А.М. Майлыбаева, Р.Ж. Мыктыбаева, К.К. Табынов, Ш.Ж. Рыскелдинова

ИЗУЧЕНИЕ БИОЛОГИЧЕСКОЙ АКТИВНОСТИ ШТАММА ВИРУСА ИНФЕКЦИОННОГО БРОНХИТА ПТИЦ «Н-120»

По результатам исследований на инфекционный бронхит птиц вводили вирусный штамм «Н-120» в дозе 1000 lgЭИД$_{50}$/0,2см^3 в аллантойсную жидкость 10-дневного эмбриона, что показало ее биологическую активность 5,78±0,08 lgЭИД$_{50}$/см^3.

При вводимой дозе 100000 ее биологическая активность составила 6,70±0,14 lgЭИД$_{50}$/см^3.

Соответственно при вводимой дозе 600000 lgЭИД$_{50}$/0,2см^3 биологическая активность составила 6,87±0,08 lgЭИД$_{50}$/см^3, где смертность эмбрионов увеличилось.

Считаем самой высокой биологической активностью является вводимая доза вируса 10000 ЭИД$_{50}$, 0,2 см^3, активность которого составляет 7,03±0,08

Ключевые слова: Coronavirus avia, Coronaviridae, E. Coli, инфекция, эмбрион, «Н-120», «Н-52», «АМ», аллантоисная жидкость, биологическая активность.

A.M. Mailibaeva, R. Zh. Myktybayeva, K.K. Tabynov, Sh. Zh.Ryskeldinova

RESEARCH OF BIOlOGICAL ACTIVITY STAMM “H-120” VIRUSES OF INFECTION BRONCHITIS OF BIRDS.

Results of research for infection bronchitis of birds injection viruse stamm “H-120” in dose 1000 lg EID$_{50}$/0,2 cm^3 in allontoise solution for 10 days embrion shows biological activity 5,78±0,08 lgEID$_{50}$/0,2 cm^3.

For injection dose 100000 biological activity shows 6,70±0,14 lgILD$_{50}$/0,2 cm^3.

Among injection dose 600000 lgEID$_{50}$/0,2 cm^3 biological activity shows 6,87±0,08 lgEID$_{50}$/0,2 cm^3 death embrions increases.

Biological activity injection dose of viruse 1000 lgEID$_{50}$/0,2 cm^3 activity shows 7,03±0,08.

Key words: Coronavirus avia, Coronaviridae, E. Coli, infection, embryos, «Н-120», «Н-52», «АМ», allantoisny liquid, biological active.

UDC 619.381:615.31

A.E. Paritova, N.B. Sarsembayeva

Kazakh National agrarian university

THE INFLUENCE OF ZEOFISH ON THE FATTY ACIDS PROFILES OF THE FISH

This article presents the results of the influence of zeolites as feed additives on the fatty acids profile of fish. Research was conducted for 63 days using rainbow trout from Turgen village (Kazakhstan). The studied material was zeolitic tuff from the Chankanay deposit as an additive to RGM-2M feed. They were fed with normal diet, and normal diet supplemented with 1%, 2%, 3% and 4% of natural zeolites. Additionally, lipid contents and FA compositions were studied. Amino acid compositions were studied. The results of this study confirmed that zeolites have a positive effect on the fatty acid composition.

Key words: zeolite, feed additives, Oncorhynchus mykiss, fatty acid composition

Introduction

Zeolites are crystalline, hydrated aluminosilicates of alkali and alkaline earth cations, consisting of threedimensional frameworks of SiO_4 and AlO_5 tetrahedra linked through the shared oxygen atoms. Both natural and synthetic zeolites are porous materials, characterized by the ability to lose and gain water reversibly, to adsorb molecules of appropriate cross-sectional diameter (adsorption property, or acting as molecular sieves) and to exchange their constituent cations without major change of their structure (ion-exchange property) [1,2]. The exploitation of these properties underlies the use of zeolites in a wide range of industrial and agricultural applications and particularly in animal nutrition since mid-1960s [3].

