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Study on the Mutton Freshness Using Multivariate Analysis Based on Texture Characteristics

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Abstract. Aiming at discrimination and prediction of mutton freshness by texture profile, the texture parameters of mutton stored at 1°C, 4°C and Room temperature were analyzed. The analysis methods of Canonical Discriminant Analysis (CDA) and Principal component analysis (PCA) were used to analyze texture parameters of mutton. The results of PCA showed that mutton sample stored at three temperatures clustered into groups according to their freshness, changing along the direction of PC1. Better classification results were found by CDA. The changing trends of mutton freshness were described by Multiple Linear Regression (MLR) and Partial Least Square analysis (PLS), and effective predictive models were found for indices of days stored, TVB-N and pH using texture parameters. With optimum analysis methods, texture parameters could classify and predict freshness of mutton stored at three temperatures. Texture profiles were proved to be a fast and objective tool for the prediction of mutton freshness.

Keywords: Mutton, Freshness, Texture parameters, multivariate analysis

1 Introduction

It is well known that a country's meat consumption and demands for healthier diets increase reflect its degree of development. Trends for healthier meat could result in increasing demands for meat with high protein, low fat and good taste, such as mutton, which could lead to a diet with better nutritional (lower fat and cholesterol) [1,2] and sensorial features (flavor, juiciness, tenderness). The consumption of mutton has a significant increase in China in recent years. However, the shelf life of mutton sold as whole carcasses or as bone-in cubes at retail store is subject to variation of storage temperatures, contamination of microorganisms, enzymes and the processing conditions, which damage the functional, sensorial and nutritional quality of mutton[3]. It is of great importance to monitor the changes of mutton freshness during storage.

Several techniques are used to monitor the deterioration of meat, including the use of microbiological techniques, detection of metabolite concentrations, spectroscopy methods, electronic tongue and electronic nose, and sensorial panels. The microbiological techniques were used to evaluate consequences of bacteria-bacteria interactions [4, 5], monitor the spoilage and freshness of meat, and study on the initial microflora and total number of bacteria changes stored at different temperatures [6]. For spectroscopy methods, near infrared reflectance spectroscopy (NIR) [7], fourier transform near infrared spectroscopy [8] and fluorescence spectroscopy [9] were shown to be effective for meat freshness assessment. The metabolic concentrations of protein, lipids, fat acid, carbohydrates, such as ATP [9], glucose and derived compounds or biogenic amines [10], detected by spectroscopy methods and physicochemical analysis, were used to express meat freshness. The electronic tongue and electronic nose showed their ability in freshness evaluation of pork [11, 12] and beef [13]. The senses of taste, smell, touch, sight and hearing were used to describe food properties by sensorial panels. The juiciness, tenderness, flavor and color of meat were determined by sensorial panels [14, 15]. However, there were few works done on the texture changes along with freshness of meat.

For the physical properties, the texture profiles, as combination of sensory perception, mechanical properties and geometric features, could directly describe the organization status of food texture characteristics in the form of numerical description. Texture Profile Analysis (TPA) [16] is a very useful method for texture analysis. The texture parameters of hardness, springiness, chewiness and cohesiveness were obtained by TPA, and these parameters were used to describe the eating quality of food and freshness assessment of fruits, fishes and meat. The relationships between TPA parameters of meat and sensory characteristics were studied aiming at finding the correlation of texture parameters and sensory analysis [17, 18]. TPA was used to monitor food quality and control of food quality, food shelf life [19] and consumer preference studies, modification formulation and processing process [20].

The studies of meat texture were mostly on the differentiation and substitution of sensory perception of raw meat and processed meat products. However, little detailed information is available on quantitative determination of physicochemical parameters of meat freshness, and correlation of texture parameters with physicochemical indexes [21, 22], except for the relationship between freshness and elasticity of fish [23] and chicken [24].

The potential use of Texture Profile Analysis for detection of mutton freshness was conducted by texture analyzer in this study. The objectives were: (1) investigation the use of a texture analyzer to monitor the freshness of mutton stored at temperatures of 1°C, 4°C and room temperature, with the help of multivariate analysis methods, (2) build models for the prediction indications of mutton freshness, (3) develop an objective method for detection of mutton freshness.

