THE IMPACT OF IODINE ON BIOCHEMICAL BLOOD PARAMETERS IN LAYING HENS

Rasa Bobinienė, Diana Gudavičiūtė, Manefa Miškinienė
Research Laboratory of Biological Diversity and Technologies, Vilnius Pedagogical University, Studentų str. 39, Vilnius LT-08106, Lithuania, tel. +37052757095
Corresponding author: bamlab@vpu.lt

Summary. In the areas, where the biosphere is deficient in iodine, the feed for domestic animals and laying hens should be supplemented with larger than the recommended doses of the trace element iodine. That would enable to concentrate the reserves of this element in the production, thus enriching human nutrition with iodine. The goal of the trial was to investigate the changes in the amount of the thyroid hormones, proteins and fats in the blood and blood serum of laying hens by using a stable concentrated preparation "Jodis" instead of the usual potassium iodide in the feed. For the trial, three equal groups of laying hens were randomly formed, each containing 40 hens from 30 to 47 weeks of age. The laying hens of Group 1 (control group) were fed with the standard diet supplemented with a recommended daily dose iodine i. e. 1 mg I/kg feed in the form of potassium iodide. The laying hens of Groups 2 and 3 (experimental groups) were fed with the standard diet where potassium iodide was replaced by a dry stable iodine supplement "Jodis". The amount of iodine in the diet given to laying hens in Group 2 was 1 mg I/ 1 kg feed, and in Group 3 − 4 mg I/ 1 kg feed, respectively.

There were significantly increased levels of thyroglobulin and free thyroxine, and decreased levels of free triiodothyronine in blood, and triglicerides in blood serum of experimental laying hens (Groups 2 and 3) compared to controls (Group 1). The HDL and LDL cholesterol level in blood serum of experimental hens was lower than in controls, but the difference was not statistically significant.

Keywords: iodine, thyroid hormones, cholesterol, triglycerides, proteins, laying hens.

JODO KIEKIO LESALUOSE ĮTAKA VIŠTŲ KRAUJO BIOCHEMINIŲ RODIKLIŲ POKYČIAMS

Rasa Bobinienė, Diana Gudavičiūtė, Manefa Miškinienė
Vilniaus pedagoginis universitetas, Studentų g. 39, Vilnius LT-08106
tel. +370 527 5709; el. paštas: bamlab@vpu.lt

Santrauka. Šalyje, kur jodo suvartojama nepakankamai, naminiai gyvuliai ir paukščiai jo turėtų gauti daugiau nei rekomenduojama ir taip koncentruoti šio elemento atsargas produkcijoje.

Bandymo tikslas – ištirti skydliaukės hormonų, baltymų ir riebalų kiekio kraujyje kaitą, vietoj įprasto kalio jodido vištų lesaluose naudojant sausą stabilaus jodo papildą „Jodis".

Bandymui su 30–47 savaičių vištomis dedeklėmis buvo sudarytos 3 grupės po 40 vištų. Pirma grupė – kontrolinė, o kitos – bandomosios. Pirmos grupės vištos dedeklės buvo lesintos standartiniais kombinuotaisiais lesalais, o antros ir trečios grupių vištos dedeklės gavo tokios pat sudėties lesalus, kaip ir kontrolinės grupės, tik įprastas kalio jodidas buvo pakeistas sausu, geriau pasisavinamu stabilaus jodo papildu „Jodis". Antros grupės vištų dedeklių lesaluose stabilaus jodo dozė buvo 1 mg I/1 kg lesalų, o trečios grupės lesalai papildyti 4 mg I/1 kg lesalų.

Tyrimo metu bandomųjų grupių vištų kraujyje nustatėme padidėjusį tiroglobulino (Tg) ir laisvo tiroksino (FT4) kiekį. Laisvo trijodtironino (FT3) abiejose bandomųjų grupių vištų kraujyje nustatyta mažiau, nei kontrolinės grupės. Tirtas ir bendras cholesterolio kiekis vištų dedeklių kraujo serume. Nustatyta, kad bendras jo kiekis bandomųjų grupių vištų kraujo serume buvo mažesnis, nei kontrolinės grupės. DTL (didelio tankio lipoproteinų) ir MTL (mažo tankio lipoproteinų) cholesterolio skirtumas tarp kontrolės ir bandomųjų grupių nebuvo statistiškai patikimas. Kraujo trigliceridų bandomųjų vištų kraujo serume buvo mažiau, nei kontrolinės grupės paukščių.

