

INFLUENCE OF RAPESEED MEAL ON PRODUCTIVITY AND HEALTH OF BROILER CHICKS

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(Received January 4, 2001; accepted December 13, 2001)

Research was focussed on investigating the influence of different quantities (0, 10, 20 and 30%) of rapeseed meal (RM), 00-cultivar Silvia on production results, as well as on the morphological and pathohistological changes in the internal organs of chicks during the course of the experiment, measured on the 21st and 42nd days of the 42-day experiment. The experiment involved 120 chicks divided into four groups: a control group (C) and three experimental groups (E₁₋₃). It was found that chicks in Groups C, E₁ and E₂ realised significantly ($P < 0.05$) higher gains than those in Group E₃, both after the test period and at the end of the experiment. No significant differences with regard to feed conversion were found between groups of chicks. Throughout the experiment chicks in Groups E₁₋₃ were found to have a significantly ($P < 0.05$) heavier liver. Compared to Groups E₁₋₃, Group C chicks had a significantly ($P < 0.05$) heavier gizzard after 21 days, but following the finisher diet Group E₃ had a significantly ($P < 0.05$) lighter gizzard. Compared with birds in Group C, those in Groups E₂₋₃ had significantly heavier ($P < 0.05$) unevacuated intestines when fed the starter diet, and those in Groups E₁₋₂ showed a similar result when fed the finisher diet. Chicks in Group E₃ had a significantly lower grill weight than those in Groups C, E₁ and E₂, both in the first half of the experiment and at the end. The starter diet did not result in any differences in the quantity of abdominal fat among groups, but following the finisher diet Groups E₂₋₃ showed significantly less abdominal fat in comparison to Groups C and E₁ ($P < 0.05$). Group C showed a significantly higher carcass yield than Groups E₁₋₃ ($P < 0.05$) in the first half of the experiment and at the end. Groups E₁₋₃ manifested a slight to medium hypertrophy of thyroid epithelial tissue as well as slight thymus hypertrophy and slight atrophy of the cloacal bursa follicles.

Key words: Broiler chicks, rapeseed meal, production indicators, morphological and pathohistological changes of internal organs, hypothyroidism

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Rapeseed meal is a by-product of the rapeseed oil production process. Rapeseed, genus *Brassica*, is the oldest oil-yielding crop in Europe and whose production is widespread throughout the world. Rapeseed meal contains between 32 and 35% of crude protein (Grbeša et al., 1994). Modern technological processes ensures that following oil extraction the meal by-product possesses higher values of certain amino acids, notably methionine and cystine but especially lysine (Grala et al., 1994). Rapeseed also contains goitrogenic substances (glucosinolates). While the glucosinolate content in earlier cultivars of rapeseed exceeded 150 $\mu\text{mol/g}$, modern 00 cultivars contain less than 25 $\mu\text{mol/g}$. Former rapeseed cultivars also contained 50% of erucic acid, whereas today that level is 1.4% in total fatty acid content (Mustapić and Pospišil, 1995). In parallel with the introduction of new 00 cultivars to agricultural production, numerous experiments with feeding diets were carried out involving broiler chickens, where different quantities of rapeseed meal were fed as a source of protein and energy. The high content (13.5%) of crude fibre (Mawson et al., 1993), as well as of glucosinolate, tannin and phytate (Korol et al., 1994), proved to be a limiting factor in the application of greater quantities of rapeseed meal in chicken feed. Wetscherek et al. (1993) fed their chicks with a fodder diet consisting of modern-day 00 cultivars of rapeseed meal in quantities of 0, 8, 16 and 24% and with a feed diet containing 24% rape cake. They found that a diet based on levels of 16% or more of rapeseed meal, and 24% of rape cake, resulted in significant growth reduction, while feed conversion was lower only in chicks fed on 24% rapeseed meal. No significant changes were observed either in slaughterhouse results or in organoleptic properties when using increased levels of rapeseed meal in the daily diet. Quantities of 8% or more of rapeseed meal in the feed influenced the colour of meat, which was considerably lighter compared to birds whose feed contained no rapeseed meal.

