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THE INFLUENCE OF NITRATES ADDITION TO MILK ON CHOSEN PROPERTIES OF CHEESE

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Key words: nitrates and nitrites, cheese ripening, coliform bacteria

The nitrates content in cheese was proportional to the amount of saltpetre added to the milk, and this relationship remained unchanged for a long time during cheese ripening if the number of coliform bacteria did not exceed 10^3 g. The nitrates reduction rate was proportional to the number of coliform bacteria. The nitrites content grew (up to a point) with the increase of the number of these bacteria. The saltpetre addition to the milk reduced the multiplication rate of coliform bacteria in the cheese only when the milk used to produce the cheese was strongly contaminated with these bacteria. In such a case the advantageous effect of saltpetre on cheese quality was also apparent.

INTRODUCTION

In some countries, saltpetre is still being added to milk, mainly in the production of Dutch type rennin cheeses (maximally 0.02% of KNO_3), to inhibit the gas-forming direction of fermentation due to coliform bacteria, and the butyric fermentation caused by anaerobic spore-forming bacteria.

It is generally believed that saltpetre additions in cheese making do not affect the number of coliform bacteria, but merely alter fermentation metabolism, reducing gasses production [8]. There have been model studies in which the saltpetre addition was in fact found to stimulate the development of these bacteria [3, 9].

In this research we studied the transformations of nitrates and nitrites during Dutch type cheese ripening depending on various saltpetre additions to the milk, the variability of the number of coliform bacteria in cheese, and the effect of the saltpetre additions on these bacteria.

MATERIAL AND METHODS

Four series of experiments were performed, each involving three repetitions of Edam-type cheese production with various amounts of saltpetre added to the milk (0,5, 10 and 20 g KNO_3 per 100 l of milk). The milk used to produce cheese in series 1, 2 and 4 was pasteurized at 74°C ± 1°C for 10-15 s, while in series 3 the milk was not heat treated. The cheese was produced on quarter-technological scale in the laboratory (series 1, 3, 4) or in a dairy plant (serie 2) according to the standard procedure for Edam cheese.

The contents of nitrates and nitrites, coli titre, water content, titratable acidity, and pH were determined in the cheeses after salting and during ripening.

The contents of nitrates and nitrites were determined according to the Polish Standard PN-81/A-86234; the remaining determinations were done according to standards PN-73/A-86232 and PN-77/A-86031. The organoleptic preperties of the studied cheeses were assessed according to a five-point scale.

RESULTS AND DISCUSSION

In the first series of experimentes efforts were made to keep the number of coliform bacteria in the cheese as low as possible. The results are listed in Table 1 and illustrated in Figs. 1 and 2 showing changes in NO_3^- and NO_2^- contents. As planned, the coliform bacteria content in these cheeses was fairly low: 10-10^3/g (to facilitate results interpretion, the coli titre was conventionally expressed as the approximate number of coliform bacteria). The nitrates content in the cheeses was proportional to the saltpetre addition to milk practically throughout the

Table 1. Changes of selected features of Dutch type cheese produced in good hygienic conditions from milk with various additions of saltpetre

KNO_3 addition (g/100 l of milk)	Time of ripening (weeks)	Acidity		Water content (%)	Coli titre	Organoleptic assessment after 6-8 weeks of ripening (5-point scale)
		pH	°SH			
0	S*)	5.02	50.5	50.5	10^{-2}	4.0
	3	5.18	50.0	48.2	10^{-2}	
	6	5.14	54.5	45.6	10^{-2}	
	9	5.10	52.0	43.3	10^{-2}	
5	S	5.04	60.1	46.7	10^{-2}	3.9
	3	5.22	56.6	42.3	10^{-2}	
	6	5.23	59.0	41.2	10^{-2}	
	9	5.23	60.2	40.1	10^{-2}	
10	S	5.08	59.5	47.5	10^{-2}	4.0
	3	5.20	56.7	42.5	10^{-2}	
	6	5.18	58.0	41.0	10^{-2}	
	9	5.21	59.9	40.1	10^{-2}	
20	S	5.08	62.5	45.6	10^{-2}	4.1
	3	5.23	58.0	41.2	10^{-2}	
	6	5.24	57.0	40.2	10^{-2}	
	9	5.22	59.4	39.3	10^{-2}	

