A deterioration of shelf life may be pertained to a lack of appropriate sensory attributes, or the appearance of a health hazard. Meat products are primarily exposed to the action of microorganisms. The rate of changes during storage, which may lead to microbial contamination of products, and changes in their sensory attributes are affected by the properties and microbial contamination levels of raw materials, the nature and conditions of the technological process, the character of the product and the amount and type of additives. However, in case of the tested wiener type sausages, the quality of which was modified by colouring with cured blood or a betanin solution and which were aromatized using a mixture of spices (lovage root, dried garlic, 1.5:1.0:4.0) no accelerated storage changes were observed in comparison to the reference sausage. The applied modifications of the technological process do not mask storage changes in the analysed sausages. A distinct deterioration of sensory quality characteristics preceded by 3 days the alarm level of microbial contamination of sausages (an increase in the total aerobic bacteria counts to the level above $1 \times 10^6$).

Key words: sausages with the addition of blood plasma, modification of flavour and colour, storage life

INTRODUCTION

The application of protein preparations as an additive in the production of wiener type sausages has become a common practice. It results from several technological and economic factors. Having consumer preferences in mind, it is however necessary to produce processed meat products with traditional sensory characteristics, in spite of the altered formulation. Several substances may be used to improve the colour or aroma and taste of protein preparations or products produced with their addition [Duda 2001, Kamińska and Góra 1987].

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Natural substances which have been traditional components of the diet are least objectionable for consumers. The beet root pigment, cured blood and herbs meet this requirement. However, each change in the composition of raw materials and the course of the technological process may lead to a change in the storage life of the product. Storage life is most frequently defined as the duration of the period from the moment of production to the time when the undergoing changes deteriorate quality indexes of the product below a certain limit, e.g. requirements of a respective standard. A loss of storage life may refer to a lack of appropriate sensory attributes, a loss of nutritive value or the occurrence of a health hazard [Kołożyn-Krajewska 1998, Libelt 1984 a, b]. Changes in the product may be manifested in biochemical changes, chemical reactions and physical phenomena. Meat products are exposed first of all to the action of microorganisms. The rate of changes during storage, possibly leading to the microbial contamination of products and changes in their sensory attributes are affected by the properties and level of the microbial contamination of raw materials, the specific character and conditions of the technological process, the nature of the product, the amount and type of additives and the efficiency, with which recontamination is eliminated [Brud 2000, Kołożyn-Krajewska 1998, Kostrzewa 1996, Pełczyńska and Szkucik 1993].

This paper is the continuation of previous done and reported studies [Krysztofiak 2004, 2005].

THE AIM OF THE STUDY

The aim of this study was to determine whether colouring agents and spices introduced to the sausages for improving their consumer quality have any effect on the character of storage changes and possibly whether they mask these changes.

EXPERIMENTAL MATERIAL AND THE DESIGN OF THE EXPERIMENT

Three types of sausages were analysed in the study: sausage with a 15% meat protein replacement with non-modified blood plasma (“O”), sausage with a 15% meat protein replacement with aromatized blood plasma and coloured with blood (“K”), and sausage with a 15% meat protein replacement with aromatized blood plasma and coloured with betanin (“B”). All these sausages were produced in accordance with the plant formulation on the basis of the composition and proportions of the control sausage with no blood plasma added: pork class III – 50%, beef class II – 25%, fine fat – 25%, curing mixture – 2%, spices – 0.3% (black pepper, chili peppers, nutmeg), water – 40% batter weight. Model sausages (“O”, “K”, “B”) were produced by removing from the formulation of the control sausage the amount of meat which contained 15% protein found in the batter. This protein was substituted with protein of aromatized porcine blood plasma. In sausages “K” and “B” 0.9% cured blood or 1% betanin (30% solution) were also added to the batter. Sausage “O” was not coloured. Moreover, water and fat levels in the raw material composition were also appropriately adjusted, so that the basic composition was as consistent as possible with the composition of sausage with no blood plasma added. Next the sausages were stored for 11 days at the temperature of...
approx. 6°C and at relative humidity of 85%. In that time sensory examinations and microbiological tests were performed in order to determine the effect of the conducted modifications of colour and flavour on sensory quality of sausages and on changes occurring during storage.