Intensive aquaculture continues to expand, which requires high quality protein sources. Fish meal is major and increasingly expensive component of salmon and trout feeds, since it has high levels of digestible protein and energy, excellent amino acid and fatty acid profiles. CLNP as feed additive can improve the effects of feed [4].

The lipid contents and fatty acid compositions (% of total fatty acids) of rainbow trout fed with four different ratios of clinoptilolite were studied. The fatty acid compositions of fish in groups ranged from 26.81% to 27.93 % saturated fatty acids, 25.35–31.435 % monounsaturated and 32.99–40.185 % polyunsaturated. Among them, those occurring in the highest proportions were oleic acid (C18:1*n9*, 19.85-22.27 %), palmitic acid (C16:0, 15.60-16.56 %), linoleic acid (C18:2*n6*, 11.43-18.88 %), cis-4,7,10,13,16,19-docosahexaenoic acid (C22:6*n3*, 13.36-15.52 %), stearic acid (C18:0, 4.25-4.75 %), palmitoleic acid (C16:1, 3.53-4.59 %), cis-5,8,11,14,17-eicosapentaenoic acid (C20:5 *n3*, 3.11-3.39 %), and myristic acid (C14:0, 2.56-2.85 %). The findings demonstrated that fatty acid compositions of the groups depend on feed, age, environmental conditions, and effects of feed additives like clinoptilolite. In this respect, it demonstrated that clinoptilolite can be added to fish feeds [3].

The purpose of the research is to analyze the fatty acids profile composition of fish meat while using Zeofish in their diet as feed additives.

Materials and methods

Investigations were carried out at the Department of Veterinary-Sanitary Examination and Hygiene, at the Kazakhstan-Japan Center of the Kazakh National Agrarian University and at the trout farm in Turgen village during 2011-2013 years.

Fatty acids analysis of fish meat

The fatty acid profile of trout samples was determined as fatty acid methyl esters (FAMEs). Prior to analysis, the head, tail, fins, and viscera of the fish were removed. The edible tissue was filleted with the skin left on and homogenized. Fish samples were prepared using direct saponification with KOH/methanol followed by a derivatization with (trimethylsilyl) diazomethane by the method of Aldai et al. (2006).

FA composition was analyzed by GC Agient with an autosampler (Agilent HP 6890 N, USA) equipped with a flame ionization detector and a Supelco-SP-2330 fused silica capillary column (30 m, 0.25 mm i.d., 0.20 mm film thickness of polyethylene glycol) (Bellefonte, PA). The oven temperature was 140 °C, held at 5 min, raised to 200 °C at a rate of 4 °C/min and to 220 °C at a rate of 1 °C/min, while the injector and the detector temperature were set at 220 °C and 280 °C, respectively. The sample size was 1 μl and the carrier gas was controlled at 16 psi. The split used was 1:100. The individual FAMEs (fatty acid methyl esters) were identified according to similar peak retention times using standard mixture Supelco 37 Component FAME Mix.

All data were subjected to one-way variance analysis (ANOVA) using the Statistica 8.0 software environment to test the effects of the experimental diets. Duncan's multiple range test and critical ranges were used to test differences among the individual means. The differences were regarded as significant when $P<0.05$. All of the results are expressed as the means±S.D.

Results and discussion

It is known that fish meat does not contain significant amounts of lipids, however the study of its fatty acid content is of significant interest, not from the perspective of determining its biological value, but as the indicator whose change is indicative of cell abnormalities in biochemical processes. It was found that zeolite mainly increases the level of polyunsaturated fatty acids in fish meat.

The biochemical composition may be affected depending on the species of fish, environmental factors, size, age, and diet Fish can be a source of essential fatty acids. In this study, FA contents (% of total FAs) in rainbow trout fed with RGM-2M with zeolite from experimentals and control groups was given in Table 3.

Of the fatty acids analysed, there were higher and lower levels of fatty acids in experimental fish than in the control group.