Raktažodžiai: jodas, skydliaukės hormonai, cholesterolis, trigliceridai, proteinai, vištos dedeklės.

Introduction. Over thousands of years iodine was washed from the soil in many mountainous as well as continental regions. It has been estimated that about 800 million people worldwide are deficient in iodine (Benoist *et al*., 2008). There are about 50 mg of iodine in the human body. 75% of iodine is in thyroid hormones. Iodine concentration in the thyroid gland is 30 times higher than in the blood. A person has to get about 150 µg of iodine with food daily. The human body accumulates a reserve of iodine; therefore, when iodine is not available from the environment, its reserves may be sufficient for approximately 2 months (Gardner and Shoback, 2006).

Thyrotropin (TSH), a hormone of the front part of the pituitary, regulates the production of thyroid hormones. It is one of the most important indicators of the thyroid gland function. It stimulates the secretion of free thyroxine (FT4) and free triiodothyronine (FT3) in the body and also stimulates the thyroid gland growth. The thyroid hormones thyroxine and triiodothyronine regulate the growth processes and activate metabolism, stimulate the activity of the body's functional systems. Under the impact of thyroid hormones, protein synthesis in the body

intensifies. The carbohydrate intake in the alimentary canal depends on these hormones, and thyroid hormones have influence on fats metabolism. In case the amount of these hormones in the body reduces, metabolism gets slower, the quantity of fats increases, and a reserve of fats accumulates (Leonard and Visser, 1986; Nobikuni *et al*., 1989; Kaneko et al., 1997; Nixon *et al*., 1988).

In order to eliminate the problem of iodine deficiency in food and to maintain the health of the population, it is necessary to look for ways how to supplement the diet of laying hens with the stable iodine preparations and to ensure good growth of laying hens as well as to promote the opportunities for consuming the iodine-enriched eggs and poultry meat (Kepalienė *et al*., 2006). One of the main conditions, which the growth of meat resources depends on, is certainly highly nutritious feed for animals and laying hens. Absence or shortage of vital biologically active substances in their feed negatively affects the state of the laying hens' health, their productivity, and feed conversion. In the areas where insufficient consumption of iodine is widespread, animals and laying hens should get more iodine than required, thus concentrating the residues of this element in milk, eggs, and meat (Lichovnikova *et al*., 2003; Flachowsky, 2007).

In case of iodine deficiency, hens lay fewer eggs, the foetal weight reduces, fewer chickens are hatched and they are weak, with an increased thyroid gland (Stanley and Bailey, 1989).

The goal of our study was to investigate the changes of biochemical parameters in the blood and blood serum of laying hens by using a stable concentrated preparation "Jodis" instead of the usual potassium iodide in the feed.

Materials and methods. The investigations were carried out with Hisex brown line combination hens. During the trial with the laying hens at the age of 30 weeks, 3 groups were formed, each of them containing 40 laying hens. The laying hens of Group 1 (control group) were fed with the standard compound feed (C), containing potassium iodide (KI) as the source of iodine, and the laying hens of Groups 2 and 3 (experimental groups) were fed with the standard diet where potassium iodide was replaced by a dry stable iodine supplement "Jodis" with increased assimilation of iodine. The amount of iodine in the diet given to laying hens in Group 2 was 1 mg I/ 1 kg feed, and in Group 3 − 4 mg I/ 1 kg feed, respectively. The laying hens were kept in cages, they were fed and had free access to water via automatic equipment.

The investigations of thyroid hormones were carried out by the biochemistry analyzer "ELECSYS 2010" (Roche Diagnostics). The hens' total blood protein, the amount of triglycerides and cholesterol were determined by the analyzer Cobas INTEGRA 400 Plus. Blood tests were performed with the laying hens at the age of 47 weeks.

Husbandry conditions for laying hens were complying with good commercial practices and with the Law of the Republic of Lithuania on the Care, Keeping and Use of Animals as well as secondary legislation – Order of the State Food and Veterinary Service of the Republic of Lithuania "On Veterinary Regulations on Breeding, Handling and Transportation of Laboratory Animals" and "On the Use of Laboratory Animals in Scientific Experiments" (Law of the Care, Welfare and Use of Animals, 2002).

