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Effect of extractive substances from spruce needle biomass on egg production and quality

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Extractive substances of spruce needles were produced from a forestry by-product – green biomass of spruce needles.

Investigations were carried out to evaluate the effects of the additive of biologically active substances from spruce needles (total extractive, separate, neutral extractive and acidic extractive substances) on laying hens' productivity and egg quality, to assess their effects on the innovative composition of hen eggs. The feeding trial was conducted with a cross Lohmann Brown laying hens by adding the total, neutral and acid extractive substances of spruce needles to the composition of trial group hens' diet at the amount of 0.04–0.05 %. The control group did not receive additives of extractive substances.

Using spruce needle extractive substances increased egg production on average by 2.10–4.86 %, egg weight by 3.86–7.50 %, and decreased feed conversion by 10.76–12.55 % in comparison with the control group ($p < 0.05$). The use of the additive of neutral extractive substances in hens' diet improved the content of α-linolenic acid by 0.35 %, total carotenoids by 2.31 mg kg^{-1}, and α-tocopherol by 3.20 mg 100 g^{-1}, but decreased the cholesterol level by 178.06 mg 100 g^{-1} in egg yolk in comparison with the control group (commercial eggs).

By using additives of neutral extractive substances in hens' diet it was possible to obtain hen eggs of an innovative composition.

Introduction

Forestry by products – green biomass of needles, non-lignified shoots and twigs of the Latvian major conifer species Norway spruce *Picea abies (L)* H. Karst contain significant amounts of natural biologically active substances.

The major active ingredients of biologically active substances from spruce needle green biomass are chlorophyll and other compounds: antioxidants, vitamins, minerals, phytosterols, polyprenols, squalene, sodium salts of resin acids (balsamic compounds), essential oils, etc. [6, 8]. The previously mentioned biologically active substances exert a broad therapeutic and prophylactic influence on the poultry and on the human organism [1].

A complex of natural biologically active substances can be extracted with a nonpolar organic solvent from spruce needle biomass.

Upon including the biologically active substances contained by spruce needle extracts into the poultry diet, they are transferred from the feed to poultry eggs. Researchers [3, 7, 10] have confirmed that the transfer of biologically active substances from feed to production takes place, resulting in products of innovative composition, enriched with biologically active substances.

In the 1970s, only a biologically active complex composed of total extractive substances called chlorophyll–carotene paste was extracted from pine needles and pine needles biomass. A small number of investigations were carried out on the chlorophyll–carotene paste utilization in poultry feeding to improve productivity and hen egg quality [11].

No investigations of the improved egg quality and functional eggs produced by using pine needle total extractive substances (chlorophyll–carotene paste) have been carried out.

According to our data, spruce needle biomass contains a higher amount of different biologically active substances (antioxidants, fatty acids, etc.). In this aspect, from total extractive substances of spruce needle biomass, neutral and acid extractive substances can be extracted.

The aim of our investigations was to evaluate the influence of spruce needle total extractive and separately neutral extractive and acid extractive substances on laying hen productivity and egg quality by specifying the increase of the content of n-3 fatty acid and antioxidant carotenoids in egg yolk and to confirm its possible use to obtain hen eggs of an innovative composition.

Materials and methods

The feeding experiment was conducted with cross Lohmann Brown laying hens aged 24 to 36 weeks.

Two hundred laying hens were weighed and distributed into four groups (Table 1). Laying hens were reared in floor pens (n = 50).

Table 1. Experimental design

Group	Basic diet*	Feeding programme
		Additives of biologically active substances
1st group–control	Basic diet	-
2nd group–trial	Basic diet	Total extractive substances from spruce needle biomass
3rd group–trial	Basic diet	Neutral extractive substances from spruce needle biomass
4th group–trial	Basic diet	Acid extractive substances from spruce needle biomass

*Layer Management Guide. Lohmann Brown – classic requirements.

The basic diet contained wheat, barley, rapeseed oil, rapeseed oil cake, monocalciumphosphate, vitamine–mineral premix for layers, salt, DL-methionine, L-lysine HCl, Carophyll Yellow, Carophyll Red. The basic diet ingredient composition was the same for all groups. The content of the basic diet for all groups was similar – nutritive value of crude protein (16.26 %), fat (6.82 %), crude fibre (5.53 %), ash (13.96 %), methionine (0.38 %), lysine (0.76 %), Ca (3.70 %), P (0.76 %), Na (0.17 %), vitamins and microelements according to the requirements of the cross Lohmann Brown hens. There were no additives of extractive substances from spruce needle biomass in the control group diet.