The FA contents of fish in control and experimental groups ranged from 26.25% to 27.37% saturated fatty acids (SFAs), 30.04–31.29% monounsaturated (MUFAs) and 35.44–36.71% PUFAs (Table 1).

Table 1

Profile of fatty acids in lipids in muscle tissue of rainbow trout (control and experimental group, % of total fatty acid).

Fatty acid composition, g 100 g^{-1}	Control group (n =20)	Experimental group			
		1%	2%	3%	4%
C 12:0 (Lauric acid)	0,04±0.03^{a}	0,04±0.02^{a}	0,05±0.02^{a}	0,05±0.02^{a}	0,04±0.02^{a}
C 13:0 (Tridecanoic acid)	0,02±0.02^{a}	0,01±0.04^{a}	0,03±0.02^{a}	0,02±0.02	0,01±0.023^{a}
C 14:0 (Miristic acid)	3,27±0.06^{a}	3,85±0.07^{a}	3,76±0.03^{a}	3,72±0.03^{a}	3,45±0.02^{a}
C 15:0 (Pentadecanoic acid)	0,37±0.03^{a}	0,37±0.05^{a}	0,37±0.04^{a}	0,36±0.05^{a}	0,36±0.03^{a}
C 16:0 (Palmitic acid)	15,99±0.05^{a}	16,16±0.08^{a}	16,05±0.07^{a}	16,22±0.10^{a}	15,34±0.08^{a}
C 17:0 (Heptadecanoic acid)	0,68±0.02^{a}	0,58±0.03^{a}	0,53±0.05^{a}	0,52±0.02^{a}	0,48±0.03^{a}
C 18:0 (Stearic acid)	3,42±0.13^{a}	3,25±0.02^{a}	3,44±0.04^{a}	3,15±0.04^{a}	3,25±0.05^{a}
C 20:0 (Arachidic acid)	0,22±0.03^{a}	0,21±0.02^{a}	0,16±0.02^{a}	0,23±0.02^{a}	0,21±0.02^{a}
C 22:0 (Behenic acid)	1,54±0.14^{a}	1,26±0.05^{a}	1,32±0.05^{a}	1,48±0.05^{a}	1,53±0.04^{a}
C 23:0 (Tricosanoic acid)	0,04±0.02^{a}	0,04±0.06^{a}	0,03±0.03^{a}	0,04±0.02^{a}	0,04±0.02^{a}
C 24:0 (Lignoceric acid)	1,35±0.02^{a}	1,40±0.03^{a}	1,44±0.04^{a}	1,25±0.05^{a}	1,54±0.03^{a}
Total SFAs	**26,94**	**27,17**	**27,18**	**27,04**	**26,25**
Monounsaturated Fatty Acids					
C 14:1 (Myristoleic acid)	0,23±0.06^{a}	0,23±0.04^{a}	0,24±0.03^{a}	0,23±0.03^{a}	0,21±0.02^{a}
C 16:1 (Palmitoleic acid)	5,30±0.02^{a}	5,29±0.08^{a}	5,25±0.05^{a}	5,36±0.04^{a}	5,95±0.02^{a}
C 17:1 (cis 10 –heptadecenoic acid)	0,26±0.06^{a}	0,23±0.02^{a}	0,21±0.02^{a}	0,23±0.02^{a}	0,35±0.03^{a}
C 18:1 n9 (Oleic acid)	20,98±0.09^{ab}	20,27±0.11^{ab}	21,72±0.10^{ab}	21,52±0.07^{ab}	20,08±0.08^{ab}
C 20:1 (cis -11- eicosenoic acid)	2,43±0.02^{a}	2,41±0.05^{a}	2,35±0.04^{a}	2,41±0.03^{a}	2,41±0.02^{a}
C 24:1 (Nervonic acid)	1,46±0.02^{a}	1,61±0.06^{a}	1,52±0.06^{a}	1,53±0.02^{a}	1,48±0.02^{a}
Total MUFAs	**30,66**	**30,04**	**31,29**	**31,28**	**30,48**
Polyunsaturated Fatty Acids					
C 18:2 n6 (Linoleic acid)	11,06±0.02^{a}	11,11±0.02^{a}	11,24±0.03^{a}	11,35±0.02^{a}	11,45±0.03^{a}
C 18:3 n6 (γ-linolenic acid)	0,29±0.03^{b}	0,42±0.02^{b}	0,35±0.02^{b}	0,28±0.02^{b}	0,26±0.02^{b}
C 18:3 n3 (Linolenic acid)	1,86±0.02^{a}	1,99±0.05^{a}	1,89±0.04^{a}	1,94±0.03^{a}	1,86±0.04^{a}
C 20:2 (cis-11,14-eicosadienoic acid)	0,58±0.02^{b}	0,50±0.03^{b}	0,45±0.04^{b}	0,55±0.04^{b}	0,65±0.04^{b}
C 20:3 n6 (cis-8,11,14-eicosatrienoic acid)	0,24±0.07^{a}	0,23±0.07^{a}	0,24±0.07^{a}	0,21±0.06^{a}	0,23±0.07^{a}

