THE EFFECT OF DIFFERENT PRESERVATION CHEMICALS AND STORAGE TEMPERATURES ON CHEMICAL COMPOSITION AND MICROBIOLOGICAL SAFETY OF MILK

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Abstract
Milk is a product that due to its natural components is very unstable at temperatures higher than 4°C. Because of its high nutritive value milk is an ideal medium for the rapid multiplication of bacteria, particularly under unhygienic production and storage at ambient temperatures. For any dairy industry to make good dairy products, high quality of raw materials is essential.

Milk quality control is based on the use of approved tests to ensure the application of approved practices, standards and regulations concerning the milk and milk products. The tests are designed to ensure that milk products meet accepted standards for chemical composition and microbiological safety. Milk testing and quality control is an essential component of any milk processing industry.

The aim of this research was to analyze the effects of different types of preservatives (azidiol and bronopol) and different storing temperatures (+4°C and room temperatures) on the maintenance of chemical and microbiological properties of milk.

According obtained results the best maintenance of chemical and microbiological properties of milk was noted on samples preserved with azidiol and stored at (+4°C).

Key words: preservatives, storage temperature, milk quality control

Introduction
Cow’s Milk contains water (87.25%) , fat (3.80%), protein (3.50%), lactose (4.80%) and minerals (0.65%)(Eckleset et al., 1951). Milk contains considerable amount of water soluble vitamins and fat soluble vitamins. Because of the large amount of water, vitamins and slightly acidic pH milk is also good food for the growth of different microorganisms either desirable or undesirable . On the other hand, Undesirable microorganisms may disturb the actual flavor or texture, and also some of them if they are pathogenic, can cause different diseases in humans (Akram).

Milk is almost sterile when is secreted from a healthy udder. The natural inhibitors in milk (e.g., lactoferrin and lactoperoxidase) prevent significantly bacterial contamination for the first three to four hours after milking, at ambient temperatures. Cooling to 4 °C within this period maintains the original quality of the milk and is the method of choice for ensuring good -quality milk for processing and consumption.

In Macedonia, most of the milk comes from small farms in rural areas. Due to traditional handling in farms in not sufficiently hygienic conditions, spoling nature nature, milk is often
spoiled, particularly in summer season.

In the milk industry it is needed to take the samples of milk for further analyses either microbiological or chemical. But milk samples can be spoiled by because of the long road to the laboratory especially when the temperatures are high. In order to avoid microbial contamination of milk, increase od normal bacterial flora, and to keep normal compositiotion of milk, milk samples taken for analysis of quality and safety of raw milk are needed dto be preserved. Different types of preservation methods are used worldwide.

Chemical milk preservation has been practiced for a long time since milk fat testing became a standard and routine exercise. Several substances and formulations have been used in the past, but the search for the ideal milk sample preservative continues. The search for preservatives depends upon the purpose for which it is required. Different types of preservatives may be required in different situations. If the sample requires short term preservation, for example, for carrying it from the farm to the laboratory for analysis, a preservative which provides small increase  of sample shelf life of sample is needed.( Upadhyay, Goyal, Kumar, Ghai, Singh)

Aim of this work was to compare efficiency of different chemical preservatives used for preservation for milk samples for laboratory analysis. Accuracy and precision are important in analysis of the milk because obtained results are not only indicators for safety of milk, they are also indicator of milk quality and present the basis for calculation of milk price.

In the past most commonly used preservatives were hydrogen peroxide, formaldehyde, potassium dichromate, mercuric chloride, boric acid or their combinations. These days most commonly used are bronopol and azidiol.

**Materials and methods**

**Sampling procedure**

Milk from local farms was collected and it was tested at a local laboratory for dairy products. Milk samples were grouped in following groups:

1. Containers with fresh raw milk without added preservatives.
2. Containers with added azidiol and stored at temperature of 4°C for 3days
3. Containers with added azidiol and stored at temperature of 4°C for 10 days
4. Containers with added azidiol and stored at a temperature of 20 °C for 3days
5. Containers with added azidiol and stored at temperature of 20°C for 10 days
6. Containers with added bronopol and stored at a temperature of 4 °C for 3days
7. Containers with added bronopol and stored at temperature of 4°C for 10 days
8. Containers with added bronopol and stored at a temperature of 20 °C for 3days
9. Containers with added bronopol and stored at a temperature of 20°C for 10 days

Analysed parameters in raw milk were total bacterial number and content of fat, protein, lactose and dry matters.

Chemical testing was made by using the infrared spectroscopy on a MilkoScan FT6000 TM and for analysis of total bacterial number BactoScan TM FC, which works on flow cytometry. The results were calculated and statistically processed with Microsoft Excel.

**Results and discussion**

Bronopol is a formaldehyde-releasing preservative particularly because of its high activity against gram-negative bacteria especially Pseudomonas aeruginosa. It can stabilize bacterial fora minimum of one year under normal storage conditions and no photodecomposition is reported. However, it decomposes more rapidly at elevated temperatures and alkalinity.

