

MICROBIOLOGICAL CONTROL OF GOAT MILK FROM SIBIU AREA REGARDING THE HYGIENE ASSURANCE AND FOOD SAFETY

CONTROLUL MICROBIOLOGIC AL LAPTELUI DE CAPRĂ DIN JUDEȚUL SIBIU ÎN VEDEREA ASIGURĂRII IGIENEI ȘI SIGURANȚEI ALIMENTARE

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The main objective concerning food safety is touching the highest possible degree of protection for human health and consumers interests regarding the food. Because of that, we analyze goat milk from three different county and the tests were performed searching the Salmonella presence. In our tests we detect an insignificant presence of Salmonella but this microorganism can be eliminated by specific methods so the food safety will be ensured.

Key words: goat milk, food safe, Salmonella

Introduction

The quality and safety of the alimentary products have become a right of the consumers, with direct effects on the quality of life, and the problems based on the quality and the safety of the products is in the center of attention for organisms set up for defending the consumers interests.

The UE strategy regarding the food safety is based on four important elements:

- norms regarding the safety of nourishment and animal fodders
- independent and widely accessible scientific consultancy
- measures for implementing the rules and control processes
- recognizing the consumer's right to choose on the basis of full information on the origin and contents of food.

Materials and Methods

The milk contains approximately 87,5% water, and the rest of 12,5% is a completely dry substance. The milk components are found in various forms: in

emulsion (fat, pigments and liposoluble vitamins), in colloidal dispersion (protean substances) and in solution (lactose, mineral salts, pigments and water-soluble vitamins). The milk proteins are composed of casein (80%-85%), lactalbumin (10%-12%) and lactoglobulin (5%-8%). They represent the most valuable element of the milk, they are complete proteins, that is why they contains the eight essential aminoacids.

In order to avoid environmental contamination and the sample taken into work, it is recommended to work in special rooms or niches with rolling flux. Certain products known to contain very few microorganisms will always be examined first, followed by another product, known to have a higher contamination.

Samples will be handled in order to avoid any risk of contamination. To achieve this the following precautions will be taken:

- when not working in the niche, work near the flame
- the milk will be collected in sterile containers
- any tool that is used must be sterile.

There were taken 25 g (ml) of the sample to be analyzed and added 225 ml pre-enrichment media (half-Fraser broth) and placed in thermostat at $30^{\circ}\text{C} \pm 1^{\circ}\text{C}$ for incubation for $24\text{h} \pm 2\text{h}$. After the incubation of the initial suspension there were transferred 0.1 ml of the resulting culture in a 10 ml test-tube with secondary enrichment media (Fraser broth). The sown media was incubated, for $48\text{h} \pm 2\text{h}$ at 35°C or 37°C . The plates with selectively sown isolation medias (3 plates corresponding to the place of origin: Rasinari, Cristian and Miercurea Sibiului, Sibiu County communes) shall be incubated at 37°C for 24 hours with the lid down and not in contact with the walls or with the upper part of the incubator. Typical colonies of Salmonella were cultivated on Wilson-Blaire agar and on Kristensen agar.

In order to confirm, from each plate containing each of the two selective medias, it is taken at least one colony considered to be typical or suspect. If there is found a plate with less than five typical or suspect colonies, there will be kept all typical or suspicious colonies. The selected colonies will be sown, on the priorly dry surface of the nutritious agar plates, so as to achieve the development of isolated colonies. The so inoculated plates are sown, in a thermostat at $37 \pm 1^{\circ}\text{C}$ for 24 ± 3 hours. For biochemical and serological confirmation pure cultures are used. There will be sown, with each of the cultures obtained from the kept colonies, the following medias: TSI Agar, Agar with urea, Media for L-lysine decarboxylation. The agar column will be sown by pricking, and the slope by striation. They will be incubated in a thermostat at $37 \pm 1^{\circ}\text{C}$, for 24 ± 3 hours.

To prove the presence of the β -galactose it will be dispersed a drop from the suspect colony, in a test-tube containing 0,25 ml saline solution, add a toluene drop and stir the content of the test-tube. The test-tube will be introduced in a thermostable water bath, at 37°C and left to stay for several minutes (about 5 minutes). Add 0,25 ml of reactive for proving the presence of β -galactosidase and mix. Reintroduce the test-tube in the thermostable water bath, at 37°C and let in for 24 ± 3 hours. For the Voges-Proskauer (VP) reaction on the media it is sown

from the suspect colony in a test-tube containing 3ml of VP media. It is incubated in a thermostat at $37\pm 1^{\circ}\text{C}$, for 24 ± 3 hours. After the incubation, add two drops of the creatine solution, three drops of the α -naphthol alcoholic solution and then two drops of the potassium hydroxide, stirring after adding each reactive.

To prove the presence of the indole on the media, a test-tube containing 5 ml of tryptophan is sown with the suspect colony. It is incubated in the thermostat at $37\pm 1^{\circ}\text{C}$, for 24 ± 3 hours. After the incubation add 1 ml of Kovacs reactive.

Results and Discussion

Microbiological confirmation on TSI agar culture media is indicated as follows:

- on the column:
 - yellow – glucose-positive (fermentation of the glucose);
 - red or unchanged – glucose-negative (absence of glucose fermentation);
 - black – formation of sulfuretted hydrogen;
 - bubbles or clefts – formation of gas from glucose,
- on the slope:
 - yellow – lactose- and/or sucrose-positive (fermentation of lactose and/or sucrose);
 - red or unchanged – lactose and sucrose-negative (neither lactose, nor sucrose ferment). Typical culture of Salmonella correspond to an alkaline slope (red), with formation (in 90% of the cases) of sulfuretted hydrogen (blackening of the agar). When a lactose-positive Salmonella is being isolated, the slope of the TSI agar is yellow. Therefore, a preliminary confirmation of the Salmonella cultures doesn't have to be based only on the results obtained on the TSI agar.

