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Fish processing technology

**EFFECTS OF GLUTAMIC-LACTIC ACID MIXTURE AND ITS  
COMBINATION WITH ASPARGIC ACID AND WITH ALANINE ON  
MATURATION RATE OF SALTED BALTIC HERRING**

**WPLYW MIESZANINY KWASU GLUTAMINOWEGO I KWASU  
MLEKOWEGO ORAZ JEJ POŁĄCZENIA Z KWASEM  
ASPARAGINOWYM LUB ALANINĄ NA SZYBKOŚĆ DOJRZEWANIA  
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Glutamic-lactic acid mixture and its combination with aspartic acid and with alanine were used to obtain effect on maturation rate of salted Baltic herring. Those substances were added in amount of 0.17% each per fish weight. The maturation of salted herring carcasses was evaluated organoleptically and by chosen chemical indices such as non-protein and amine nitrogen content. Results proved the carcasses salted with glutamic-lactic acid mixture and with combination of those acids and alanine were mature after 4 weeks, while treated with the mixture of aspartic, glutamic and lactic acids after 6 weeks. The carcasses treated with the salt only did not mature during 10 weeks of the study period.

INTRODUCTION

Salted fish products, those weakly salted (up to 10% NaCl in meat) in particular, are in great demand among consumers. However, the deficiency of the traditional raw materials for such products makes it necessary to look for technological solution which would turn the available fish species into salted products of flavour characteristics typical of properly matured salted fish. Maturation of salted fish is an enzymatic process, for which reason meat of some fish species, influenced mainly by its own enzymes (Kosova 1969; Knochel and Huss 1984) acquires desirable properties without the need for thermal treatment.

One of quality optimisation methods in fish salting is the use of proteolytic enzymes, mainly of microbiological origin (Konstantinova and Pachomova 1970; Szołtysek et al

1984; Chabowska and Wawerek 1986) or of preparations obtained from viscera of fish properly maturing during salting (Lisovaja and Nechamkina 1976; Sluckaja et al. 1983). In addition, maturation can be also stimulated by increasing the activity level of some native enzymes. Šenderiuk and Chlopkova (1973) reached this effect by using hydrochloric acid, whereas Jasińska (1989), in her studies on optimisation of Baltic herring salting, applied lactic acid which, apart from acidifying the enzymes' medium, favourable affected the flavour and shelf life of the product.

As a result of enzymatic decomposition of muscle tissue proteins of salted fish, the content of low molecular weight nitrogen compounds, mainly amino acids and peptides, increases. The muscles tissue of mature salting herring was also found (Kiesvaara 1975) to contain considerable amounts of alanine and glutamic acid. Chlopkova and Šenderiuk (1973) as well as Plorinia and Leonova (1970) demonstrated a considerable increase in aspartic acid to take place in the properly maturing salted herring. Addition of glutamic-lactic acid mixture, too, was found to accelerate maturation of salted herring (Jasińska, 1989).

The data reported by the authors referred to above and those produced during the author's own research were used to obtain effects of glutamic-lactic acid mixture in combination with aspartic acid and with alanine on maturation rate of salted Baltic herring.

## MATERIAL AND METHODS

The raw material used was the Baltic herring individuals 25–29 cm long and weighing 130–190 g. They were at maturity stage V (Maier's scale).

The chemical composition of fish used for salting was as follows: 83.1%—water, 14.7%—protein, 2.4%—fat.