Researches undertaken indicate that good results can be achieved in broiler rearing if birds are fed diets containing 10 to 15% of rapeseed meal (Fritz et al., 1993; Kolodizej, 1995; Jamroz, 1995). Haščik et al. (1994) replaced soybean meal in feed diet with 50 and 100% rapeseed meal, the result being lower final weights of broiler chicks. Koncicki et al. (1991) found that increased levels (20 and 30%) of rapeseed meal in broiler feed diets cause increased activity of serum enzymes (ALT, AP) and a rise in cholesterol levels, while haematological indicators remain unchanged. In comparison to the control group, chicks fed on rapeseed meal had a significantly lower final body weight and demonstrated lower feed conversion (6–13.2%). Research carried out by Khan et al. (1996) found that the addition of 10 and 15% of rapeseed meal in broiler chick feed diet leads to a significant enlargement of the thyroid gland. Yu et al. (1995) found that the addition of 12% and more of rapeseed meal to feed diet results in pathological changes in kidney, and 14% or more causes a haemorrhagic syndrome.

The aim of this paper was to establish the influence of different levels of Silvia 00-cultivar rapeseed meal (10, 20 and 30%), i.e., of glucosinolate levels

(2.18, 4.36 and 6.54 mmol/kg in the diet), on body weight and feed conversion, as well as on the occurrence of morphological and pathohistological changes to internal organs of experimental birds during the course of a 42-day experiment, linking this to the manifestation and level of hypothyroidism.

Materials and methods

Animals

The experiment involved 120 sexed, one-day-old Ross chicks (60 males and 60 females). The chicks were divided into 4 groups – one control group (C) and three experimental groups (E₁₋₃) – each comprising 30 birds, all of which were wing tagged.

Diet and feeding

During the first three weeks of the experiment birds were fed a starter diet, followed by a further three weeks on a finisher diet (total of 42 days). The control group (C) was not given rapeseed meal, while at the same time the quantities given to the experimental groups differed. Feed given to Group E₁ contained 10% (2.18 mmol/kg glucosinolate), Group E₂ 20% (4.36 mmol/kg glucosinolate) and Group E₃ 30% (6.54 mmol/kg glucosinolate) of rapeseed meal.

Concentrations of total and individual glucosinolates in rapeseed grain before extraction were determined by the ISO 9167-1-1992 method of high-performance liquid chromatography (ISO-9167-1-1992). Five major glucosinolates were identified. Four were alkenyl: 2-hydroxybut-3-enyl (progoitrim), but-3-enyl (gluconapin), pent-4-enyl (glucobrassicapin), 2-hydroxypent-4-enyl (gluconaoiferin); while one was indole: 4-hydroxyindol-3-ylmethyl (4-hydroxyglucobrassicin). The sum of all individual glucosinolates provided us with total concentrations. Composition of both starter and finisher diets for trial fattening was based on the chemical composition of raw materials. Tables 1 and 2 present the chemical composition of rapeseed meal and level of individual glucosinolates, while Table 3 presents the chemical composition of the starter and finisher diets. For the first seven days broiler chicks were fed from round, plastic floor troughs and then, until the end of the experiment, from round, galvanised, suspended troughs with a capacity of 10 kg. Fresh water was supplied twice daily from round, plastic water troughs, which were regularly washed and disinfected. Feed and water were provided *ad libitum*.

Table 1
Chemical composition of rapeseed meal – cultivar Silvia (%)

Moisture	Ash	Crude protein	Crude fat	Crude fibre	NFE*	Ca	P
11.9	6.6	33.4	2.3	11.5	34.3	0.68	0.73

*NFE = nitrogen-free extract

Table 2
Concentration of glucosinolates in Silvia rapeseed meal

Glucosinolate type	mmol/kg	Percentage (%)
2-Hydroxybut-3-enyl	13.17	60.39
But-3-enyl	5.81	26.64
Pent-4-enyl	1.34	6.14
2-Hydroxypent-4-enyl	0.62	2.84
4-Hydroxyindol-3-ylmethyl	0.21	0.96
Others	0.63	3.03
Total glucosinolates	21.78	100.00

Housing and management

Chicks were kept in accordance with the recommended Ross technology. Birds were weighed on days 1, 21 and 42, always at the same part of the day and in identical group sequence. Feed consumption was checked every time when the chickens were weighed, separately for each group.

