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PRINCIPLE COMPONENT ANALYSIS USED TO DESCRIBE COLOUR CHANGES IN MEAT PRODUCTS

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INTRODUCTION

Colour is one of the most important quality attributes of meat and meat products. Development of meat products and product technology emphasizes the importance of a correct description of the colour. Various colour measuring instruments are commercially available. A common feature of these instruments is expression of colour in a three dimensional space. The three coordinates applied to describe the ultimate colour, differ according to the system used. The two three component systems, i.e., CIE (1976) L* (Lightness), a* (red-green), b* (yellow-blue) and L* (Lightness), C* (Chroma), H° (hue) are currently the most used (Rodriguez-Lopez *et al.*, 1992; Chizzolini *et al.*, 1993; Shahioli and Pegg, 1992). The CIE L*C*H* colour system uses cylindric instead of Cartesian coordinates. Although chroma and hue are arithmetic derivatives of a* and b*, they supply additional information about the total chromaticity and the colour tone. Consequently, the most accurate description of the colour of meat and meat products is obtained when all five parameters are used. Thus, measurement of colour and colour stability with different production parameters, relative to light source, intensity and time exposure leads to considerable number of single results. Principal component analysis (PCA), one of the multivariate analysis methods, allows graphical presentation of data with a large number of variables in one projection plane (Marida *et al.*, 1980).

The present study was undertaken to demonstrate the potential of PCA as a tool to describe the colour and colour stability of norwegian dry fermented sausage, relative to light intensity and time of exposure.

MATERIAL AND METHODS

Sausages

Fermented dry sausages were made as described by Næs *et al.* (1991). The colour measurements of sausages were carried out twice, after 14 (PI) and 35 (PII) days of ripening.

Sample preparation

Five slices, 1cm of thickness, were cut to represent each one of the sausage batches. Immediately, after cutting, three replicate colour measurements were taken on a fresh surface and then samples were placed into "Petri dishes". The samples were kept at 4±0.5°C, exposed to 400 and 1000 Lux of fluorescent white light (Osram L 36W/31 Warmtone Lumilux) and air. Colour stability was determined by colour measurement after 0,1,3,6 and 24 hours.

Colour evaluation

A Minolta Chroma Meter CR-200 (Minolta Camera Co., Japan) with an 8mm viewing port was used to measure the three colour coordinates. Chromaticity was expressed as CIE (1976) L*a*b*, where L8 corresponded to lightness, a* to red/green chromaticity and b* to yellow/blue chromaticity. The instrument was adjusted against a white standard (L*=97.4, a*=-0.5, b*= 2.2) using D_{65} illuminant. The total colour difference (E) between two exposing times for

sausage was calculated from equation E_{CIE} (L*a*b*)= $[L^{*2} + a^{*2} + b^{*2}]1/2$. Additionally, the chroma (C*) and hue (H°) were derived from the primary coordinates a* and b*, according to Hunter & Harold, (1987).

Data analysis

The data were first evaluated traditionally by analysis of variance, using GLM procedures (SAS Institute, Inc., 1985). The significance of the main variables (time, lux, ripening period) and first-order interaction between them was determined by an F-test. The mean values (75 measurements) of colour coordinates (L8, a*, b*, C* and H°) for each exposure time at two illuminations (400 lux and 1000 lux) and for two ripening periods were subjected to Principal Component Analysis (PCA), in order to find the most important variables and the patterns in the data set. All variables are treated simultaneously (Mardia *et al.*, 1980). Original data are decomposed and transformed from a coordinate system of many dimensions into the new coordinate system with fewer dimensions. The new coordinates axes are called principal components (factors). The transformation consists in projecting of the samples in space on to an x/y plane. The variables are mean-centred by substraction of the mean value and scaled. Then the data are modelled by orthogonal lines best fitted (least squares) to the samples in the space spanned by the original variables. Factors are interpreted mainly by score- and loading=plots. Scores are an orthogonal projection of the samples on factors (PC) line, and the distance to origin is a score value of each sample. The relationship between the original variables and the new factor are called loadings. The differences between the localization of the sample in the new and the old system are regarded as the residuals. They represent the variability, which is interpreted as noise. In our study PCA was performed using Unscrambler software (Camo, 1992). The number of significant factors from PCA was determined by leverage correction (Martens and Næs, 1989).

The results of variance analysis presented in Table 1, are showing effect of exposure time (T), illumination intensity (L) and ripening period (P) on colour parameters, L8,a*,b*,C* and H°.
Exposure time and light intensity significantly influenced all colour parameters, but largest variations were observed in the hue (H°) and redness (a*). Yellowness (b*) was independent on the light intensity, but significantly affected by the ripening period. Variance analysis revealed that variation of lightness (L*) was mainly caused by the ripening period. The interaction between illumination time and intensity of light was the greatest for H°, but also significance for a* and C* values. The total colour differences (E) of the displayed sausages, between exposure time and initial time, was calculated on the rectangular coordinates L*, a* and b*.

