1450 nm) referred to the moisture content, where the absorption got lower gradually. The average intensity of spectra (integral) represented the same slow-decreasing characteristic.
- The average absorption on the outer surfaces of O_2-packed samples were increasing (darkening) after unpacking, but after 20 minutes remained stable.
- The average absorption on the outer surfaces of vacuum-packed samples were increasing (darkening) after unpacking, but after 10 minutes, it showed a slow-decrease.

To sum up:

- In case of fresh-cut measurement, the changing was rather slow, so the spectral measurement can be done as soon as the temperature is getting to be stable.
- In case of measuring packed samples, it is worth to inspect through the see-through foil, or after unpacking, the measurement must be done as soon as possible.

In case of the **ageing experiment**, the intensity values were normalized further to focus only to the spectral properties. Two significant ranges were found, where the change of spectra was monotonous while the other parts remained the same. The first range was between 740 and 1040 nm, while the second one (1400 - 1500 nm) might refer to the moisture content since one of its significant absorption peaks is 1450 nm. The average intensity showed slow decreasing during the 4-week ageing time.
The absorption spectrum of the samples depends on the location from fore rib to rump. The PLS models resulted good correlation on instrumental data (factors=17, R^2=0.9762) and also on hyperspectral data (factors=19, R^2=0.9166) for predicting the location (Fig. 3).
Since the location was mostly effected on the average intensity of absorption spectra, and the ageing time effected on the spectral changes, for prediction of the ageing time, henceforth we normalized all spectra.

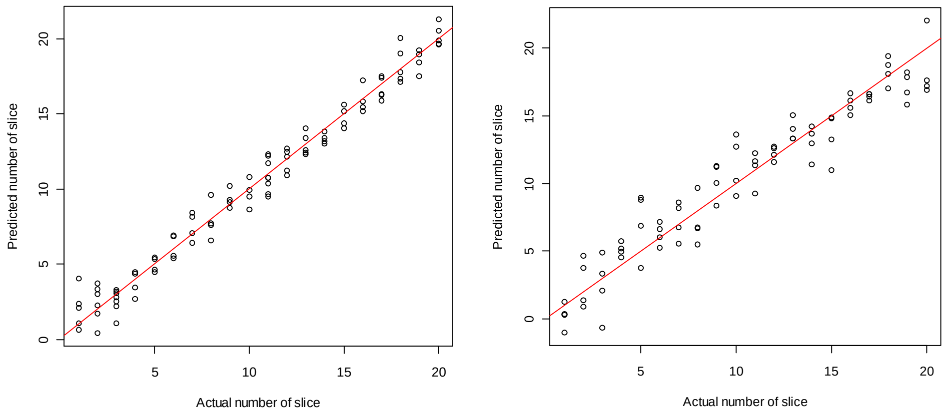

FIGURE 3: Influence of location on the base of spectral and hyperspectral data

After normalization the PLS prediction model of ageing time also resulted good correlation on the base of instrumental- (factors=17, R^2=0.9803) and hyperspectral (factors=15, R^2=0.9857) measurements as well (Fig. 4).
Even though the hyperspectral acquisition has generally much more noise, these reflected spectra were also enough to calibrate PLS regression models for ageing time prediction.

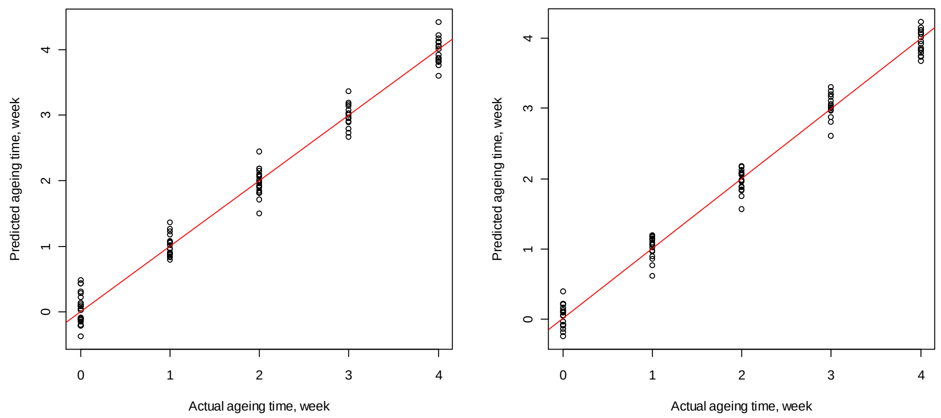

FIGURE 4: Prediction of aging time on the base of spectral and hyperspectral data

5. Conclusion

Conventional spectroscopy and hyperspectral imaging can both efficiently determine the ageing time of beef sirloin.

The hyperspectral method can be further improved by avoiding the effect of uneven surface with use of diffuse illumination (instead of line), measuring the samples under glass.

On the base of hyperspectral measurement, classification and PLS regression model, the significant wavelengths of an inspected property can be calculated, and those might be used by a non-contact multispectral measurement in an industrial application.

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