Measuring and pathohistological examinations

The first trial slaughtering of chicks was carried out on day 21 of the experiment, three male chicks being selected at random from each group. Final weighing was carried out at the age of 42 days. After 12 hours of rest and fasting the birds were slaughtered and left in a hanging position to drain the blood. Once feathers were removed the chicks were processed according to the method of Gobic and Ljubić (1970); (hereinafter: 'grill processing'), which included cleaned carcass without head, neck, edible giblets and legs as carcass yield. Legs were severed at the tarsal joint. The carcass included lungs, kidneys, sex glands, cloacal bursa and the pectoral section of windpipe and gullet, as well as the two last cervical vertebrae. Abdominal fat comprised adipose tissue from the abdominal cavity and fat lining the gizzard and small intestine. Gizzards were separated from proventriculus, opened and cleaned and, with keratinised layer removed, were weighed. Intestines were weighed in an unevacuated state. Bile ducts and gallbladder were carefully separated from liver and were then individually weighed.

Table 3

Composition of starter and finisher diets and their chemical analyses (%) with calculated ME

Ingredient, %	Starter groups				Finisher groups			
	C	E ₁	E ₂	E ₃	C	E ₁	E ₂	E ₃
Rapeseed meal	0.0	10.0	20.0	30.0	0.0	10.0	20.0	30.0
Corn	57.2	51.5	48.5	43.3	61.8	57.3	56.5	52.5
Soybean meal	32.0	26.0	18.5	12.5	25.8	19.0	14.0	9.0
Sunflower meal	0.0	0.0	0.0	0.0	6.0	6.0	2.0	
Fish meal	6.0	6.0	6.0	6.0	1.0	1.0	1.0	1.0
Fat	1.8	3.8	4.5	6.0	2.2	3.8	3.8	5.0
Dicalcium phosphate	1.1	0.8	0.6	0.3	1.1	1.1	1.1	1.0
Limestone	1.1	1.2	1.2	1.2	1.2	1.0	0.8	0.7
Iodised salt	0.3	0.2	0.2	0.2	0.4	0.3	0.4	0.4
DL-Methionine	0.1	0.1	0.02	0.0	0.02	0.02	0.0	0.0
Vitamin-mineral premix	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Total	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0
Analysis as fed**								
Moisture	12.1	11.8	11.2	10.8	12.1	11.4	11.1	10.9
Crude protein	22.7	22.1	22.2	22.7	19.4	19.9	19.9	19.0
Crude fat	4.3	5.8	6.8	8.1	4.9	6.2	6.3	8.0
Crude fibre	3.7	4.3	4.3	7.6	4.3	5.8	5.7	4.8
Ash	5.4	5.2	5.5	5.4	5.1	5.2	5.3	5.1
Ca	1.1	1.2	1.2	1.1	0.9	1.0	1.0	1.0
P	0.7	0.7	0.7	0.7	0.7	0.7	0.7	0.7
NFE***	51.9	50.8	50.1	45.4	54.3	51.5	51.7	52.3
Metabolisable energy, MJ/kg&	12.4	12.5	12.4	12.4	12.3	12.3	12.3	12.3
Total glucosinolate, mmol/kg	0.0	2.2	4.4	6.5	0.0	2.2	4.4	6.5

Provided per kilogram of mix: vitamin A, 15,000 IU; vitamin D₃, 2,000 IU; vitamin E, 30 mg; vitamin K₃, 2 mg; vitamin B₁, 1 mg; vitamin B₂, 6 mg; niacin, 30 mg; D-pantothenic acid, 12 mg; vitamin B₆, 3 mg; vitamin B₁₂, 0.01 mg; biotin, 0.1 mg; folic acid, 0.5 mg; choline chloride, 500 mg; Fe, 500 mg; Cu, 8 mg; Mn, 8 mg; Zn, 50 mg; J, 0.5 mg; Co, 0.2 mg; Se, 0.15 mg; virginiamycin, 10 mg; coccidiostatic Amprol + (only in starter diet), 125 mg.