**METHODOLOGY**

Sensory examinations of changes in desirability of sausages during their storage in combination with microbial analyses of the product constitute the basic method to determine the quality and ensure the safety of this group of foodstuffs [Konopka and Pełczyńska 1994, Libelt 1984 a, b]. The course of changes in sensory desirability of sausages was investigated by observing changes in the appearance of the link (the condition of the surface), colour at the cross-section and aroma. Tests were performed on the 1st, 4th, 5th, 6th, 7th, 8th and 11th day after production. Sensory quality evaluation of sausages was performed using a 5-point scale, prepared according to the guidelines in respective standards [PN-ISO 4121:1998, PN-ISO 5492:1997]. Microbial analyses were performed according to the recommendations contained in literature sources [PN-A/82054:1973, PN-A/82055-6:1994]. The total level of aerobic microorganisms and the most likely count of anaerobic bacteria endospores were assessed. The instrumental colour measurement was conducted using a spectrophotometer. Light reflection degree R was measured every 10 nm and the results were expressed in the CIE L*a*b* system. The obtained testing results were analysed statistically.

**DISCUSSION OF RESULTS**

It was found that during the storage of sausages changes in the analysed indexes were similar, although the sample with no colouring substances added (“0”) had a less intense colour. A distinct deterioration in all the analysed quality attributes occurred on the 5th day of storage, although the overall desirability might be considered satisfactory. After that time the adverse changes were accelerated, manifested primarily in the changes in colour at the cross-section and later also in aroma, along with each successive 24 hours. On the 6th day of storage the quality of sausages was assessed as unsatisfactory. The observed changes during storage were similar to the course of changes typical of scalded sausages [Libelt 1984 b].

The course of changes during storage was also followed by observing the dynamics of changes in the total counts of aerobic bacteria and anaerobic bacterial spores. Mean values of the total counts of aerobic bacteria per 1g sausage during storage are presented in Table 1. In modified sausages “K” and “B” microbial contamination after the completion of the production cycle (“initial”) was higher than in the reference sausage “0”. This difference results probably from the contamination of additionally introduced colouring agents and spices [Kostrzewa 1996, Pelczyńska and Szkucik 1993, Wieczorkiewicz-Górnik and Piątkiewicz 2001]. In spite of that fact, contamination did not exceed \(1 \times 10^6\) per 1 g, which is considered the threshold level for this type of processed meat products. The volume of the “initial” contamination of all the tested samples was at the
average level, typical of scalded sausages [Konopka and Pełczyńska 1994, Libelt 1984 a, Pełczyńska and Szkucik 1993]. The alarm level \( (1 \times 10^6/g) \) of the total microbial count was reached by the model sausages only after 11 days of storage. It may be stated that the addition of modified blood plasma and cured blood or betanin does not constitute a hazard for the standard microbial purity of experimental sausages (Fig. 1).

Table 1. Mean values of the total aerobic bacteria count (in 1 g sample) during storage of sausages produced with the addition of non-modified blood plasma and flavour- and colour-modified plasma