The spruce needle total extractive, neutral and acid extractive substances were added in doses of 0.04–0.05 % to the diet of 2, 3, 4 groups. The spruce needle neutral and acid extractive substance additives were obtained by extraction from the total extractive mass. The additives were used in the form of paste and were dark-green in colour.

The main indices of poultry productivity and egg quality were recorded and evaluated during the investigation. The gas chromatography method was used for the analysis of fatty acids, α-tocopherol and carotenoids, and the Blur colorimetric method was used for cholesterol analysis. The morphological quality of eggs was evaluated employing the Futura–Eggs Quality Measure software.

The statistical analysis was performed using SPSS 17. Statistical significance was declared at $p < 0.05$.

Results and discussion

The chemical composition of spruce needle biomass extractive substances is shown in Table 2.

Table 2. Chemical composition of substances extracted from spruce needle biomass (natural sample)

Parameter	Total extractive substances	Neutral extractive substances	Acids extractive substances
Total lipids, %	0.79	0.72	0.74
Fatty acids, % of total lipids			
Palmitic acid	5.60	4.02	4.06
Stearic acid	6.57	2.19	3.59
Oleic acid	5.61	7.66	4.14
Linoleic acid	2.56	4.04	1.54
α-Linolenic acid	1.01	1.02	1.04
Carotenoids, mg kg^{-1}	2070.40	2607.64	161.66
α-Tocopherol, mg 100 g^{-1}	77.00	110.15	28.00

A comparison of fatty acid content in spruce needle extractive substances indicates that neutral extractive substances and acid extractive additives contain a little less palmitic, stearic fatty acids in comparison with the respective components of total extractive substances.

The concentration of oleic acid, linoleic acid, carotenoids and α-tocopherol was higher in the additive of neutral extractive substances in comparison with total extractive substances.

It was anticipated that the additives of biologically active substances after their transfer from feed to poultry organism will take part in metabolism processes and favourably influence hens' productivity and egg quality with the possibility to obtain innovative quality eggs.

The effects of extractive substances from spruce needle biomass on egg production, eggs mass, feed intake and feed conversion are shown in Table 3.

Table 3. Effects of substances extracted from spruce needle biomass on the production of laying hens

Production parameters	Control group	Additives of		
		total extractive substances	neutral extractive substances	acid extractive substances
Egg production, %	94.15	96.25	98.39	99.01
± to control	100.00	102.10	104.24	104.86
Egg mass, g/hen/per day	54.31	59.62	60.05	59.32
% to control	100.00	109.77	110.56	109.22
Feed intake, g/hen/per day	152.00	145.80	150.13	145.97
Feed conversion* kg kg^{-1}	2.79	2.44	2.49	2.46
% to control	100.00	87.45	89.24	88.17

*Feed conversion = kg of feed : kg of egg mass during the stydy period.

Feeding diets with added extractive substances from spruce needle biomass had positive effects on egg production parameters: egg production increased by 2.10–4.86 %, egg mass by 9.22–10.56 %, and feed conversion decreased by 10.76–12.55 % as compared with the control group ($p < 0.05$).

The obtained data indicate that natural biologically active substances of spruce needle extractives favourably influenced hens' organism metabolic processes and increased their productivity.

Egg quality morphological parameters such as yolk weight, albumen height, Haugh Unit (HU) value and shell thickness were not affected by feeding diets with extracts from spruce needle biomass (Table 4).

By using additives of substances from spruce needle biomass in hen diets, egg weight increased by 2.23–4.33 g or by 3.86–7.50 % in comparison with the control group ($p < 0.05$).

Table 4. Effect of substances extracted from spruce needle biomass on egg morphological quality

Egg component	Control group	Additives of		
		total extractive substances	neutral extractive substances	acids extractive substances
Weight, g	57.69	62.02	61.10	59.92
% to control	100.00	107.50	105.91	103.86
Yolk weight, %	26.19	26.11	27.37	26.80
Albumen height, mm	6.78	6.69	7.31	7.37
Haugh Unit (HU) value	82.01	80.80	84.38	84.41
Shell thickness, mm	0.36	0.36	0.36	0.35

A higher average egg weight (62.02 g) was obtained by feeding a diet with an additive of total extractive substances.