C = control group; E₁ = group given 10% rapeseed meal (RM), (2.18 mmol/kg glucosinolate); E₂ = 20% RM, (4.36 mmol/kg glucosinolate); E₃ = 30% RM, (6.54 mmol/kg glucosinolate); ** Official methods were used throughout (A.O.A.C., 1984); ***NFE = nitrogen-free extract; &Calculated data (Allen, 1993)

All weighing was performed using Mettler P1200 scales with a tolerance of ± 0.5 g. Subsequent to the starter and finisher diet the following organs were subjected to pathohistological tests: heart, liver, kidneys, spleen, cloacal bursa, thymus, pancreas, testicles, suprarenal gland and thyroid gland. Organs were kept in a 10% formalin solution at room temperature for a period of 48 h, fixation being the first stage of specimen preparation. Next, the organs were dehydrated with alcohol and inserted into paraffin. Tissue samples of 6 μm thickness were taken (serial sections), stained with haematoxylin and eosin (HE) and examined under an optical microscope.

Statistical analyses

All statistical analyses were performed using the GLM procedure (SAS, Institute Inc., 1996).

Results

Rapeseed meal used in the experiment was the Silvia cultivar. Meal contained no detectable concentration of erucic acid. Chemical analysis established that rapeseed meal contained a tot of 21.82 mmol/kg of glucosinolates, most of them alkenyl (Table 1). Among the 6 established groups of glucosinolate those most widely present were 2-hydroxybut-3-enyl (60.39%) and but-3-enyl (26.64%), while the least represented was 4-hydroxyindol-3-ylmethyl (0.96%).

Body weight and feed conversion

Body weight and feed conversion for birds aged 1, 21 and 42 days are presented in Table 4. Following starter diet (21 days), and finisher diet (up to day 42), findings were as follows: chicks in experimental group (E₃) were significantly ($P < 0.05$) lighter than those in the first (E₁) and second (E₂) experimental groups. Following a starter diet (21 days) chicks in Group E₂ showed the best feed conversion, while Groups E₁, E₃ and C demonstrated a somewhat lower feed conversion. After the finisher diet (42nd day of experiment) the best feed conversion was achieved by birds in the control group (C), while birds in all experimental groups (E₁₋₃) manifested a slightly lower feed conversion.

Internal organs, grill processing, abdominal fat and carcass yield

Table 5 presents weights of internal organs, grill processing, abdominal fat and carcass yield of broiler chicks measured after the starter diet.

It was found that chicks in the control group (C) had a significantly lighter ($P < 0.05$) liver weight in comparison with those in all three experimental groups. A statistically significant difference was also found in average gizzard weight between Group C and the three experimental groups. Average intestine weight of

birds in Groups E₂ and E₃ was significantly heavier ($P < 0.05$) than in birds in the control group (C) and the experimental group E₁. Significantly ($P < 0.05$) lower grill weight was measured in Group E₃ chicks in relation to those in Groups C, E₁ and E₂. No significant difference was found in the quantity of abdominal fat between all the groups of broiler chicks. Birds in the control group (C) did have a significantly higher ($P < 0.05$) carcass yield than birds in all three experimental groups.

Table 4

Average body weight (g) and average feed conversion (kg/kg) of growing chicks in the experiment (n = 30)

Days of trial	Groups of animals							
	C		E ₁		E ₂		E ₃	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Average body weight (g)								
1	49	3.1	47	4.6	48	3.4	47	3.6
21	620 ^{a,b}	81.8	663 ^{a,c}	69.2	656 ^{b,d}	74.2	619 ^{c,d}	63.1
42	1919 ^a	429.3	1892 ^b	358.1	1868 ^c	378.1	1716 ^{a,b,c}	220.5
Average feed conversion (kg/kg)								
21	1.68		1.65		1.63		1.67	
42	1.92		1.95		2.03		2.03	

a:a, b:b, c:c, d:d = $P < 0.05$; C = control group; E₁ = group given 10% RM, (2.18 mmol/kg glucosinolate); E₂ = group given 20% RM, (4.36 mmol/kg glucosinolate); E₃ = group given 30% RM, (6.54 mmol/kg glucosinolate)

Table 6 shows the weights of internal organs, grill processing, abdominal fat and carcass yield of chicks in experimental groups at the end of the experiment.