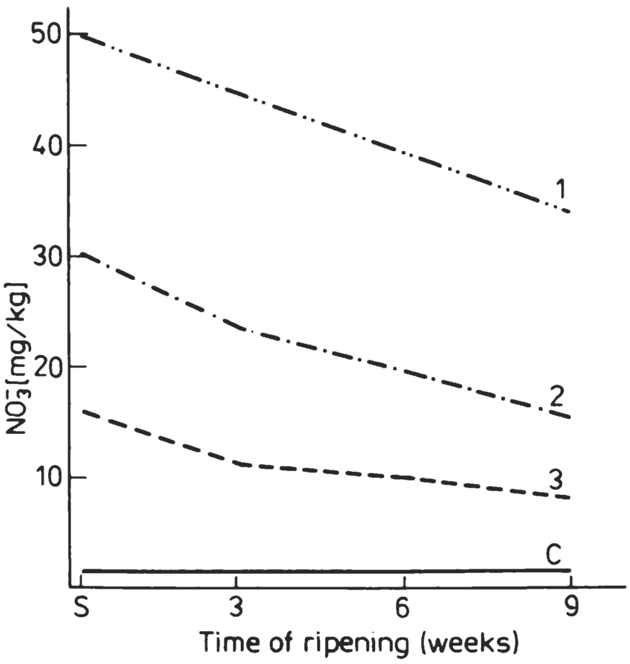

Fig. 1. Changes of nitrates content during ripening of Dutch type cheese with relatively low coliform number; S — cheese after salting; additions of potassium nitrate in g/100 l milk: 1 — 20, 2 — 10, 3 — 5, C (control) — 0

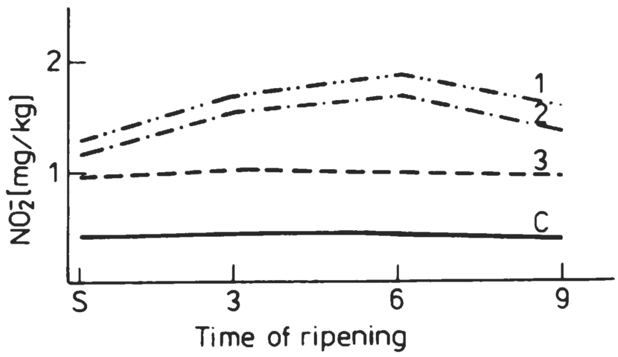

Fig. 2. Changes of nitrites content during ripening of Dutch type cheese with relatively low coliform number. See Fig. 1 for explanations

period of ripening. A characteristic feature of these cheeses was the very slow rate of nitrates reduction during ripening: after six weeks of ripening, the NO_3^- content was at the relatively high level of 25-35 mg/kg of the cheese produced from milk with 0.02% of saltpetre. A similar content of nitrates in Dutch type cheeses produced in good hygienic conditions was found by Goodhead et al. [6]. It is worth noting that the Polish regulations concerning food additives allow a maximum level of 50 mg KNO_3/kg of cheese, which corresponds to 30.6 mg NO_3^-/kg [7]. Six-week Edam cheese produced from milk with 0.01% saltpetre conforms to this requirement.

The content of nitrites in cheeses produced in series 1 experiments was low (1.0-1.5 mg NO_2/kg) which may be accounted for by the low number of coliform bacteria which, as was demonstrated in subsequent experiments, are the main factor responsible for nitrates reduction in cheese.

The magnitude of the saltpetre addition was found to have no effect on the number of coliform bacteria during the entire period of ripening of cheeses produced in good hygienic conditions. This is in agreement with the view that saltpetre is not bactericidal but merely alters the metabolism of bacteria [2, 9, 12].

Organoleptic assessment revealed no signiffcant differences between cheeses produced from milk with different saltpetre additions. Given the small number of coliform bacteria in cheeses from series 1 of experiments, the risk of blowing was minimal.

The conclusions from this series of experiments are that in cheeses produced in good hygienic conditions (coliform number not exceeding 10^3/g) nitrates decomposition is very slow, and saltpetre added to the milk does nothing to limit the adverse effect of coliform bacteria.

In series 2 of experiments, suitable volumes of pasteurized milk with rennet and starter were transferred from industrial tanks to small stainless steel containers in which cheese production was continued with various doses of saltpetre added to the inidividual milk aliquods. The results of this series are listed in Table 2, and the changes in NO_3^- and NO_2^- contents are illustrated in Figs. 3 and 4.