Nevertheless, from the marketing point of view and according to egg weight distribution [9], most profitable were eggs obtained from hens fed with a neutral extractive substance additive (Table 5).

Table 5. Expected egg grades (%) for different egg weight

Egg weight, g	Categories of eggs	Control group amount of eggs, %	Additives of		
			total extractive substances, %	neutral extractive substances, %	acid extractive substances, %
73 >	XL	0.34	14.44	-	1.94
63–72.9	L	23.99	20.56	46.87	50.00
53–62.9	M	74.66	65.00	53.13	47.57
> 53	S	1.01	-	-	0.49
Total L + M		98.65	85.56	100	97.57

One hundred percent of eggs obtained from hens to which a neutral extractive additive was fed were realized in L and M weight categories, similarly as using acid extractive substance additives.

Using total extractive substance additive, less of L weight category eggs (by 26.31–29.44 %) were obtained and more eggs were obtained in the XL weight category in comparison with neutral and acid extractive substances. It is possible that total extractive biologically active substances influenced the ovulation process and the distribution of eggs according to weight categories differed correspondingly.

The spruce needle extractive additives in hen feed did not influence the total content of saturated, monounsaturated and polyunsaturated fatty acids in eggs yolk (Table 6).

Applying additives of neutral extractive substances in hen diet, the levels of polyunsaturated fatty acids – arachidonic, α-linolenic acid and docosahexaenoic – changed in egg yolk.

The additives influenced a decrease of arachidonic acid in eggs yolk by 0.35 %, but at the same time they increased the content of α-linolenic and docosahexaenoic acids by 0.35 % and 0.29 % from total lipids in comparison with the control group. Similarly as noted in references [2, 3], natural biologically active substances from a neutral extractive influenced fatty acid reduction and synthesise processes in the poultry organism and its content in egg yolk.

One of the innovative quality tests of eggs is the Σ (n – 6) and Σ (n – 3) fatty acid content and ratio in egg yolk. Σ (n – 6) fatty acid amount in egg yolk was within 17.72–18.54 % and Σ (n – 3) content of fatty acids was varied from 4.08 to 4.68 %. The Σ (n – 6) content of fatty acids was a little higher in control group eggs and when using total extractive substance additives in hens' diet, but a little less when using neutral and acid extractive substances.

Table 6. Effect of spruce needle biomass extractive substances on egg fatty acid level

Fatty acids, % of total lipids	Control	Additives of		
		total extractive substances	neutral extractive substances	acid extractive substances
Saturated (SFA)	32.81	31.72	32.52	33.27
Monounsaturated (MUFA)	40.86	39.65	39.95	40.06
Polyunsaturated (PUFA)	22.54	22.67	22.44	22.14
n – 6 fatty acids:				
Linoleic acid	15.44	15.53	15.00	14.74
γ-Linolenic acid	0.06	0.16	0.11	0.07
Eicosatrienoic acid	0.23	0.21	0.27	0.25
Arachidonic acid	2.73	2.64	2.38	2.66
Σ (n – 6)	18.46	18.54	17.76	17.72
n – 3 fatty acids:				
α-Linolenic acid	1.38	1.37	1.73	1.66
Eicosapentaenoic (EPA)	0.10	0.11	0.09	0.08
Docosapentaenoic (DPA)	0.37	0.29	0.34	0.31
Docosahexaenoic (DHA)	2.23	2.36	2.52	2.37
Σ (n – 3)	4.08	4.13	4.68	4.42
Σ (n – 6) : Σ (n – 3)	4.52:1	4.48:1	3.85:1	4.08:1

Table 7. Content of cholesterol and carotenoids in eggs

Egg quality	Control	Additives of		
		total extractive substance	neutral extractive substance	acid extractive substance
Yolk weight, g	15.10	16.19	16.72	15.46
Cholesterol, mg100 g	685.83	615.00	507.77	710.41
Cholesterol, mg/egg	103.56	99.56	84.89	109.82
Carotenoids, mg/kg	5.22	5.84	7.53	4.10
Carotenoids, mg/egg	0.078	0.094	0.126	0.063
α-Tocopherol, mg/100 g	3.70	4.88	5.70	5.99
α-Tocopherol, mg/egg	0.55	0.79	0.95	0.92

At the same time, the Σ (n – 3) fatty acid content in egg yolk was higher by 0.60 % when using in hens' diet additives of neutral extractive and by 0.34 % when using acid extractives in comparison with the control group. Such fatty acid changes could be connected with the influence of natural biologically active substances on fatty acid metabolism in hen organism and its transport from the feed to egg yolk.