Following the finisher diet (42nd day) chicks in Group C had a significantly lighter ($P < 0.05$) liver than those in all three experimental groups. Chicks in Group E₃ had a significantly lighter ($P < 0.05$) stomach in comparison to Groups C, E₁ and E₂. The intestine weight of chicks in Groups E₁ and E₂ was significantly heavier ($P < 0.05$) than that of chicks in control group (C) and experimental group E₃. Grill weight in chicks of Group E₃ was significantly lower ($P < 0.05$) than in Groups C, E₁ and E₂. Chicks in Groups C and E₁ had a significantly higher ($P < 0.05$) quantity of abdominal fat in comparison with those in Groups E₂ and E₃. The best carcass yield was found in birds from in the control group (C), and was significantly ($P < 0.05$) lower in all the three experimental groups (E₁₋₃).

Table 5

Weights of some organs, grill processing, abdominal fat (g) and carcass yield (%) in chicks after a 21-day growing period (n = 30)

Parameter	Group of animals							
	C		E ₁		E ₂		E ₃	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Liver	15.00 ^{a,b,c}	1.69	18.50 ^a	2.00	17.25 ^b	1.04	19.63 ^c	2.33
Gizzard	17.63 ^{a,b,c}	5.18	15.88 ^a	1.55	15.25 ^b	2.77	13.50 ^c	2.67
Unevacuated intestines	40.88 ^{a,c}	5.19	41.63 ^{b,d}	3.78	53.13 ^{a,b}	5.03	47.00 ^{c,d}	4.41
Grill processing	426.38 ^a	15.77	419.88 ^b	4.61	420.25 ^c	11.37	417.8 ^{a,b,c}	33.99
Abdominal fat	4.50	3.51	5.13	2.42	6.75	1.98	6.75	1.83
Carcass yield, %	63.50 ^{a,b,c}	2.39	62.38 ^a	0.74	62.00 ^b	1.31	61.88 ^c	1.55

a:a, b:b, c:c, d:d = P < 0.05; C = control group; E₁ = group given 10% rapeseed meal (RM), (2.18 mmol/kg glucosinolate); E₂ = group given 20% RM (4.36 mmol/kg glucosinolate); E₃ = group given 30% RM (6.54 mmol/kg glucosinolate)

Table 6

Weights of some organs, grill processing, abdominal fat (g) and carcass yield (%) in chicks after a 42-day growing period (n = 30)

Parameter	Group of animals							
	C		E ₁		E ₂		E ₃	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Liver	36.5 ^{a,b,c}	8.1	39.4 ^a	7.1	41.3 ^b	10.5	38.7 ^c	4.4
Gizzard	29.6 ^a	4.5	30.2 ^b	4.8	30.8 ^c	5.2	28.1 ^{a,b,c}	5.2
Unevacuated intestines	85.5 ^{a,c}	18.1	105.4 ^{a,b}	24.8	112.3 ^{c,d}	12.4	96.0 ^{b,d}	12.4
Grill processing	1239.3 ^a	294.7	1241.7 ^b	250.5	1213.2 ^c	268.2	1120.7 ^{a,b,c}	159.4
Abdominal fat	32.2 ^{a,b}	11.8	32.5 ^{c,d}	12.3	30.2 ^{a,c}	8.1	27.4 ^{b,d}	8.1
Carcass yield, %	67.1 ^{a,b,c}	1.6	65.5 ^a	1.5	64.8 ^b	2.1	65.2 ^c	2.0

a:a, b:b, c:c, d:d = P < 0.05; C = control group; E₁ = group given 10% rapeseed meal (RM), (2.18 mmol/kg glucosinolate); E₂ = group given 20% RM (4.36 mmol/kg glucosinolate); E₃ = group given 30% RM (6.54 mmol/kg glucosinolate)

Pathohistological examination

Histological tests revealed pathological changes only in some internal organs and only in chicks in the experimental groups. Having completed the starter diet (day 21), chicks in Group E₁ were found to have mild-level hypertrophy of the thyroid gland epithelium, while those in Groups E₂ and E₃ had a medium-level hypertrophy. Mild hyperplasia of the thymus epithelium was found only in birds in

Group E₃. No pathological changes were found in heart, liver, kidneys, spleen, cloacal bursa, pancreas, testicles and suprarenal gland in any of the groups.

Upon completion of the finisher diet, i.e., at the end of the experiment, chicks in Groups E₁ and E₂ showed mild hypertrophy of the thyroid gland epithelium, as well as mild atrophy of the cloacal bursa follicles. Chicks in Group E₃ also had a mild atrophy of the cloacal bursa follicles, as well as mild thymus hypertrophy and medium-level hypertrophy of the thyroid gland epithelium.