C 20:3 n3 (cis-11,14,17-eicosatrienoic acid)	0,58±0.02[b]	0,58±0.02[b]	0,51±0.02[b]	0,56±0.02[b]	0,58±0.02[b]
C 20:4 n6 (Arachidonic acid)	0,87±0.02[a]	0,69±0.07[a]	0,79±0.07[a]	0,71±0.06[a]	0,87±0.07[a]
C 20:5 n3 (cis-5,8,11,14,17-eicosapentaenoic acid) EPA	3,86±0.02[a]	4,05±0.03[a]	4,11±0.03[a]	4,12±0.03[a]	4,21±0.03[a]
C 22:2 (cis 13,16 –docosadienoic acid)	1,05±0.06[b]	1,05±0.02[b]	1,02±0.02[b]	1,02±0.02[b]	1,06±0.02[b]
C 22:6 n3 (cis-4,7,10,13,16,19-docosahexaenoic acid) DHA	15,05±0.02[a]	16,09±0.02[a]	15,05±0.02[a]	15,35±0.02[a]	15,34±0.02[a]
Total PUFAs	**35,44**	**36,71**	**35,65**	**36,09**	**36,51**
PUFAs/SFAs	1,32±0.02[a]	1,35±0.02[a]	1,31±0.05[a]	1,33±0.05[a]	1,39±0.03[a]
Ó n6	12,46±0.07[a]	12,45±0.06[a]	12,62±0.03[a]	12,55±0.07[a]	12,81±0.08[a]
Ó n3	21,35±0.05[b]	22,71±0.04[b]	21,56±0.05[b]	21,97±0.05[b]	21,99±0.03[b]
n6/n3	0,58±0.02[a]	0,55±0.02[a]	0,59±0.05[a]	0,57±0.05[a]	0,58±0.03[a]
DHA/EPA	3,90±0.08[a]	3,97±0.08[a]	3,66±0.07[a]	3,73±0.05[a]	3,64±0.06[a]
Unidentified	6,96±0.02[a]	6,08±0.02[a]	5,88±0.03[a]	5,59±0.02[a]	6,76±0.04[a]
The letters in the same line show the differences in the results of statistical analysis.					

Lipids in the fatty muscle tissue of trout fed with feed containing 2 and 3% zeolite contained the most saturated fatty acids. The most monoene acids were contained in lipids of the muscle tissue of fish fed with feed with the addition of 2 and 3% zeolite. The level of SFAs was comparable, and MUFAS was significantly lower than the level oberved by Łuczyńska (Łuczyńska et al. 2011). The most n-6 polyene fatty acids were noted in the muscle tissue of fish fed with feed with the addition of 1% zeolite, however this content was greater in all experimental groups in comparison to the control group.