The most intensive discoloration was observed during the first hour of exposure. Sausage slices exposed to 1000 lux, underwent discoloration almost twice as fast as those exposed to 400 lux, particularly during the first hour. These results supports the findings of Iversen (1984), Andersen (1988) and Rodriguez-Lopez (1992), who observed remarkable colour deterioration already in the first hours.

The mean values of colour coordinates L8,a*,b*,C* and H° obtained for each exposure time (o,1,3,6 and 24 hours) at two illumination levels (L=400 lux, H=1000 lux) in two ripening periods (I=14 days, II=35 days), have been subjected to principal component analysis (PCA). It was possible to describe 94.6% of total variation of the original variables with 2 factors (PC's). Each factor was composed of a combination of the original variables. The first factor accounted for 61% of the variance. The second factor explained 33% of the variance. Factor 3 increased explanation as little as 5% and is omitted in the present discussion.

One-vector score plot (Figure 2) for each mean value reveals that factor 1 is mainly connected to the illumination time and light intensity, whereas factor 2 primarily is related to the ripening period.

The two-vector loading plot, illustrates contributions of the variables to the PC-factors (Figure 3) Variables with high absolute values gave a high contribution to factors. The first factor was mainly based on hue angle (H°), which contributed to high positive loading for this factor, as well as redness (a*) and chroma (C*), which gave high negative loadings. The yellowness (b*) also contributed to the explanation of first factor, though to a smaller degree.

The second factor appeared to be composed of lightness (L*) and partly of yellowness (b*).

Two-vector score plot is an even better graphical display of the major variation/covariation pattern of the colour alternations.
Score value of each object (mean value of 75 measurements) was calculated for every factor (Figure 4). This score plot for factor 1 vs 2 illustrates the colour changes during light exposure of sausage slices very well.

At the starting point the surfaces of sausages featured a red tone, expressed by high score values for redness and chroma on one hand and low hue value on the other hand. When the samples were exposed to light and air, colour changes in terms of rectangular (L*,a*,b*) and cylindrical (L*,C*,H°) coordinates took place.

A notable decrease of redness and chroma, with a simultaneous increase of hue and yellowness, was observed during the whole illumination period, particularly in the sausage exposed to 1000 lux. The objects have moved gradually, along factor 1 axis, from left side of the score plot towards the right side. Distinct discoloration of samples subjected to 1000 lux (H°) was observed. Exposure surfaces moved towards the yellow-brown-grey appearance. Factor 2, which explained 33% of the variations was closely related to L* (lightness) and slightly to b* (yellowness). During the illumination the surface of sausages not only changed colour type or tone, but also became darker, which was emerged after 24 hours. Besides, there were revealed differences in lightness between two ripening periods. It seemed that factor 2 was most strongly linked to the ripening period. In PCA similar objects are located close to each other. It is easy to notice that samples illuminated with 1000 lux, reached a certain discoloration level faster than those which were exposed to 400 lux. For example I-H-1 was like I-L-3, i.e. the same changes were obtained for samples exposed to 1000 lux after 1 hour as for samples held 3 hours under 400 lux.

Furthermore, principal component analysis made it possible to separate two groups from the data set. They were related to the ripening periods. Factor 2 delimited marked division between samples from two ripening periods. Sausages analyzed after 35 days of ripening (II), were distinctly darker than after 14 days (I).
The results obtained in the present study, confirmed existing problems with discoloration of cured meat displayed in light (Iversen *et al.*, 1984; Andersen *et al.*, 1988; Rodriguez-Lopez *et al.*, 1992). Discoloration of dry fermented sausages is caused by photo-oxidation of red nitric oxide myoglobin (NOMb) to grey-brown metmyoglobin (MMb). The nitric oxide pigments are photochemically unstable and will upon light exposure, in the presence of oxygen and/or peroxides loose its attractive red colour through light induced colour degradation.

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Table 1. Variance analysis of colour parameters. F-value[a] .

Variance source	Colour parameters L*	a*	b*	C*	H°
Time (T)	*** 8.91	*** 1038.65	** 78.44	*** 479.65	*** 1544.07
Lux (L)	*** 44.41	*** 678.62	0.87	*** 408.91	** 573.45
Ripening (R)	*** 871.15	*** 65.89	*** 210.63	*** 128.52	*** 71.38
T x L	1.21	*** 45.09	1.31	*** 20.22	*** 84.98
T x P	** 4.59	0.30	1.05	0.49	1.39
L x P	2.09	0.83	** 9.14	0.23	*** 15.70

[a] significant effect at the (**) P<0.01 or (***) P<0.001.