Table 2. Changes of selected features of Dutch type cheese produced in a dairy plant from milk with various additions of saltpetre

KNO_3 addition (g/100 l of milk)	Time of ripening (weeks)	Active acidity pH	Water content (%)	Coli titre	Organoleptic assessment after 6 weeks of ripening (5-point scale)
0	S*)	5.20	44.2	10^{-5}	4.0
	2	5.03	44.0	10^{-5}	
	6	5.26	42.0	10^{-4}	
	10	5.74	40.1	10^{-5}	
10	S	5.00	45.0	10^{-3}	4.0
	2	4.96	45.0	10^{-5}	
	6	5.26	42.3	10^{-5}	
	10	5.64	41.0	10^{-5}	
20	S	5.00	45.0	10^{-4}	4.0
	2	4.96	45.0	10^{-5}	
	6	5.26	42.1	10^{-4}	
	10	5.64	41.0	10^{-4}	

*) S — cheese after salting
The fat content in cheeses after 6 weeks of ripening was 48% of dry matter

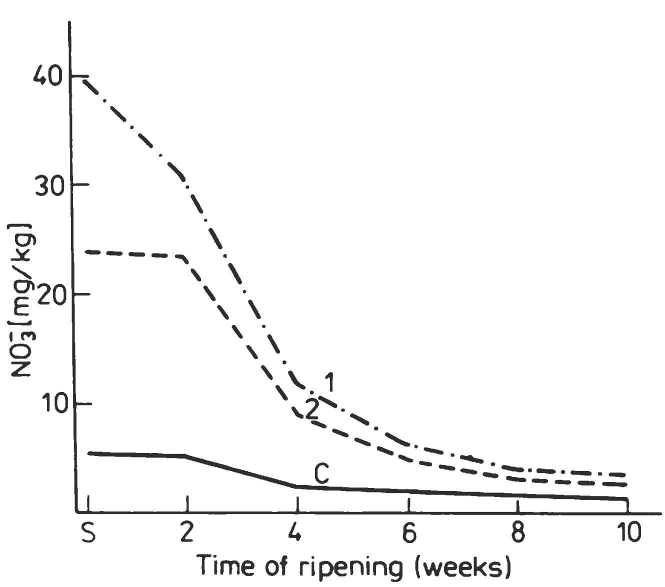

Fig. 3. Changes of nitrates content during ripening of Dutch type cheese with high coliform number. See Fig. 1 for explanations

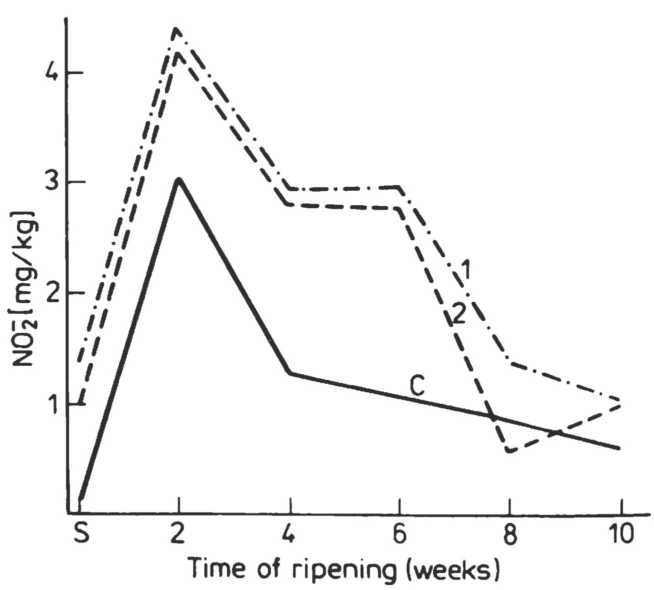

Fig. 4. Changes of nitrites content during ripening of Dutch type cheese with high coliform number. See Fig. 1 for explanations

Nitrates reduction in the series 2 cheeses was very intense with their content dropping to below 10 mg NO_3^-/kg after six weeks of ripening (0.02% saltpetre in the milk). Also in this series of experiments, the nitrates content in the cheese was correlated with the amount of saltpetre added to the milk, with the proportionality being closer in the first six weeks of ripening.

The nitrates decomposition in series 2 cheeses, several times more rapid than in cheeses of series 1, is most probably due to the much greater number of coliform bacteria in the former (10^3-10^5/g) than in the latter (10-10^3/g). The higher coliform number in series 2 cheeses was most probably also responsible for the higher content of nitrates in them during ripening (1.0-4.5 mg NO_2^-/kg of cheese), given the nitrates-decomposition ability of these bacteria [1, 3, 4]. Galesloot [5] claims that the factor responsible for the nitrate-to-nitrite reduction in cheeses is xantine oxidase, but this observation appears to be correct only in the case of cheeses characterized by high hygienic quality. When large numbers of coliform bacteria are present in the cheese, the contribution of xantine oxidase in nitrates reduction is probably small.