The fish were processed to carcasses and were divided into 4 batches 4 kg each. They were salted in stoneware containers using identical amount of salt 400 g / 4 kg fish in each case. The first batch was treated with salt only and was set aside as a control. The second batch was treated with mixture of glutamic and lactic acids (0.25% of each substance as per fish weight). The third batch consisted of herring salted with salt enriched with a mixture of glutamic, lactic and aspartic acids (0.17% of each as per fish weight). The fourth batch was treated with salt enriched with glutamic and lactic acids and alanine (0.17% of each compound as per fish weight). The batches thus prepared were stored at 5°C ( $\pm 1^\circ$ ) for 10 weeks. The fish were sampled after 2, 4, 6, 8 and 10 weeks of storage for the organoleptic assessment and chemical assays. Minces were prepared from skinned and filleted carcasses to be used in further chemical assays. The following assays were made in TCA extracts obtained in two stage method according to Kořakowski (1973):

—non-protein nitrogen content, Kjeldahl technique,

—amine nitrogen, Pope-Stevens method in Fik's (1979) modification.

A number of additional assays included:

—total nitrogen content, Kjeldahl method,

—water content, drying at 105°C for 3 h,

—NaCl content, Volhard method, after salting process was terminated,

—muscle tissue pH, potentiometrically.

The organoleptic evaluation was carried out by a panel using a 5 grade scale and taking the following quality indices into account: appearance, texture, taste, smell and desirability of the product. The organoleptic assessment criteria are presented in Table 1. The results were treated statistically with Student's *t* test.

Table 1

Organoleptic assessment criteria

Score	Quality Factor				
	Appearance	Texture	Taste	Smell	Desirability of the product
5	very good	very delicate, juicy	fully mature	very good	highly desirable
4	good	tender, juicy	mature	good	desirable
3	sufficient	cohesive, juicy	maturing	sufficient	not fully desirable
2	acceptable	tough	half-raw	acceptable	desirable to some degree
1	not acceptable	strong tough, dry	raw	not acceptable	definitely undesirable

## RESULTS AND DISCUSSION

The muscle tissue NaCl content after salting (2 weeks) ranged from 7.4% to 8.0%. The organoleptic evaluation (Tab. 2) showed a positive effect of the additives used on maturation rate and on the flavour properties of the salted fish. Those carcasses treated with salt only (the control) did not acquire properties of a mature product during the whole period of study (10 weeks). The carcasses salted with the addition of the glutamic-lactic acid mixture and with the combination of those acids and alanine were mature after 4 weeks, while treatment with the mixture of aspartic, glutamic and lactic acids yielded a mature product after 6 weeks. The fish treated with salt only showed symptoms of spoilage and they were assessed as a definitely non-desirable product. On the other hand, the fish salted with the additives and kept for the same period of time were of a very good quality: the carcasses salted with addition glutamic-lactic mixture scored highest; the fish salted with the mixture of both acids combined with alanine, scored high as well, while slightly lower scores were given to the fish salted with the mixture of aspartic, glutamic and lactic acids as the mixture caused the meat to brighten much more than was the case with other additives.

Table 2

Scores of organoleptic assessment of salted herring carcasses

Quality factor	Kind of sample and salting time (weeks)																			
	Control					Glutamic-lactic acid mixture					Aspartic, glutamic and lactic acids mixture					Glutamic, lactic acids and alanine mixture				
	2	4	6	8	10	2	4	6	8	10	2	4	6	8	10	2	4	6	8	10
Appearance	5	5	4.5	4	2.4	4.5	4.2	4	4	3.8	4.6	4	3.8	3.5	3.5	5	4.5	4.5	4	4
Texture	3	3	3	3.4	—	3.5	3.9	4	4.2	4	3	3.5	4	4	4 M	3	4	4	3.6	4
Taste	2	2.5	2.6	2.4 S*	—	3	4	4.5	5	5	3.5	3.6	4	4	4	3.1	5	4.5	4	4.5
Smell	2	2.3	3	2 S*	1.8 S**	2.8	2.6	3	3.3	3.5	3	2.5	3	3	3	2.7	3	3	3	3.6
Desirability of the product	2	2.2	2.4	2	1	3	4	4.5	5	5	3	3.5	4	4	4.5	3	4.7	4	4	4.5

S—musty-putrid; \*—weakly sensible; \*\*—clear; M—buttery texture; the other explanations see Tab. 1.

