

Valuation of *Staphylococcus aureus* pathogen in the fresh milk of the Tirana (Albania)

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SUMMARY

During 2004 we have analyzed 120 milk samples, which were taken from two different factories in Tirana. Out of these, 60 samples are unpasteurized milk, the other 60 samples are taken from the same milk, after the process of pasteurization. These samples have been analyzed for the presence of pathogens, which can be very dangerous for the consumers' health. From the isolated pathogens of three samples of milk, we identified the presence of: *Staphylococcus aureus*, which represent 5% of the samples of unpasteurized milk or 2.5% of all the verified milk. The above results show that the unpasteurized milk taken from these two factories is represented with the presence of *Staphylococcus aureus*, which is considered a dangerous pathogen for the consumers.

Keywords: fresh milk, pathogene, *Staphylococcus aureus*

INTRODUCTION

Fresh milk is thought to be one of the main sources of food intoxication in people. The sources of this pollution in fresh milk are numerous and are connected with factors such as: the animals' hygiene, the hygienic-sanitary conditions during process of milking, the conditions of accumulation, transportation, etc. Milk is a product which favors the growing types of microorganisms. The level of pollution of this product from the process of milking up to the consumer is growing of microbes. Such process is in direct correlation with the time keeping processes and the temperature of the environment. (Nagle, 1999)

High temperatures in the environment increase the number of microorganisms in a logarithmic scale for the volume unit. The presence of pathogenic microorganisms in fresh milk can become a consequence of different infective processes in the animal breast (clinical and subclinical mastitis) or in its body. (Ingalls, 2000). Furthermore milk can be contaminated indirectly from the mastitis manipulations of breast as well as the pollution of the product during milking.

Cows with mastitis are used as a resource in transmitting pathogenic microbes in, like those of *Staphylococcus* type and other different pathogen.

(Ingalls, 2000). To eliminate these dangerous pathogens mentioned above, the milk must follow a strict pasteurization process. The pasteurization is a thermic process used to decrease the level of pollution of microorganisms in milk. This is a method of milk healthiness which guarantees the destruction of all the vegetative forms of microorganisms, excluding those of the spore forms. (Bramley, 2000)

MATERIAL AND METHODS

Nowadays in order to measure bacterial pollution in milk, there are a variety of methods used to identify microorganisms (Kramer, 1991). The routine methods used in the laboratories are oriented towards the standard methods. (Robert, 1992). In this study we have used the standard methods, such as: (91/180/EEC). The study is carried out in the Institute of Veterinary Research, Department of Microbiologic Control of Food.

The raw milk samples for the analysis have been taken from two factories in Tirana. The aim of this study is to show the pollution of milk caused from *Staphylococcus aureus* before and after the pasterization process. There have been 120 milk samples analyzed. Out of them 60 came from fresh milk and 60 samples taken from the same milk after the pasterization process.

The samples are taken by sterile items using laboratory sterile bottles at 500ml in volume. The study was carried out for the 12 months of the year 2004. For each sample there has been a strict documentation, such as: the quantity, the date, the place of collection, transportation, etc.

There have been prepared the diluted decimal 10^{-1} up to 10^{-6} for each sample. The process has started at 25 ml of milk and 225 ml of diluted substance. The analytical process used the above decimal dilution because the number of microbes under the first dilution is extremely large.

Afterwards the procedure was carried out acknowledging other analytical indicators it. The analytical procedure uses the standard methods ISO (ISO 11 866-2).

The presence of *S. aureus* is achieved by analyzing the samples of unpasteurized and pasteurized milk, according to the ISO methods (ISO 8261:2001 (E), IDF 122:2001 (E)).

The occurrence and measurements of *Staphylococcus aureus* is achieved by transferring 1 ml of each pre-arranged dilution in each two separate agar selective plates. For this purpose the Baird Parker Agar terrain has been used. The planted plates are incubated at 37°C for 48 hours. (Microbiology of food).

The counting and identification is being done after the incubation. At least 5 typical and atypical colonies were chosen, from which is achieved the microscopic preparation and the coloring by the Gram method. The suspicious colonies are planted in the agar-blood terrain, to see the hemolysis phenomena. The *Staphylococcus aureus* pathogens have appeared β -hemolytic.

Furthermore, we have followed with the test of biochemical identification of suspicious colonies. This test is achieved with the API *Staph* system. The confirmation of the above pathogens is attained, by the coagulosis test. As a result, the suspicious colonies have been passed in the infusion of brain and heart, BHI (Brain Heart Infusion), for 24 hours in 37°C.

In order to accomplish this test there was used: commercial rabbit plasma, which is distributed in serologic tubes in 0.3-0.5 ml volume. The confirmation of coagulative mass is attained in the incubated tubes after 4, 6 and 24 hours incubation. The coagulation of plasma results a positive test and it confirms the presence of *Staphylococcus aureus*. (ISO 10560:1993 (E)).

RESULTS AND DISCUSSIONS

During 2004 we have analyzed 120 milk samples, which were taken from two different factories in Tirana. Out of these, 60 samples are unpasteurized milk, the other 60 samples are taken from the same milk, after the process of pasteurization. These samples have been analyzed for the presence of pathogens, which can be very dangerous for the consumers' health. From the isolated pathogens of three samples of milk, we identified the presence of: *Staphylococcus aureus*, which represent 5% of the samples of unpasteurized milk or 2.5% of all the verified milk. The above results show that the unpasteurized milk taken from these two factories is represented with the presence of *Staphylococcus aureus*, which is considered a dangerous pathogen for the consumers.

Unpasteurized milk is a source of microbe's contamination even though with the presence of dangerous pathogens.

Table 1. Number of milk samples and its analytic indicators.

| Type of sample | Nr. of analyzed samples | Analytic indicators | |
|--------------------|-------------------------|---------------------|------|
| Unpasteurized milk | 60 | | |
| Pasteurized milk | 60 | Staph. aureus | Etc. |
| Total | 120 | | |

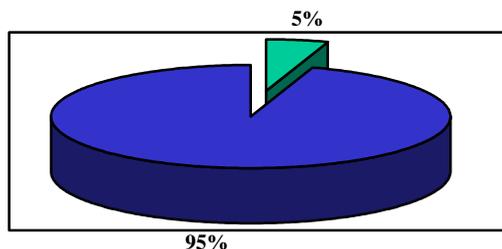
Table 2. Positive cases of *Staph. aureus* in milk samples.

| Samples | Isolated pathogen: <i>Staph. aureus</i> |
|--------------------|---|
| Unpasteurized milk | 3 samples |
| Pasteurized milk | - |

5% of the unpasteurized milk samples resulted positive for *Staph. aureus* presence.

Table 3. Percentage of the positive cases and type of samples.

| Analytic indicators | Nr. of positive samples | Percentage of total of samples | Percentage of total of samples of unpasteurized milk | Percentage of total of samples of pasteurized milk |
|------------------------------|-------------------------|--------------------------------|--|--|
| <i>Staphylococcus aureus</i> | 3 | 2.5 % | 5 % | - |

Graph 1. Positive cases of *Staph. aureus*, isolated and identified in milk samples.

CONCLUSIONS

1. Unpasteurized milk has resulted with microbe contamination.
2. The presence of *Staphylococcus aureus*, up to 5% of total unpasteurized samples is a dangerous index for health consumers
3. The presence of *Staphylococcus aureus* is not connected with high temperature of the environment (spring-summer) because the tow factories had respected the cooling chain process.

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