2 Experiments and Methods

2.1 Experimental Samples

All the meat samples used were obtained from local market. For pretreatment, the fat and connective tissues were removed before detection, and the mutton samples were cut into cubes of 20 mm ×20 mm×15 mm. Cuboids of mutton were stored at 1 °C, 4 °C and room temperature of 20 ±2 °C, respectively. The samples were brought to room temperature before analyzed by TPA and physicochemical methods for freshness indications. Samples detected at the day they were taken to the lab were named d0, samples which were stored for 1 day were named d1, samples which were stored for 2 day were named d2, and so on d3, d4, d5, d6, d7, d8, d9, d10, and d11.

2.2 Experimental Methods

The texture analyzer of TMS-Pro (Food Technology Corporation, USA) , equipped with a 1000 N load cell, was used for texture measurements. The software of V1.13-002 Texture Lab Pro (Food Technology Corporation, Virginia,USA) was used to acquire and analyze data. TPA analysis was conducted according to the method of Tian [19] with some modifications. All the measurements were carried out with 2 compression cycle test, with cylinder probe of 15 mm diameter, 40% compression of the original sample height. 5 s was left between the two compression cycles. The force-time deformation curves of mutton sample were gained with 1 N trigger force at speed of 60 mm min⁻¹. The hardness, chewiness, cohesiveness, springiness and gumminess of mutton were calculated from the force-displacement curve for each sample. Eight replicates were measured for TPA parameters of mutton for each treatment.

For tenderness, a digital muscle tenderness meter (C - LM3, Tenovo international co., Limited, China) was used to measure the tenderness of treated mutton samples. Tenderness measurement was evaluated with three replicates by cutting through the sample perpendicular to fibre direction, and expressed in form of shear force. Shear force was calculated as the average shear force of three samples.

As freshness indicator of mutton, content of total volatile basic nitrogen (TVB-N) was detected according to the method of semimicro-Kjeldahl determination in GB/T 5009.44[25] and pH was detected according to the method in GB/T 9695.5[26]. 3 duplicates were conducted for each sample, and the average of 3 duplicates were used.

For data processing, the texture characteristic parameters of mutton were analyzed by multivariate data analysis methods to discriminate the samples according to the freshness of mutton. Principal Component Analysis and Canonical Discriminant Analysis were used for visualization of mutton with different freshness using texture parameters. The changing trends of mutton freshness were described by Multiple Linear Regression and Partial Least Square analysis, and the predictive results were compared. For data processing, the software program of the SAS (V8, SAS Institute Inc., Gary, USA) was used.

3 Results and Discussion

3.1 Changes of mutton freshness

The deterioration of mutton samples was monitored by TVB-N, which is the typical indicator of meat freshness. pH of the sample was detected as a reference indicator for mutton freshness. The changing trends of TVB-N and pH of mutton samples stored at 1 °C, 4 °C and room temperature were shown in Fig.1 and Fig.2. The value of TVB-N exceeded the limit level of 25 mg/100g at the 2nd day for samples stored at room temperature, the 8th day for samples stored at 4 °C and 1 °C. The pH exceeded over the limit level of 6.7 at the 2nd day for samples stored at room temperature, the 7th day for samples stored at 4 °C, and the 8th day for samples stored at 1 °C. The TVB-N and pH of mutton stored at three different temperatures increased with storage time. Faster changes were observed in higher storage temperature, which is in accordance with the changes of meat freshness by Nan [27].