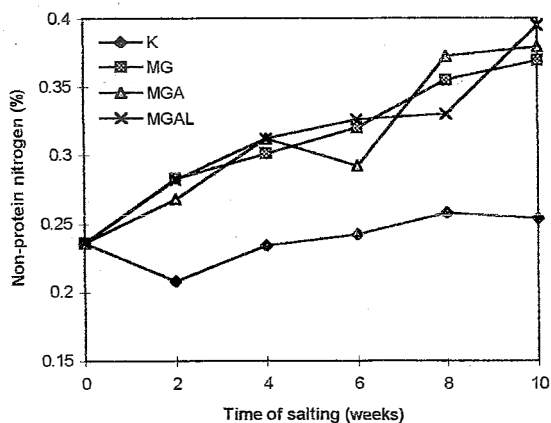


Fig. 1. Effects of glutamic-lactic acid mixture and its combination with aspartic acid and with alanine on non-protein nitrogen content in the salted herring muscle tissue. K—control, MG—mixture of glutamic and lactic acids, MGA—mixture of aspartic, glutamic and lactic acids, MGAL—mixture of glutamic and lactic acid and alanine.

the fish salted with the addition of glutamic and lactic acids showed an about 36% increase; an about 24% increase was recorded in the fish salted with the mixture of aspartic, glutamic and lactic acids, whereas in fish salted with salt only a 2.5% increase only was observed.

Differences in the non-protein nitrogen content of the muscle tissue of fish treated with salt only and those salted with additives were significant and highly significant, whereas differences between various treatments involving additives were non-significant (Tab. 3).

Table 3

Results of pair-wise comparisons with Student's *t*-test non-protein nitrogen content in the herring carcasses muscle tissue salted with glutamic-lactic acid mixture and its combination with aspartic acid and with alanine

Reference sample	Sample under comparison		
	Glutamic-lactic acid mixture	Aspartic, glutamic and lactic acids mixture	Glutamic, lactic acids and alanine mixture
Control	4.4860a**	3.8192*	4.0823**
Glutamic-lactic acids mixture		0.1173	0.4132
Aspartic, glutamic and lactic acids mixture			0.3501

a—parameter *t*,

\*— $\alpha \leq 0.05$ ,

\*\*— $\alpha \leq 0.01$ .

The organoleptic changes found were reflected in the chemical processes. The muscle tissue non-protein nitrogen content (Fig. 1) in the fish salted with additives was much higher than that in tissues treated with salt only during the whole period of study. After 6 weeks of storage, when the fish salted with additives were mature, their non-protein nitrogen content increased by 38% in the fish salted with the mixture of glutamic and lactic acids with alanine;

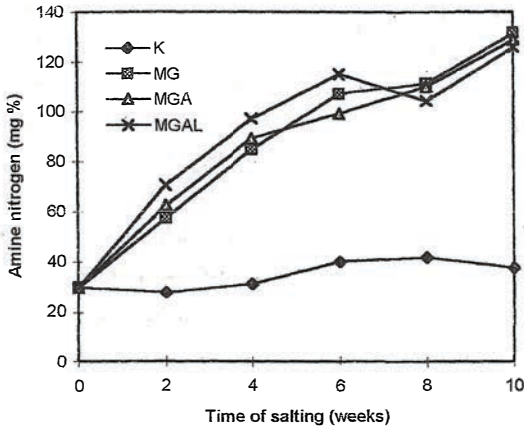


Fig. 2. Effects of glutamic-lactic acid mixture and its combination with aspartic acid and with alanine on amine nitrogen content in the salted herring muscle tissue  
For explanations see Fig. 1.

The amine nitrogen content (Fig. 2), similarly to non-protein nitrogen, was much higher in the fish salted with additives than in those treated with salt only over the whole period of study. During storage, the amine nitrogen content increased in all batches; however it was clearly higher in the fish salted with additives than in the salt - only treatment.