The very similar level of nitrites and the similar course of nitrites content changes in cheeses made from milk supplemented with 0.01 and 0.02% of saltpetre may be an indication that the content of these compounds depends on factors other than the nitrate addition to milk. Considering also the fact that the posible effect of saltpetre on cheese quality manifests itself by the action of nitrites formed in situ, one may draw the conclusion that a 0.01 % addition of saltpetre to the milk ought to provide a sufficient inhibition of the gas-forming direction of fermentation, since in favourable conditions the nitrites content is then almost the same as when 0.02% of saltpetre is added to the milk.

This conclusion cannot, of course, be confirmed by results of series 2 of experiments since there were no organoleptically determined differences between saltpetre-containing and control cheeses. It may be that the coliform number in these cheeses was too low for the adverse effect of the bacteria on organoleptic properties of the product to become apparent. This would be in agreement with Sukhotskene et al. [11] who believe that the negative effect of coliform bacteria on cheeses occurs when their number exceeds 10^5/g.

On the other hand, one cannot preclude the advantageous effect of saltpetre on organoleptic properties of the cheeses from this series of experiments, since in control cheeses the nitrites content was also found to be fairly high in the initial period of ripening. The relatively high nitrates content in the initial milk which may sometimes occur (e.g. up to 5 mg/kg) could have sufficed to produce in the control cheeses an amount of nitrites which could already affect the metabolism of coliform bacteria.

In experiment series 3 the cheeses were produced from unprocessed milk of low microbiological quality. The coliform number in these cheeses was 10^5-10^6/g, and nitrates changes in them were several times faster than in cheeses produced from pasteurized milk in the first two series of experiments. As can be seen in Table 3, already after salting the nitrates level is low: about 1, 2, 5 and 16 mg

Table 3. Changes of selected features of Dutch type cheese produced from raw milk with various additions of saltpetre

KNO_3 addition (g/100 l of milk)	Cheese after salting						Cheese after 2 weeks of ripening						
	water content (%)	NO_3^- mg/kg	NO_2^- mg/kg	acidity		coli titre	water*) content (%)	NO_3^- mg/kg	NO_2^- mg/kg	acidity		coli titre	Organoleptic assessment (5-point scale)
				pH	°SH					pH	°SH		
0	45.1	1	0.5	5.32	63.3	10^{-6}	44.0	—	0.3	5.36	64.0	10^{-6}	3.0
5	45.0	2	0.8	5.35	63.6	10^{-6}	44.5		0.3	5.41	64.0	10^{-6}	3.0
10	43.8	5	2.7	5.36	61.3	10^{-5}	43.6	—	0.6	5.41	62.0	10^{-5}	4.0
20	44.4	16	7.9	5.36	62.3	10^{-5}	44.1	3	1.1	5.41	62.0	10^{-5}	4.0

*) Fat content was 45% of dry matter, and salt content was 1.8%

Table 4. Effect of coliform bacteria addition to the milk and different additions of saltpetre on selected characteristics of Dutch type cheese

KNO_3 addition (g/100 l of milk)	Cheese after salting						Cheese after 2 weeks of ripening						
	water content (%)	NO_3^- mg/kg	NO_2^- mg/kg	acidity		coli titre	water*) content (%)	NO_3^- mg/kg	NO_2^- mg/kg	acidity		coli titre	Organoleptic assessment (5-point scale)
				pH	°SH					pH	°SH		
0	45.8	1	0.2	5.39	68.6	10^{-7}	45.1	—	—	5.44	70.3	10^{-6}	3.0
5	45.1	2	0.4	5.39	69.3	10^{-7}	43.7	—	—	5.43	69.6	10^{-6}	3.0
10	45.9	6	1.1	5.43	70.3	10^{-6}	43.3	—	0.1	5.48	70.0	10^{-5}	3.5
20	46.1	22	1.4	5.43	71.3	10^{-6}	44.2	2	0.3	5.48	70.6	10^{-5}	4.0

*) Fat content in the cheese was 45% of dry matter, and salt content was 1.5%

NO_3^-/kg of cheese for saltpetre additions to the milk of 0, 5, 10 and 20 g KNO_3/100 l, respectively. After three weeks of ripening trace amounts of these compounds (2-4 mg NO_3^-/kg) were detectable practically only in cheeses produced from milk with 0.02% of saltpetre.