Other organs tested showed no pathological changes in any of the groups of chicks in the experiment, which could be linked to the feed used, and through it to the quantity of glucosinolates in the diets fed to the chicks.

Discussion

The values found through chemical analyses of the 00-cultivar Silvia rapeseed meal fall within the parameters of chemical analyses of rapeseed meal (crude protein 32.5–37.8%; crude fat 1.8–4.4%; crude fibre 9.7–12.3% and ash 4.5–6.2% in 88% of dry matter) produced from cultivars used in Poland (Pastuszewska, 1992). The level of total glucosinolates in the seed (11.2) and in the meal (21.78 mmol/kg) produced from 00-cultivar Silvia rapeseed is below the average values found in the five Polish 00-cultivars with a low level of glucosinolates (Korol et al., 1994). Chemical analysis established that the glucosinolate structure of the Silvia cultivar is similar to that found in Polish and Canadian rapeseed cultivars (Slominski, 1998).

Rapeseed meal glucosinolates and products of their hydrolysis, respectively, i.e., thiocyanate ion (SCN⁻) and aglycone components, have a significant impact on the activity of the thyroid gland. They inhibit iodide transport across the basement membrane, while at the same time facilitating iodide discharge from the thyroid trap. Glucosinolate ingestion leads to a hypothyroid condition (primary hypothyroidism) characterized by increased thyroid activity due to the elevated level of the serum thyroid-stimulating hormone (TSH) and reduced circulating levels of thyroid hormones 3,5,3'-triiodothyronine (T₃) and tetraiodothyronine (T₄). The active form is T₃. It affects the transcription of target genes by way of the thyroid receptor (T₃R) and the thyroid response element (TRE) (Mendelson, 2000). Disruption in thyroid gland activity can affect body growth in two ways. First, it reduces levels of T₃ – which is of key importance for the expression of a gene that encodes membranes Na⁺-K⁺-pump or Na⁺-K⁺ ATPase (Griffin, 2000) accounting for 30% or more of the total ATP consumption of animal cells (Alberts et al., 1998). A diet containing glucosinolates, which has an inhibiting effect on gene expression of the above-mentioned ATPase, thereby reducing the body's oxygen consumption, could provide a distorted picture of their anabolic effect. The greatest grill weight in Group E₁ could be the result of such

activity. A part of the improved growth observed in the course of grill processing in chicks in Group E₁ could also be attributed to increased water retention in skin (myxoedema). Second, thyroid hormones act as tissue growth factors. T₃ could be responsible for the expression of the hormone growth gene since its receptors in the anterior lobe of the pituitary gland in rats and chickens manifest special properties (Griffin, 2000). In the absence of thyroid hormones, secretion of growth hormone is depressed (Ganong, 2001). Depending on its intensity, a protracted hypothyroidism inevitably causes stunted growth, the extreme form of the condition being dwarfism. Significantly stunted growth, alongside the significantly smallest grill weight ($P < 0.05$), was already observed in chicks in Group E₃ as early as the 21st day, while the same result was found at the end of the experiment. Towards the end of the experiment, chicks in Groups E₁ and E₂ also began to show lower growth rate in relation to Group C, which ultimately showed the highest weight.

Results of chicken body weight found in this experiment fully agree with results of researches conducted on broiler chicks by Chrappa et al. (1991), Khan et al. (1996) and Zeb et al. (1999), where chicks were fed a diet containing different quantities and cultivars of rapeseed meal, but they differ from results obtained by Richter et al. (1996) who fed their broiler chicks with feed diets containing only 5% of rapeseed meal, the outcome being reduced growth rate and body weight.

The behavioural tendency of feed conversion was proportional to the level of glucosinolates found in the experimental diets. The obtained conversion values, which showed no significant differences, indicate the effects of glucosinolates on thyroid function. In our research the feed conversion values agree with those found in experiments conducted with broiler chicks by Rojas-Ramirez et al. (1985), Wetscherek et al. (1991) and Khan et al. (1996), although they differ from the findings of Zavodsky et al. (1990) and Koncicki et al. (1991) who used different rapeseed cultivars.