After 6 weeks of storage, when the fish salted with additives were mature, the amine nitrogen content in the control was by about 35% higher than the initial level, whereas an almost 4-fold increase was recorded in the batch salted with the mixture of glutamic and lactic acids with alanine, an about 3.5-fold increase occurred in the glutamic-lactic acid acid treatment and about 3.3-fold increase was observed where the mixture of aspartic, glutamic and lactic acids was applied.

Differences in the amine nitrogen content between—the control and the fish salted with additives were significant and highly significant. On the other hand, differences in this form of nitrogen between different treatments with additives were non-significant (Tab. 4).

Table 4

Results of pair-wise comparisons with Student's *t*-test amine nitrogen content in the herring carcasses muscle tissue salted with glutamic-lactic acid mixture and its combination with aspartic acid and with alanine

Reference sample	Sample under comparison		
	Glutamic-lactic acid mixture	Aspartic, glutamic and lactic acids mixture	Glutamic, lactic acids and alanine mixture
Control	3.8628a*	4.0622**	4.3789**
Glutamic-lactic acid mixture		0.2458	0.9432
Aspartic, glutamic and lactic acids mixture			1,1780

For explanations see Tab. 3.

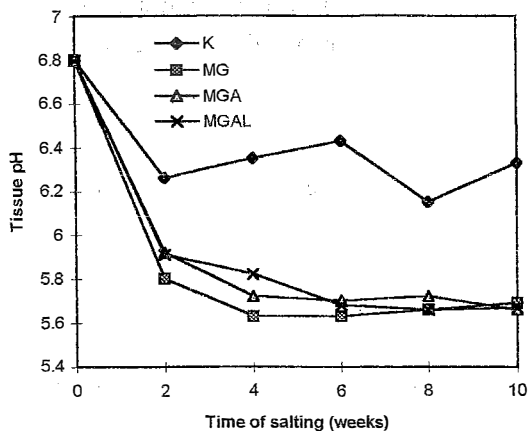


Fig. 3. Effects of glutamic-lactic acid mixture and its combination with aspartic acid and with alanine on salted herring muscle pH  
For explanations see Fig. 1.

than that of the experimental treatments and generally did not drop below 6.15 over the whole period of study (10 weeks).

This study demonstrate a positive effect of the additives used on maturation rate and flavour of salted fish, which is the more important as the control failed to acquire properties of a mature product during the whole period of study (10 weeks) and symptoms of spoilage appeared at the last stage of storage. The organoleptic changes found were related to the increase in non-protein nitrogen fractions in the salted herring. Higher levels of both non-protein and amine nitrogen in the fish salted with additives during the initial period of storage was probably associated with diffusion of amino acids introduced into the tissue with salt. In the later period of storage, a higher increase in non-protein nitrogen fractions might have resulted from increased activity of proteolytic enzymes of the muscle tissue as a result of pH reduction caused by the additives used. A positive effect of acidification on proteolytic enzyme activity in i.a. muscle tissue reported too (Šenderiuk and Chlopkova 1973).

Glutamic and lactic acids have antidenaturation properties (Noguhi and Matsumoto 1975a, 1975b), due to which they can protect the structure of proteins and proteolytic enzymes enhance their activity in the presence of salt.

Prolongation of the shelf life of those carcasses salted with additives was observed as compared to the control. It can be assumed that lactic acid plays an important role here as it has some bactericidal and bacteriostatic properties and lowers pH of the medium, whereby conditions less favourable for putrid bacteria occur (Zaleski 1985).