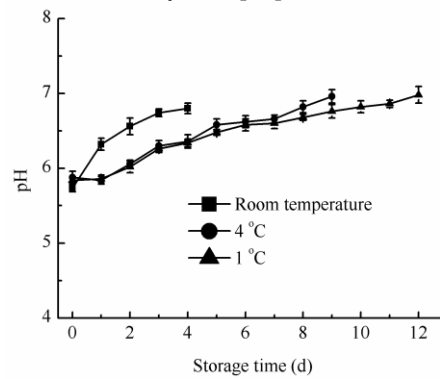


Fig. 1. Trend for pH versus storage time (day) at three temperatures

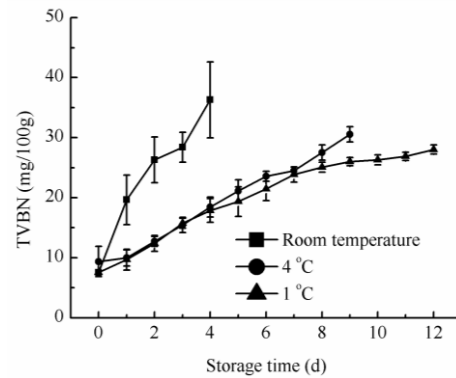


Fig. 2. Trend for TVB-N versus storage time (day) at three temperatures

3.2 Texture characteristic parameters of mutton changed with freshness

The texture of mutton changed according to the deterioration of mutton when stored, and the changes of texture parameters and shear force were presented in Fig.3 and Fig.4.

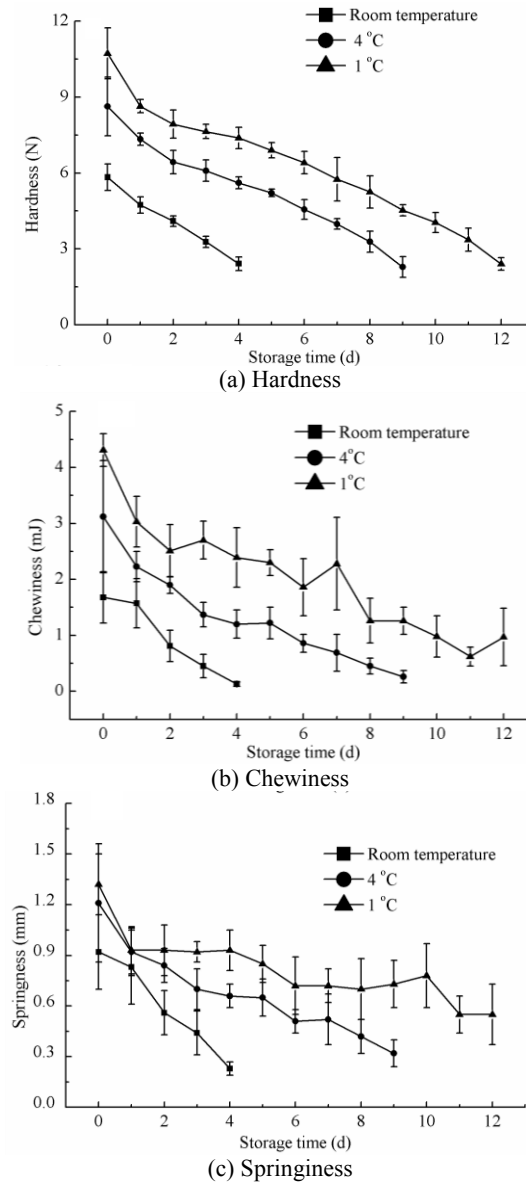


Fig. 3. Trend for texture profiles of mutton versus storage time (days) for three storage temperatures

Of all the variables measured, the values of hardness, chewiness, springiness all decreased over days at three storage temperatures. The shear force increased as the

freshness of mutton decreased, indicating the tenderness decreased with the increasing of days stored. The higher the storage temperature was, the faster these parameters changed. Same decreasing trends of hardness, chewiness and springiness to storage time were observed for grass carp [28] and chicken [19].