<table>
<thead>
<tr>
<th>Storage time, days</th>
<th>Sample “0”</th>
<th>Sample “K”</th>
<th>Sample “B”</th>
</tr>
</thead>
<tbody>
<tr>
<td>Czas przechowywania, dni</td>
<td>Próba „0”</td>
<td>Próba „K”</td>
<td>Próba „B”</td>
</tr>
<tr>
<td>1</td>
<td>( 5.21 \times 10^3 )</td>
<td>( 9.80 \times 10^3 )</td>
<td>( 1.08 \times 10^4 )</td>
</tr>
<tr>
<td>4</td>
<td>( 1.13 \times 10^4 )</td>
<td>( 2.50 \times 10^4 )</td>
<td>( 2.83 \times 10^4 )</td>
</tr>
<tr>
<td>5</td>
<td>( 1.81 \times 10^4 )</td>
<td>( 3.70 \times 10^4 )</td>
<td>( 3.60 \times 10^4 )</td>
</tr>
<tr>
<td>6</td>
<td>( 3.45 \times 10^4 )</td>
<td>( 4.40 \times 10^4 )</td>
<td>( 4.13 \times 10^4 )</td>
</tr>
<tr>
<td>7</td>
<td>( 5.61 \times 10^4 )</td>
<td>( 6.70 \times 10^4 )</td>
<td>( 6.30 \times 10^4 )</td>
</tr>
<tr>
<td>8</td>
<td>( 9.93 \times 10^4 )</td>
<td>( 1.43 \times 10^5 )</td>
<td>( 1.23 \times 10^5 )</td>
</tr>
<tr>
<td>11</td>
<td>( 7.92 \times 10^4 )</td>
<td>( 1.30 \times 10^5 )</td>
<td>( 1.30 \times 10^5 )</td>
</tr>
</tbody>
</table>

Fig. 1. Changes of total bacterial count expressed as \( \log(N/No) \) during storage of sausages produced with the addition of non-modified blood plasma and flavour- and colour-modified plasma

Rys. 1. Zmiany ogólnej liczby bakterii tlenowych, wyrażone jako \( \log(N/No) \), w czasie przechowywania wędlin wyprodukowanych z osoczem niemodyfikowanym oraz z osoczem po modyfikacji smakowitości i barwy
The above conclusion was verified also by assessing the effect of the type of sausage on the microflora growth dynamics. The development of bacteria may be described by the logistic curve. However, under the conditions of the discussed experiment only the so-called exponential part of the graph is found. To describe this stage of microflora development the following function may be applied:

\[ N(t) = N_0 \cdot \exp(a \cdot t) \]

where:  
- \( N \) – microbial count after time \( t \),  
- \( N_0 \) – initial microbial count,  
- \( t \) – time,  
- \( a \) – constant.

The above equation may be presented in the linear form:

\[ \ln \frac{N}{N_0} = f(t) \]

Data presented in Table 1 were subjected to appropriate calculations to establish the correlation between the change in microbial counts after a given storage time (dependent variable) and storage time (\( t \) – independent variable).

Differences between slopes of straight lines of the calculated equations are statistically non-significant (at \( \alpha = 0.05 \)), which indicates that there is no significant effect of the type of the used blood plasma on the dynamics of microflora growth (Table 2). Anaerobic bacterial spores are found in the analysed sausages in small amounts. No statistically significant effect of the type of sausage was found on the dynamics of changes in the most likely count of anaerobic bacterial spores during the storage of sausages (Fig. 2, Table 3).

Table 2. Correlations between changes in microbial counts and sausage storage time depending on the type of sausage

<table>
<thead>
<tr>
<th>Sample</th>
<th>Regression Equation</th>
<th>( R^2 )</th>
<th>( S_{(y)} )</th>
</tr>
</thead>
</table>
| 0      | \( \ln(y) = -1.0207 + 0.5098 \cdot t \)  
\( y = 0.36033 \cdot \exp(0.5098 \cdot t) \) | 0.959* | 0.3610 |
| K      | \( \ln(y) = -0.9581 + 0.4753 \cdot t \)  
\( y = 0.38360 \cdot \exp(0.4753 \cdot t) \) | 0.931* | 0.4478 |
| B      | \( \ln(y) = -0.9601 + 0.4575 \cdot t \)  
\( y = 0.38280 \cdot \exp(0.4575 \cdot t) \) | 0.907* | 0.5079 |