Aspartic, glutamic acids and alanine used in salting are amino acids which occur in significant amounts in the meat of properly maturing salted herring (Kiesvaara 1975;

The additives used in salting caused muscle tissue pH to decrease (Fig. 3). During the first 4 weeks of salting, meat pH decreased most in the mixture of glutamic and lactic acids and least in the mixture of these acids plus alanine. Further on during storage, the meat pH of all products was at maintained a similar level of 5.63–5.7. On the other hand, the meat pH of the control was clearly higher

Šenderiuk 1976), while glutamic acid is regarded as a flavour modulator (Tyszkiewicz 1973). Thus it can be assumed that addition of those amino acids enhanced taste properties of the salted product.

### CONCLUSIONS

1. Herring carcasses salted with salt only did not acquire properties of a mature product during the whole period of study (10 weeks), whereas the fish salted with additives (a mixture of glutamic and lactic acids and a mixture of both acids and alanine) were mature after 4 weeks of storage, and the fish salted with addition of aspartic, glutamic and lactic acids were mature after 6 weeks.
2. In the muscle tissue of those carcasses salted with addition of glutamic and lactic acid mixture and with combination of this mixture and alanine or aspartic acid, much higher contents of non-protein and amine nitrogen was found than in the carcasses salted with salt only.
3. The pH of the fish salted with addition of a glutamic and lactic acid mixture and with combination with alanine or aspartic acid was clearly lower than that of the fish salted with salt only.
4. A mixture of glutamic and lactic acids and their combination with aspartic acid and alanine can be used in the industrial practice of salting fish carcasses.

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## WPŁYW MIESZANINY KWASU GLUTAMINOWEGO I KWASU MLEKOWEGO ORAZ JEJ POŁĄCZENIA Z KWASEM ASPARAGINOWYM LUB ALANINĄ NA SZYBKOŚĆ DOJRZEWANIA SOLONYCH ŚLEDZI BAŁTYCKICH

### STRESZCZENIE

Jednym ze sposobów optymalizacji jakości solonych produktów, na które istnieje wciąż duże zapotrzebowanie wśród konsumentów, jest zastosowanie w procesie solenia jako dodatków substancji, których zawartość zwiększa się w tkance mięśniowej prawidłowo dojrzewających ryb solonych lub też substancji powodujących podwyższenie aktywności enzymów własnych solonych ryb. Opierając się na powyższych założeniach postanowiono określić wpływ dodatków mieszaniny kwasów glutaminowego i mlekowego w połączeniu z kwasem asparaginowym lub alaniną na szybkość dojrzewania śledzi bałtyckich oprawionych do postaci tuszek. Użyte substancje dodano w ilości 0,17% w stosunku do masy przeznaczonych do solenia ryb. Stopień dojrzłości solonych tuszek śledziowych

określano na podstawie oceny organoleptycznej, wskaźnikiem zachodzących przemian enzymatycznych była również zawartość azotu niebiałkowego i azotu aminowego w tkance mięśniowej solonych ryb.

Przeprowadzone badania wykazały pozytywny wpływ zastosowanych dodatków na cechy smakowe, szybkość dojrzewania i trwałość solonych tuszek śledziowych. Ryby solone samą solą nie uzyskały cech dojrzałego produktu przez cały okres trwania badań (10 tygodni), tuszki solone z mieszaniną kwasów glutaminowego i mlekowego oraz mieszaniny tych kwasów z alaniną były dojrzałe po 4 tygodniach, a z dodatkiem mieszaniny kwasów asparaginowego, glutaminowego i mlekowego po 6 tygodniach solenia. Również zawartość azotu niebiałkowego i azotu aminowego była znacznie większa w rybach solonych z dodatkami niż samą solą, a występujące różnice były statystycznie istotne. Zastosowane substancje powodowały także obniżenie pH tkanki mięśniowej solonych ryb, wpływające na przyspieszenie dojrzewania i przedłużenie trwałości solonych śledzi.

Uzyskane wyniki wskazują na możliwość praktycznego wykorzystania mieszaniny kwasów glutaminowego i mlekowego w połączeniu z kwasem asparaginowym lub alaniną podczas solenia tuszek śledziowych.

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