Azidiol is commonly used in all milk-testing laboratories. It contains two components:sodium azide and chloramphenicol. Coli-form bacteria and Staphylococci are sensitive to sodium azide while Salmonella spp., E. coli, Listeria sp. and S.aureus are more sensitive to chloramphenicol.

The samples were analyzed and average of the results was calculated and it is shown in the following tables.
Table 1. Analyses of samples of fresh raw milk without added preservatives

<table>
<thead>
<tr>
<th>Samples without preservative - first day</th>
<th>CFU/mL</th>
<th>Fat (%)</th>
<th>Protein (%)</th>
<th>SNF (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>$3.585 \times 10^3$</td>
<td>3.71</td>
<td>3.28</td>
<td>8.41</td>
</tr>
</tbody>
</table>

Table 2. Chemical and microbiological analyses of samples with added azidiol and bronopol after 3 days at temperatures of 4°C

<table>
<thead>
<tr>
<th>samples with azidiol added</th>
<th>CFU/mL</th>
<th>Fat (%)</th>
<th>Protein (%)</th>
<th>SNF (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>$3.373 \times 10^3$</td>
<td>3.70</td>
<td>3.26</td>
<td>8.37</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>samples with bronopol added</th>
<th>CFU/mL</th>
<th>Fat (%)</th>
<th>Protein (%)</th>
<th>SNF (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>$3.654 \times 10^3$</td>
<td>3.67</td>
<td>3.19</td>
<td>8.31</td>
</tr>
</tbody>
</table>

Table 3. Chemical and microbiological analyses of samples with added azidiol and bronopol after 3 days at temperatures of 20°C

<table>
<thead>
<tr>
<th>samples with azidiol added after</th>
<th>CFU/mL</th>
<th>Fat (%)</th>
<th>Protein (%)</th>
<th>SNF (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>$6.171 \times 10^3$</td>
<td>3.65</td>
<td>3.18</td>
<td>8.36</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>samples with bronopol added after</th>
<th>CFU/mL</th>
<th>Fat (%)</th>
<th>Protein (%)</th>
<th>SNF (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>$8.088 \times 10^3$</td>
<td>3.57</td>
<td>3.15</td>
<td>7.47</td>
</tr>
</tbody>
</table>

Table 4. Chemical and microbiological analyses of samples with added azidiol and bronopol after 10 days at temperatures of 4°C

<table>
<thead>
<tr>
<th>samples with azidiol added</th>
<th>CFU/mL</th>
<th>Fat (%)</th>
<th>Protein (%)</th>
<th>SNF (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>$3.177 \times 10^3$</td>
<td>3.77</td>
<td>3.28</td>
<td>8.40</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>samples with azidiol added</th>
<th>CFU/mL</th>
<th>Fat (%)</th>
<th>Protein (%)</th>
<th>SNF (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>$1.970 \times 10^3$</td>
<td>3.76</td>
<td>3.30</td>
<td>8.46</td>
</tr>
</tbody>
</table>

The samples of milk after 10 days of storage at 20°C were spoiled and they were not taken for further analysis.

According obtained results presented in tables (1,2,3,4), is noted that after 3 days of storage results are much better with both preservatives azidiol and bronopol at temperatures of 4°C compared to the ones stored at 20°C. In terms of total number of microorganisms the samples with azidiol stored at 4°C are showing 6% less than the samples on first day without preservative and samples with added bronopol at 4°C 1.9% more. The samples at 20°C with azidiol shown 72% higher and samples with bronopol shown 125% higher number of bacteria compared with samples without preservative. These results shown that the effect of both preservatives is low at higher temperatures of storage. Comparison of samples without preservative with samples stored in period of 10 days at 4°C with azidiol and samples with bronopol is 11% and 45% less respectively. These results shown that bronopol is less stable on longer period of storage than azidiol.

Regarding the changes of chemical composition of milk, obtained results have shown that the samples stored for 3 days and preserved with azidiol at 4°C shown less deviation than the samples with bronopol. Also the samples stored at 20°C shown that bronopol is less effective. On other hand the samples stored in period of 10 days at 4°C the results did not shown significant differences between the results of the two preservatives. Samples with applied preservatives shown similar results with samples without preservatives, which means that both preservatives are effective in keeping unchanged composition of milk.

Conclusion

Adding preservatives in milk samples helps the dairy industry to take and keep them from spoiling during transportation from the farm to the laboratory. According obtained results it is best to keep the milk at temperatures of 4°C and maintain unchanged chemical composition and avoid microbiological contamination it is best to use azidiol. If the samples are supposed to
be used after a longer period (10 days) the best way is to keep them at 4° C and to use azidiol or bronopol as preservative. But if the samples are kept at ambient temperatures for longer period of time (10 days), the preservatives are not effective, because of milks’ natural decomposition in such environment. In that case preservatives can not stop the spoiling of the samples.

References

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