The data was processed by applying statistical biometry methods and using Statistica for Windows, Version 6.0 (StatSoft Inc.).

Results and Discussion. TSH concentration in the blood is mostly inversely proportional to the concentration of FT4 and FT3. The data of our investigations demonstrated that the amount of TSH in blood of the laying hens of Group 2 was 0.01 mU/mL, or on 14.29% lower (Table 1), compared to the controls ($P<0.01$), and the amount of TSH in blood of the laying hens of Group 3 decreased on 0.02 mU/mL (28.58%), compared to controls ($P<0.001$).

Table 1. **The amount of thyroid hormones in blood serum of laying hens**

Group No.	Feeding characteristics	TSH mU/mL	Tg ng/mL	FT3 pg/mL	FT4 ng/dL
1	C + KI (1 mg I/ 1 kg feed)	0.07±0.006	0.35±0.034	0.74±0.017	4.35±0.378
2	C + "Jodis" (1 mg I/ 1 kg feed)	0.06±0.027*	0.37±0.012	0.66±0.065	5.49±0.376**
3	C + "Jodis" (4 mg I/ 1 kg feed)	0.05±0.011**	0.39±0.019*	0.62±0.012*	4.74±0.165

Note: The difference between the control and the test groups is statistically significant with (*$P<0.01$), (**$P<0.001$).

The thyroid gland synthesizes and secretes a mixture of the hormones thyroxine T4 and triiodothyronine T3, which are important for the normal development of the body and metabolism. Most of them are bound to proteins in the blood, and only an insignificant part of them (0.02-0.04%) is free. Thyroidins, bound to proteins, form a circulating reserve of hormones. Only a free hormone is physiologically active, and its amount in blood plasma is very small (Fauci *et al*., 2008). T4 makes a functional reserve of T3 in the blood. In tissues, T4 is converted into T3, which is 3-4 times more active. As mono- and diiodthyronins bind in the thyroglobulin (Tg), which is located in the thyroid follicles, triiodothyronine and thyroxine are synthesized. Thyroglobulin is an inactive form of thyroid hormones (triiodothyronine T3 and thyroxine T4), a depot of these hormones (Beiša, 2006). During the investigation

we determined the amount of thyroglobulin in the blood of laying hens: in the blood of the hens of Group 2 it was 0.02 ng/mL or on 5.71% higher, compared to the control group. In the blood of the hens of Group 3, thyroglobulin increased on 0.04 ng/mL (11.42%), compared to the control group (P<0.001). Moreover, a higher amount of free thyroxine (FT4) was established in the blood of hens in experimental groups. The amount of this hormone in the blood of the hens of Group 2 increased on 1.14 ng/dL, or by 26.20% (P<0.001), and in the blood of the hens of Group 3 it increased on 0.39 ng/dL, or by 8.96%, compared to the control group.

It was established that the amount of free triiodothyronine (FT3) in both experimental groups was lower compared to controls. In the blood of the hens of Group 2, the amount of FT3 was lower on 0.08 pg/mL, or by 10.82%, compared to the control group, and this index of Group 3 was by 0.12 pg/mL, or by 16.22% (P<0.01) lower compared to the control group. Following the data from literature, a major part of this hormone develops not in the thyroid gland but in the periphery (liver, kidney, cells of connective tissue) (Gardner and Shoback, 2006).

The results from this study indicate, that thyroid hyperfunction in experimental groups was observed.

Total cholesterol amount and the amount of triglycerides in the blood serum of laying hens was also investigated (Table 2). It was determined that total cholesterol amount in the blood serum of hens in experimental groups was lower compared to the control group. In the blood serum of the hens of Group 2, the amount of total cholesterol was lower on 1.12 mmol/L (31.12%), and in Group 3 it was lower on 1.76 mmol/L (48.89%), compared to the control group (P<0.001). The HDL and LDL cholesterol difference between the control and the test groups was not statistically significant (P>0.05).

Table 2. **The amount of cholesterol and triglycerides in blood serum of the laying hens**

Group No.	Feeding characteristics	Cholesterol, mmol/L	Triglycerides, mmol/L
1	C + KI (1 mg I/ 1 kg feed)	3.60±0.925	20.79±7.235
2	C + "Jodis" (1 mg I/ 1 kg feed)	2.480.024**	11.63±1.419**
3	C + "Jodis" (4 mg I/ 1 kg feed)	1.84±0.127**	6.60±1.106**

Note: The difference between the control and the test groups is statistically significant with (*P<0.01), (**P<0.001).