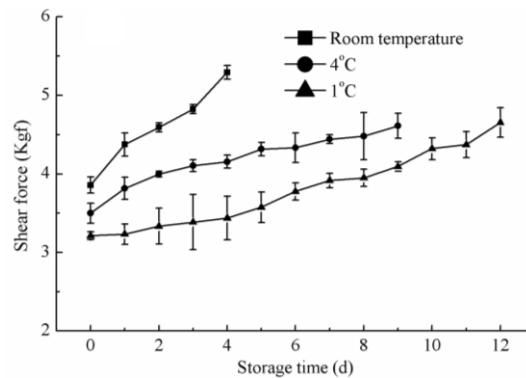
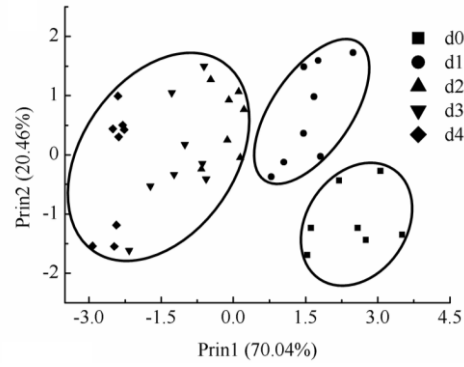


Fig. 4. Trend for shear force of mutton versus storage time (days) at three storage temperatures

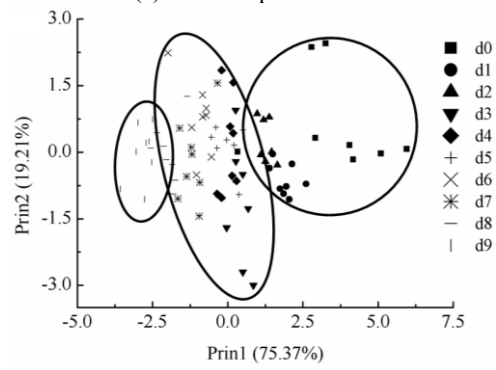
3.3 Discrimination of mutton freshness by PCA and CDA

For discrimination of mutton freshness changed along storage, PCA and CDA were used to cluster mutton samples. Hardness, chewiness, springiness and cohesiveness, obtained by texture analysis, were used as independent variables to perform the PCA and CDA analysis by SAS V8. The results of PCA for mutton samples stored at three different storage temperatures were shown in Fig.5. PC1 and PC2 accumulated 90.50%, 94.58% and 92.14% of the total variance for mutton stored at room temperature, 4°C and 1°C, respectively. As shown in Fig.5a, with PCA method, mutton samples stored at room temperature clustered into groups according to the days stored, although samples of d2 overlapped with that of d3. The freshness of mutton was clearly discriminated into three groups, with d0 as fresh group, d1 as secondary fresh group, d2, d3 and d4 as deteriorated meat, which were same with freshness level indicated by TVB-N. As shown in Fig.5b, with PCA method, the mutton samples stored at 4°C also clustered into three groups according to the freshness of mutton, with samples of d0-d2 in the fresh group, samples of d3-d7 in the secondary fresh group, and samples of d8-d9 into the group of deteriorated meat, although samples of two adjacent days overlapped with each other. The PCA results of mutton samples stored at 1°C were shown in Fig.5c, an obvious deteriorating trend was found for clusters of mutton with different freshness, although samples stored at 1°C overlapped for the adjacent 2 to 3 days.

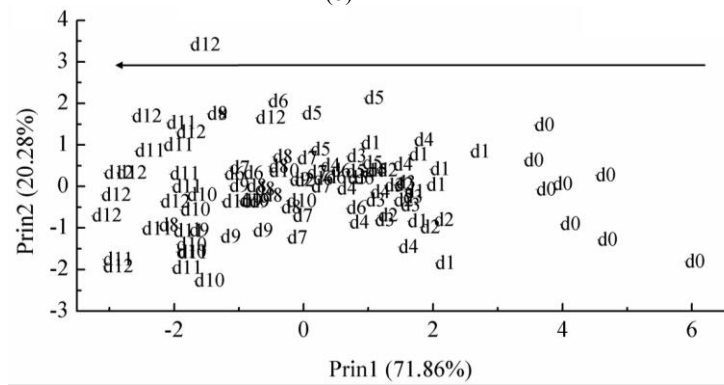
All the mutton samples were grouped into distinct clusters according to their freshness by the method of PCA, except that some samples of two adjacent days overlapped with each other. What's more, in the decreasing direction of PC1, an obvious deteriorating trend was found for clusters of mutton with different freshness for three storage temperatures. Mutton with different freshness could be discriminated by PCA.