The nitrites content in cheeses after salting was very high. For the 0.02% addition of saltpetre to milk it was over 10 mg, the mean figure being 7.9 mg NO_2^-/kg of cheese. Such a high content of nitrites is in practice never found in cheese. After three weeks of ripening, the nitrites content dropped to below 1.5 mg NO_2^-/kg.

The very rapid reduction of nitrates in the cheese may have been due not only to the presence of coliform bacteria in the raw milk but also of other nitrates-reducing bacteria groups. The psychrotropic bacteria are particularly effective in this capacity [1].

The coliform number in cheeses produced from milk with higher saltpetre doses (0.01 or 0.02%) was lower both after salting and after three weeks of ripening, this being compatible with the higher nitrites content in them. This may be evidence that large numbers of coliform bacteria or of other bacteria giving rise to large amounts of NO_2^- ions, the bactericidal effect of nitrites becomes apparent. The bactericidal effect of nitrites on coliform bacteria was studied, among others, by Sałek [9].

Photo 1. Cheeses produced from raw milk. The figures on the cheeses denote KNO_3 additions in g/100 l of milk

The organoleptic quality of cheeses from raw milk with a higher saltpetre additions was clearly superior to the analogous quality of control cheeses. A manifestation of this quality was mainly the inhibition of the intense abnormal holes formation (cf. Photo 1).

In experiments of series 4, cheeses were produced from pasteurized milk supplemented with a culture of coliform bacteria. These too were characterized by very intense transformation of nitrates and nitrites. The drop in the content of these compounds was even faster than in raw milk cheeses.

The nitrites content in these cheeses after salting did not exceed 1.4 mg NO_2^-/kg on average. It may be surmised that given the large number of coliform bacteria in the cheese (of the order of 10^7/g) the rate of nitrates and nitrites reduction was so high already before the end of salting that there was no accumulating of nitrites to the level observed in the raw milk cheeses of the third series of experiments. After two weeks of ripening, the series 4 cheeses contained only trace amounts of nitrates and nitrites.

After salting, the coliform number in cheeses produced from milk with the higher (0.01 and 0.02%) saltpetre additions was from 10 to 100 times lower than in cheeses with 0.005% or no saltpetre additions. After two weeks of ripening the differences were not as great. The fact remains, however, that when the number of coliform bacteria is high, a saltpetre addition inhibits their development, this being probably due to the temporarily increased content of the appearing nitrites which may then demonstrate their bactericidal capabilities [10].

Regarding organoleptic qualities, here too, like in the case of cheeses produced from raw milk, the cheeses with higher saltpetre additions were superior. The differences in hole formation in the cheeses from this experiments series can be seen in Photo 2.

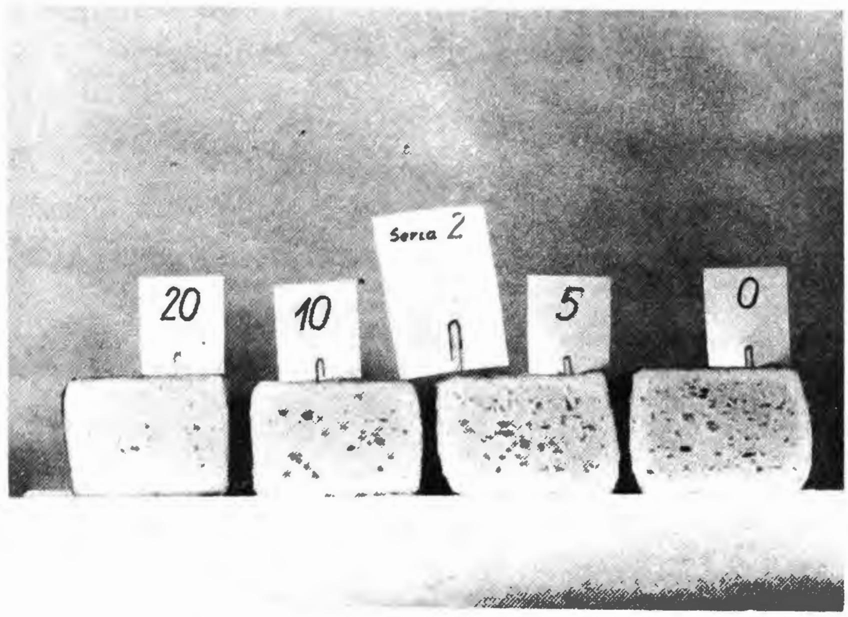

Photo 2. Cheeses produced from pasteurized milk supplemented with culture of coliform bacteria. The figures on the cheeses denote KNO_3 additions in g/100 l of milk