The ratio between Σ (n – 6) and Σ (n – 3) fatty acids was (3.85–4.52) : 1 in our trial. The optimal ratio of Σ (n – 6) to Σ (n – 3) fatty acids in the human diet is 10 : 1 to 5 : 1 [12].

The majority of human diet researchers recommend that the ratio of Σ (n – 6) and Σ (n – 3) fatty acids should be less than 5 : 1.

By using additives of extractive substances from spruce needle biomass in hens' feeding, the Σ (n – 6) and Σ (n – 3) ratio was within the recommended limits. The ratio of Σ (n – 6) and Σ (n – 3) lower the product diet value is more favourable for human organism [12].

When using in hens' feeding neutral extractive substance additives, the most favourable for human organism ratio of the Σ (n – 6) and Σ (n – 3) fatty acids (3.85 : 1) was obtained in comparison with the control group; also, the tendency of a higher Σ (n – 3) level in egg yolk was observed in comparison with the control group.

Possibly it is connected with a comparatively higher content of antioxidants in the spruce neutral extractive substance additive. Total and acid extractive substance additives contained a lower amount of carotenoids (Table 7). A similar ratio of antioxidants and fatty acid was noted by Z. Hayat et al. [3]: the higher antioxidant levels in feed, the higher the Σ (n – 3) level in eggs.

So, egg yolk carotenoid content to a large extent corresponds to its content in spruce needles. Since the neutral extractive substances contained a comparatively higher level of carotenoids, its level was higher also in egg yolk (7.53 mg kg^{-1}) in comparison with the control group ($p < 0.05$). Less carotenoids contained acid extractive substances, and therefore less carotenoids (4.10 mg kg^{-1}) were noted in eggs of this group. The spruce extractive substance additive used in hens' diet increased in egg yolk the content of antioxidants α-tocopherol on average by 1.20–3.20 mg 100 g^{-1} in comparison with the control group ($p < 0.05$).

Cholesterol synthesis and reduction in the poultry organism and its further transformation and content in egg yolk were determined by the level of carotenoids, α-tocopherol, fatty acids and other biologically active substances in the additive of spruce extractive substances.

Egg yolk contained on average 710.41–507.77 mg of cholesterol in 100 g. The use of neutral extractive substance additives decreased cholesterol level by 178.08 mg 100 g^{-1} but increased carotenoids by 2.31 mg kg^{-1} and α-tocopherol content by 3.20 mg 100 g^{-1} in comparison with the control group (Table 7).

Conclusions

Spruce needle total extractive and separate biologically active neutral and acid extractive substances used in the diet of laying hens improved their productivity (on average by 2.10–4.86 %), egg weight (3.86–7.50 %) and to decrease feed conversion on average by 10.76–12.55 % in comparison with control group ($p < 0.05$).

Applying spruce needle neutral extractive substances in hens' diet allowed to produce eggs of an innovative composition – with an increased level of carotenoids (on average by 2.31 mg kg^{-1}) and α-tocopherol (by 3.20 mg 100 g^{-1}), but a decreased cholesterol level (178.06 mg 100 g^{-1}) in comparison with the control group.

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EGLĖS SPYGLIŲ BIOAKTYVIŲ JUNGINIŲ ĮTAKA VIŠTŲ DEDEKLIŲ PRODUKTYVUMUI IR KIAUŠINIŲ KOKYBEI

S a n t r a u k a

Tirta eglės spyglių bioaktyviųjų junginių įtaka vištų dedeklių produktyvumui. Nustatyta, kad panaudojus eglės spyglių preparatus kiaušinių kiekis padidėja 2,10–4,86 %, jų svoris padidėja 3,86–7,50 %. Pašarų konversija sumažėja 10,76–12,55 %, palyginti su kontroline vištų grupe. Naudojant neutralaus ekstrakto medžiagas, padidėja linoleno rūgšties, karotinoidų ir α-tokoferolio koncentracija, o cholesterolio koncentracija kiaušinio trynyje sumažėja.