(a) room temperature



(b) 4°C



(c) 1°C

Fig. 5. Qualitative identification of mutton freshness by PCA based on the texture profiles for three storage temperatures

The results of CDA for mutton samples stored at three different storage temperatures were shown in Fig.6. The first two CANs explained 99.18%, 95.94% and 97.77% of the total variance of 100% for mutton stored at room temperature, 4 °C and 1 °C, respectively. As shown in Fig.6a, samples stored at room temperature were grouped into 5 clusters according to the days stored, with d0 as the fresh grade, d1 as the secondary freshness, d2, d3 and d4 as the deteriorated meat. In Fig.6b and Fig.6c, similar results were obtained. Mutton samples were grouped into 3 groups according to the freshness indicated by TVB-N value, except that few samples located into the wrong groups. With CDA, better discrimination results were found. In the descending direction of CAN1, a decreasing trend was found for freshness of mutton for three storage temperatures.

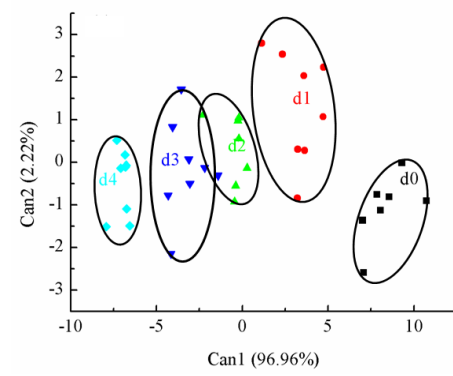
Mutton samples stored at three storage temperatures were classified according to their freshness, indicated by TVB-N, by methods of PCA and CDA, and better classification results were found by CDA.

3.4 Prediction of mutton freshness using texture characteristic parameters

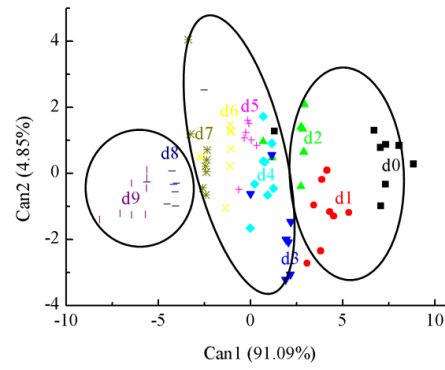
Aiming at studying the relationship between texture parameters and freshness of mutton, the method of multiple linear regressions (MLR) and partial least square regression (PLS) were used, with texture parameters as input data. The predictive results were compared to find better prediction model. The correlation coefficient of R^2 and root mean square error of RMSE were used to estimate the performance of predictive models. Lower RMSE and larger R^2 always lead to better calibration models.

For data sets containing mutton samples, 72 (6×12) samples for calibration and 24 (2×12) for validation for mutton stored at 1 °C, 54 (6×9) for calibration and 18 (2×9) for validation for mutton stored at 4 °C, 24 (6×4) for calibration and 8 (2×4) for validation for mutton stored at room temperature, were used to build the prediction model for mutton freshness, and the predictive model were validated.

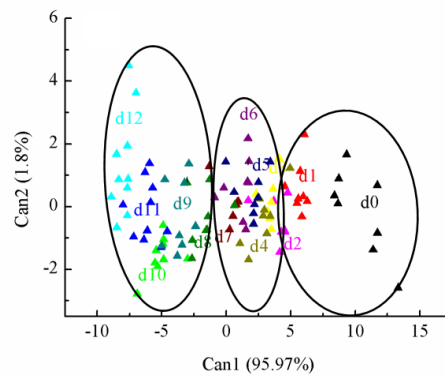
With texture parameters as independent variables, and days stored, TVB-N and pH as dependent variables, the multivariate projection method of PLS was conducted with leave-one-out technique. The accuracy was estimated using R^2 and RMSE. As shown in Table 1, good correlations for calibration data set were found between texture parameters and freshness indicators with $R^2 \geq 0.890$ for TVB-N, $R^2 \geq 0.891$ for pH, $R^2 \geq 0.941$ for days stored at three storage temperatures. The values of R^2 increased with the rising of storage temperature from 1 °C to 4 °C and then to room temperature. When these models were used to predict mutton freshness for samples in the test set, the lowest R^2 for pH, TVB-N and days stored (0.821, 0.772 and 0.831) were found for samples stored at 4 °C. In the former studies, chemical parameters were successfully predicted by method of PLS[29,30], When the PLS method was used with texture characteristics, similar results were found that the freshness of mutton stored at different temperatures could be obtained by PLS using texture parameters.