The amount of triglycerides in the blood serum of hens in Groups 2 and 3 was on 9.16 mmol/L and on 14.19 mmol/L lower compared to controls (P<0.001).

The amount of total proteins in the blood serum of hens is shown in Table 3. In Group 2, the amount of total proteins was 12.43 g/L or on 25.42%, and in Group 3 17.43 g/L or on 35.64% higher compared to controls (P<0.001).

Table 3. **The amount of total proteins in the blood serum and the total amount of nucleic acids in blood**

Group No.	Feeding characteristics	The amount of total proteins, g/L
1	C + KI (1 mg I/ 1 kg feed)	48.90±2.129
2	C + "Jodis" (1 mg I/ 1 kg feed)	61.33±6.399**
3	C + "Jodis" (4 mg I/ 1 kg feed)	66.33±12.007**

Note: The difference between the control and the test groups is statistically significant with (*P<0.01), (**P<0.001).

It was demonstrated a decreased amount of the thyrotropin hormone (TSH) in the blood of experimental hens. This is in concert with data, that TSH concentration with animal age gradually increases (Klimienė *et al*., 2008; Špakauskas *et al*., 2007). Moreover, alongside with the increase of the amount of cholesterol in blood, TSH concentration increases as well (Kepalienė *et al*., 2006). According to data of other researchers, in case of the decreased amount of TSH in the blood, thyroid hyperfunction may be suspected (Weetman, 1997; Fauci *et al*., 2008).

An increased concentration of thyroid hormones in blood itself inhibits the production of TSH in hypophysis (a negative feedback) i. e. the secretion of thyroliberin in the hypothalamus discontinues (Gardner and Shoback, 2006). Thyroid hormones activate the metabolism (Lu *et al*., 2007), and our study confirmed this. FT4 influences the secretion of TSH, and in case TSH secretion is not normal, the excess or deficiency of thyroid hormones is observed. The amount of FT4 in blood is as usual inversely proportional to the amount of TSH i. e. as one of these parameters increases, the other one has to decrease, and vice versa. An increase in free thyroxine and a decrease in TSH is an indicator of the increased function of the thy-

roid gland**.** A decrease of FT4 and an increase of TSH indicate an insufficient function of the thyroid gland. According to the data from literature, thyroglobulin ensures permanent hormones access of T4 and T3 into the blood. Under the impact of protease, T4 and T3 separate from thyroglobulin (Klimiene *et al*., 2008). On the basis of the obtained results of the investigations of thyroid hormones, it is possible to state that thyroid hyperfunction in experimental groups was observed. This led to acceleration of the main metabolism; however, oxidation and phosphorylation processes were disbalanced, less energy was accumulated in macroenergetic compounds, and more heat was emitted. Furthermore, protein metabolism and gluconeogenesis in the liver become activated (Špakauskas *et al*., 2008; Leonard and Visser, 1986; Nikolic *et al*., 2001). Our data is in agreement with the results that thyroid hormones activate protein synthesis (Kepalienė *et al*., 2006). It was stated that the amount of thyroid hormones in plasma is more closely related to the feed and the amount of selenium and iodine in the feed (Špakauskas *et al*., 2008; Wichtel *et al*., 1996). Thyroxine activates the enzyme regulating the intensity of cholesterol synthesis (Beiša, 2006).

On the basis of the data of other researchers, thyroid hormones intensify lipolysis; therefore, the concentration of triglycerides and cholesterol decreases (Nobikuni *et al*., 1989), and this is confirmed by our study.

Conclusions.

The results from this study indicate that stable iodine supplement "Jodis" activated the thyroid function in experimental hens. The amount of the hormone (TSH) in the blood serum of experimental hens (Groups 2 and 3) groups decreased on 14.29 % ($P<0.01$) and on 28.58 % ($P<0.001$), FT3 decreased on 10.82% and on 16.22 % ($P<0.01$), and FT4 increased on 26.20 % ($P<0.001$) and on 8.96 % ($P<0.01$) compared to controls (Group 1).

Diet supplementation with stable iodine supplement "Jodis" statistically significantly intensified lipolysis and protein metabolism of experimental hens compared to supplementation of diet with potassium iodine in controls ($P<0.001$).

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