Generally speaking, the positive effect of saltpetre additions to the milk on cheese quality was not apparent when the coliform number was low both in the milk and in the cheese produced from it. When the coliform number in the cheese was over 10^5/g, the population of these bacteria was reduced after salting, and the quality of the cheeses improved when 0.01 or 0.02% of saltpetre was added to the milk.

CONCLUSIONS

1. The rate of nitrates decomposition in cheese depends mainly on the coliform number. In cheese with this number below 10^3 g, nitrates reduction is very slow. After nine weeks of ripening, a 20-30% drop in nitrates content was observed in such cheeses produced from milk with 0.02% of KNO_3.

2. The content of nitrites in ripening cheese depends only slightly on the amount of saltpetre added to the milk. This content increases with the increase of the coliform number, but only up to a point; when the coliform bacteria become very numerous, both the nitrates and the nitrites vanish rapidly.

3. The saltpetre addition to the milk was not found to affect the number of coliform bacteria in the cheeses if their content was not higher than 10^5 g. In cheeses with a higher content of these bacteria, greater saltpetre additions to the milk were found to reduce their number.

4. No organoleptic differences were found in cheeses produced from milk with various additions of saltpetre when the coliform number was below 10^5 g. A positive effect of the saltpetre addition to milk was apparent in cheeses strongly contaminated with coliform bacteria.

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WSPÓŁZALEŻNOŚĆ MIĘDZY ZRÓŻNICOWANYM DODATKIEM SALETRY DO MLEKA, ZAWARTOŚCIĄ AZOTANÓW I AZOTYNÓW W SERZE, JEGO JAKOŚCIĄ I LICZEBNOŚCIĄ BAKTERII Z GRUPY COLI

Katedra Produktów Białkowych i Tłuszczowych, SGGW-AR, Instytut Mleczarstwa, Warszawa

Streszczenie

Wykonano cztery serie doświadczeń, w których prowadzono wyrób sera edamskiego z różnym dodatkiem saletry do mleka, tj. 0, 5, 10 i 20 g KNO_3 na 100 l mleka (po 3 wyroby w każdej serii). Mleko używane w serii 1, 2 i 4 pasteryzowano w temp. 74°C $\pm$ 1 przez 10-15 s, zaś w serii 3 użyto do wyrobu mleko surowe. W serii nr 4 do mleka dodawano namnożoną hodowlę bakterii z grupy coli.

Stwierdzono, że szybkość rozkładu azotanów w serze zależy głównie od liczebności bakterii z grupy coli. Przy zawartości tych bakterii wynoszącej 10^5-10^6/g sera całkowity rozkład azotanów następował już po 2 tygodniach dojrzewania (przy dodatku 0,02% KNO_3 do mleka) podczas gdy w serach zawierających nie więcej niż 10^3 bakterii/g nawet po 9 tygodniach spadek zawartości azotanów nie przekraczał 20-30% (rys. 2). Zawartość azotynów w serze w czasie dojrzewania w niewielkim stopniu zależna była od wielkości dodatku saletry do mleka serowarskiego, natomiast wzrastała do pewnej granicy wraz ze wzrostem liczby bakterii z grupy coli. Przy bardzo dużej liczebności bakterii coli zanikają szybko zarówno azotany jak i azotyny. Nie stwierdzono wpływu dodatku saletry do mleka na liczebność bakterii z grupy coli w serze, jeśli ich zawartość nie przekraczała 10^5/g sera. Natomiast przy silnym zakażeniu mleka serowarskiego tymi bakteriami obserwuje się niższą ich zawartość w serze przy wyższym dodatku saletry do mleka (tab. 3 i 4). Nie stwierdzono różnic w ocenie organoleptycznej serów otrzymanych przy różnym dodatku saletry do mleka, jeśli liczebność bakterii z grupy coli była niższa niż 10^5/g sera. Korzystny wpływ dodatku saletry do mleka na cechy organoleptyczne sera ujawnił się natomiast w przypadku serów silniej zakażonych ww. bakteriami.