(a) room temperature



(b) 4°C



(c) 1°C

Fig.6. Qualitative identification of mutton freshness by CDA based on the texture profiles for three storage temperatures

Table 1. Comparison of two predictive methods (PLS and MLR) for TVB-N, pH and days stored for mutton stored at three storage temperatures

Methods	Storage temperature	Parameters	Training set		Test set	
			R ²	RMSEC	R ²	RMSEP
PLS	Room temperature	TVB-N	0.953	2.05	0.977	2.56
		pH	0.926	0.1	0.913	0.15
		Days stored	0.968	0.25	0.94	0.43
	4 °C	TVB-N	0.93	1.85	0.821	2.91
		pH	0.912	0.11	0.772	0.17
		Days stored	0.949	0.66	0.831	1.16
	1 °C	TVB-N	0.89	2.22	0.87	2.36
		pH	0.891	0.12	0.853	0.14
		Days stored	0.941	0.94	0.923	1.03
	Room temperature	TVB-N	0.949	1.96	0.972	2.39
		pH	0.917	0.1	0.923	0.14
		Days stored	0.962	0.25	0.94	0.43
MLR	4 °C	TVB-N	0.925	1.85	0.821	2.91
		pH	0.905	0.11	0.772	0.17
		Days stored	0.945	0.66	0.832	1.24
	1 °C	TVB-N	0.885	2.21	0.87	2.37
		pH	0.888	0.12	0.853	0.14
		Days stored	0.938	0.94	0.923	1.03

The relationship between texture characteristics parameters and freshness indicators of mutton were established by MLR algorithm. Larger R² and lower RMSE indicate adequate fits. As shown in table 1, a linear correlation between texture parameters and freshness indicators was illustrated by MLR. Models were obtained with high correlation as R² ≥ 0.885 for TVB-N, R² ≥ 0.888 for pH, R² ≥ 0.938 for days stored at three temperatures. For MLR, the values of R² increased with the rising of storage temperature. When the MLR models were used to predict samples in test set, the R² were all higher than 0.8, except for pH at 4 °C (0.772). The MLR model was proved to be effective in the prediction of TVB-N, pH and days stored for mutton samples.

Models built by PLS and MLR showed high ability to predict the content of TVB-N, pH and days stored for mutton. Compared by values of R² and RMSE, better results were found by PLS with high correlation for the training and test sets. Freshness indicators, days stored were best predicted, than comes the TVB-N value. The prediction of pH was not so good.

4 Conclusions

The potential use of texture analyzer to monitor the freshness of mutton stored at different temperatures was evaluated in this study. It was found that texture parameters obtained by TPA analysis could be successfully applied for the monitor of meat deterioration and prediction of freshness indicator (TVB-N, pH, and days stored). Both PCA and CDA showed high ability in the discrimination of mutton samples with different freshness, and the results of CDA were better than that of PCA. The predictive models built by PLS and MLR showed high capacity (R^2 higher than 0.88 for all parameters in calibration data set, higher than 0.77 in validation data set) in mutton freshness prediction based on the texture parameters. The days stored were best predicted, and then comes TVB-N, and pH. What's more, PLS was more effective than MLR when they were used to study the freshness of mutton. The texture analyzer could provide the meat industry with a fast, objective and useful tool for the determination of mutton freshness.

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