Among them, those present in the highest content in the experimental group of fish were C18:1n9, oleic acid (OLA, 20,08–21,72 %), C16:0, palmitic acid (PAA, 15.34–16.22%), DHA (15.05–16.09%), C18:2 linoleic acid (LIA 11.06–11.45%), palmitoleic acid (PLA 5.30–5.69%), C16:1 EPA (3.86–4.21%), stearic acid (STA 3.15–3.44%), and C14:0, and myristic acid (MYA, 3.27–3.85%).

Epidemiological studies have shown that n-3 fatty acid intake is inversely related to cancer, cardiovascular diseases, psychiatric disorders, asthma, bone mineral density and type 2 diabetes. Because of this fact, polyunsaturated fatty acids (PUFAs) should be separated into n-3 and n-6 fatty acids. Although n-3 and n-6 PUFA levels in the two experimental groups (1% and 3% additives) were higher than in the control group, the difference was statistically significant.

The muscle tissue of trout fed with feed with the addition of 4% zeolite was characterized as the richest source of EPA. Linoleic acid was dominant in the group of n-6 fatty acids, and DHA and EPA were dominant in the n-3 group. Other researchers have made similar observations.

The proportions of FAs-n3 (21.35%; 21.56–22.71% control and experimental groups) were generally higher than those of FAs-n6 (12.46%; 12.45–12.81%). The UK Department of Health recommends an ideal n6/n3 ratio of 4.0 at maximum. Values higher than the maximum value are harmful to health and may promote cardiovascular diseases. In this study, the n6/n3 ratio was found to be 0.55–0.59 in the all experimental groups.

The recommended minimum value of the PUFAs/SFAs ratio is 0.45, which is lower than the values of 1.32 and 1.31–1.39 from the control group and the experimental groups treated with RGM-2M and additives. DHA/EPA ratios ranged from 0.72 to 6.89 in some fresh water fish species and it was equal to 1.56 in rainbow trout.

In this study, the ratio of DHA/EPA in rainbow trout fed with RGM-2M with 1% zeolite was found to be 3.97 and was greater than in control groups (3.90). In other groups, this ratio was lower and amounted to from 3.64–3.73.

No simple correlation can be found between the content of individual fatty acids and the percentage of addition of zeolite to feed on the basis of the conducted analysis. Undoubtedly, the addition of zeolite has an influence on the profile of fatty acids in lipids in the muscle tissue of Rainbow trout, and it also increases the content of n-3 and n-6 polyene fatty acids advantageously.

Conclusion

The results of this study confirmed that zeolites have a positive effect on the biochemical features of meat. A negative effect of clinoptilolite was not determined. This study showed the significance of using zeolites as a feed additive for fish, as part of a comprehensive program to control fish meat quality and to increase the level of polyunsaturated fatty acids.

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А.Е. Паритова, Н.Б. Сарсембаева

ВЛИЯНИЕ ЦЕОФИШ НА СОСТАВ ЖИРНЫХ КИСЛОТ МЯСА РЫБЫ

В данной статье изучали влияние кормовой добавки Цеофиш на состав жирных кислот мяса рыбы. В результате исследовании было установлено, что мясо рыб получавших Цеофиш с кормом по составу жирных кислот особо не отличались от здоровой рыбы (контрольной). Это говорит о том, что Цеофиш не оказывает негативного влияния на обмен веществ.

А.Е. Паритова, Н.Б. Сарсембаева

ЦЕОФИШТІҢ БАЛЫҚ ЕТІНІҢ МАЙ ҚЫШҚЫЛДАРЫ ҚҰРАМЫНА ӘСЕРІ

Бұл мақалада Цефоиш азық қоспасының балық етінің май қышқылдары құрамына әсерін зерттедік. Зерттеулер нәтижесінде азықпен бірге Цеофишті қабылдаған балық еті май қышқылдарының құрамы бойынша сау балықтардың (бақылау) етінен ерекшеленбеді. Бұл Цеофиштің зат алмасуға негативтік әсері жоқ екенін көрсетеді.