1. Microbiological confirmation on agar culture media with urea. In the case of the positive reaction, the decomposition of urea produces emission of ammonia, making the red from the phenol turn into pink, then to dark red. The reaction is frequently visible after 2-4 hours.

2. Microbiological confirmation on L-lysine decarboxylation media. Turbidity and a purple color, after the incubation, indicate a positive reaction, while a yellow color indicates a negative reaction.

3. Microbiological confirmation on culture media for proving the presence of β -galactose. A yellow color indicates a positive reaction. The reaction is usually visible after 20 minutes.

4. Microbiological confirmation on culture media for the Voges-Proskauer (VP) reaction. The formation of a pink to bright red, in approximately 15 minutes indicates a positive reaction.

5. Microbiological confirmation on culture media for proving the presence of indole. The formation of a red ring indicates a positive reaction, while a brown-yellow ring indicates a negative reaction.

Microbiological confirmation on culture media can be emphasised in the table no. 1

Table 1

Microbiological confirmation on culture medias

	Salmonella species							
	S.Typhi		S.Paratyphi A		S.Paratyphi B		Other species	
	Reaction	% b)	Reaction	% b)	Reaction	% c)	Reaction	% b)
TSI media, resulting acid because of glucose fermentation (m1)	+	100	+	100	+		+	100
TSI media, resulting gas from glucose (m2)	-d)	0	+	100	+		+	92
TSI media, resulting acid because of lactose fermentation (m3)	-	2	-	100	-		-	1
TSI media, resulting acid because of sucrose fermentation (m4)	-	0	-	0	-		-	1
TSI media, resulting sulfuretted hydrogen (m5)	+	97	-	10	+		+	92
Urea hydrolysis (m6)	-	0	-	0	-		-	1
Lysine decarboxylation (m7)	+	98	-	0	+		+	95
β -galactosidase reaction (m8)	-	0	-	0	-		-	2 e)
Voges-Proskauer (m9)	-	0	-	0	-		-	0
Formation of indole (m10)	-	0	-	0	-		-	1

a) Take from the referece SR EN ISO 6579/2003.

b) These percentages indicate that not all isolated serotypes of Salmonella give reactions marked with + or -. These percentages may vary between and within serotypes due to poisoning serotypes from different parts of the food.

c) The percentages are not given in public literature.

d) Salmonella Typhi is anaerogenic.

e) Salmonella enterica, subspecies Arizonae gives a lactose-positive or negative reaction, but it is always β -galactosidase-positive. To study these species is necessary to perform additional tests.

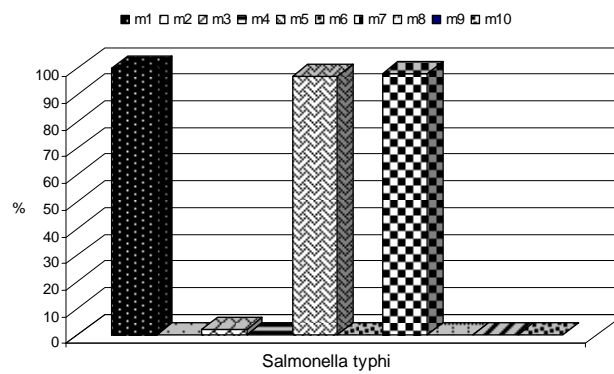


Figure 1 Microbiological confirmation for Salmonella Typhi

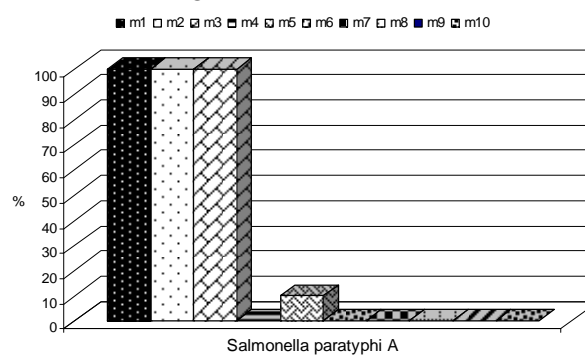


Figure 2 Microbiological confirmation for Salmonella Paratyphi A

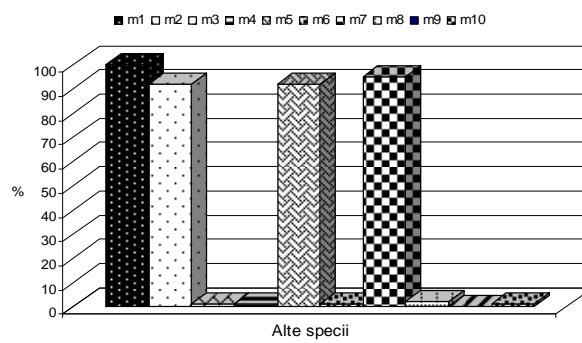


Figure 3 Microbiological confirmation for Other Species

Conclusions

After we made the tests of the milk that we collect from three different area Rășinari, Cristian and Miercurea Sibiului we detect an insignificant presence of Salmonella. The values that we found are insignificant so there aren't any contamination dangers. Salmonella is an important factor but this microorganism can be eliminated by specific methods so the food safety will be ensured.

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