*F value (\( p \leq 0.05 \)).
*Wartość F (\( p \leq 0.05 \)).
Table 3. Correlations between changes in the most likely anaerobic endospore counts and sausage storage time depending on the type of sausage

<table>
<thead>
<tr>
<th>Sample</th>
<th>Regression Równanie regresji</th>
<th>$R^2$</th>
<th>$S_{(y)}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>$\ln (y) = -0.1873 + 0.2235 \cdot t$</td>
<td>0.941*</td>
<td>0.1942</td>
</tr>
<tr>
<td></td>
<td>$y = 0.1892 \cdot \exp(0.2235 \cdot t)$</td>
<td></td>
<td></td>
</tr>
<tr>
<td>K</td>
<td>$\ln (y) = -0.1938 + 0.1815 \cdot t$</td>
<td>0.974*</td>
<td>0.1024</td>
</tr>
<tr>
<td></td>
<td>$y = 0.8230 \cdot \exp(0.1815 \cdot t)$</td>
<td></td>
<td></td>
</tr>
<tr>
<td>B</td>
<td>$\ln (y) = -0.1815 + 0.2291 \cdot t$</td>
<td>0.943*</td>
<td>0.1951</td>
</tr>
<tr>
<td></td>
<td>$y = 0.9217 \cdot \exp(0.2291 \cdot t)$</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*F value ($p \leq 0.05$).
*Wartość F ($p \leq 0.05$).

Fig. 2. Changes of mean values of the most likely anaerobic endospore counts ($\log (N/N_0)$) during storage of sausages produced with non-modified blood plasma and with flavour- and colour-modified plasma

During the storage of sausages colour changes were also assessed by determining physical colour parameters ($L^*a^*b^*$). The obtained results were subjected to the multivariate statistical analysis. However, no significant dependencies were found between the dynamics of changes in colour parameters and the application of a colouring agent.
Moreover, no significant correlation was found between the time of storage (in the analysed range) and changes in colour parameters.

The presented dependencies justify the statement that the proposed quality modification methods of experimental sausages do not cause accelerated microbial development or change the dynamics of colour changes in comparison to non-modified sausages.

CONCLUSIONS

1. The introduction of a colouring agent and an additional amount of spices resulted in an increase in the initial count of aerobic bacteria from $5.21 \times 10^3$ to $1.08 \times 10^4$, but no statistically significant effect was found of the type of sample on the growth dynamics of these bacteria.

2. The most likely count of anaerobic bacterial spores was low and no effect of the type of the sample was observed on the dynamics of the increase in this count.

3. The introduced modification of the technological process do not mask changes during storage in the analysed sausages. A distinct deterioration in sensory quality attributes preceded by 3 days the alarm volume of microbial contamination of these sausages (an increase of the total aerobic bacteria count to the level exceeding $1 \times 10^6$).

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Streszczenie. Utrata trwałości może dotyczyć braku odpowiednich cech sensorycznych lub pojawienia się niebezpieczeństwa zdrowotnego. Na tempo zmian przechowalniczych mają wpływ właściwości oraz poziom zakażenia mikrobiologicznego surowców, specyfika i warunki przebiegu procesu technologicznego, charakter produktu oraz ilość i rodzaj dodatków. W wypadku badanych kiełbasi, typu parówkowego, których jakość modyfikowano za pomocą barwienia krwią peklowaną bądź roztworem betaniny oraz aromatyzowano, używając mieszany przypraw (korzeń lubczyka, susz czosnkowy, 1,5:1,0:4,0) nie zaobserwowano przyspieszonych zmian przechowalniczych. Wyraźne pogorszenie sensorycznych wskaźników jakości wystąpiło 3 dni wcześniej niż alarmowa wielkość zakażenia mikrobiologicznego kiełbasi (zwiększenie ogólnej ilości bakterii tlenowych do poziomu powyżej $1 \times 10^6$ jkt).

Słowa kluczowe: kiełbasy z dodatkiem osocza, modyfikacja smakowitości i barwy, trwałość

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