Thyroid hypofunction results in disrupted metabolism of fats, signs of which were observed after just three weeks into the experiment. Thyroid hormone deficiency causes a decrease in the number of low-density lipoprotein receptors (LDL) on the cell surface and hypercholesterolaemia (Griffin, 2000). It also causes deficiency of tissue extracellular lipoprotein lipase (LPL), which is located on the luminal surface of capillary endothelial cells (Foster and McGarry, 2000). Due to the lower activity of LPL, the catabolism of chylomicrons and very-low-density lipoprotein (VLDL) particles is disrupted, which produces a reduction in the deposition of triglycerides into extrahepatic tissues, as well as hypertriglyceridaemia.

Liver weight in Group C chicks was significantly ($P < 0.05$) lighter than that in Groups E₁₋₃ after only three weeks of the experiment. At that point the heaviest liver weight was found in Group E₃. The same result was obtained at the

end of the experiment. Although the level of lipids in blood was not tested in the course of the experiment, we believe it rose considerably due to the fall in the LDL receptors and reduced activity of LPL. The result was an accumulation of lipids in liver, i.e., infiltration of fats, and a subsequent increase in liver weight. After three weeks the effect of this reduced function reflected only on liver, but at the end of the experiment it affected both liver weight and the quantity of abdominal fat. In Groups E₁₋₃ the quantity of abdominal fat, comprised of triacylglycerol, was insignificantly higher than in Group C (false anabolic effect), but at the end of the experiment it was significantly ($P < 0.05$) lower in Groups E₂ and E₃ than in Groups C and E₁; the lowest quantity of abdominal fat was found in Group E₃. This could be attributed to a more pronounced decrease in LPL function, which led to reduced deposition of triglycerides. Changes in liver weight and the quantity of abdominal fat resulting from hypothyroidism show that the first and more pronounced effect exerted by thyroid hormones is on the number of LDL receptors, and on LPL activity. The consequences of the assumed reduction in the number of LDL receptors and hypercholesterolaemia, as well as of disruptions in the synthesis and/or release of VLDL from liver, were – as previously mentioned – visible after 21 days.

A fall in LPL activity (not observed on day 21) appeared subsequently, and was apparent in Groups E₂ and E₃ at the end of the experiment.

Results with regard to liver weight yielded by our experiment agree with the findings of Trefny et al. (1989) and Karunajeewa et al. (1990), who found that birds fed on diets containing 14% and 17% of rapeseed meal had increased liver weight.

In our experiment chicks were fed a diet containing 20% and 30% of rapeseed meal (4.36 and 6.54 mmol/kg glucosinolates), and were found to have significantly less abdominal fat, results which coincide with those found by Würzner et al. (1989) who fed their broiler chicks diets containing 6% and more of rapeseed meal.

The fluctuating trend in the weight of an empty stomach and unevacuated intestines also resulted from the condition of hypothyroidism. Deficit of thyroid hormones slowed bowel transit, decreased motility and caused constipation, and this may contribute to the modest weight gain (Greenspan, 1997). Weights of gizzards were, in a regular pattern, inversely proportionate to the quantity of consumed glucosinolates, i.e., the level of hypothyroidism. The effect of thyroid hormones as growth factors can best be observed on gizzard weights at three weeks of age. This regularity disappears at the end of the experiment and is assumed to be due to the interfering influence of reduced basal metabolism (false anabolic effect) described previously, which could also be active in Groups E₁ and E₂, as well as due to the depressant effect on general growth rate, which continued to be dominant and visible in Group E₃.

Weight of intestines was crucially affected by their content. It can be assumed that the weight of discharged bowels would be similar in tendency to the weight of gizzards. At day 21, and at the end of the experiment, in Groups E₁ and E₂ it was exactly proportionate to the quantity of consumed glucosinolates, in other words the quantity of their content was proportionate to their motility, increasing from Group C towards Group E₂. The lower values in E₃ in relation to Group E₂ found after 21 days, and in E₃ in relation to E₁ and E₂ at 42 days of age, could be explained by poorer appetite resulting from hypothyroidism (Griffin, 2000) which, judging by the values of body weights and feed conversion, obviously existed.

The inhibiting effect of glucosinolates on thyroid function, and the effect of thyroid hormones, as well as their influence on the metabolism of fats and on digestive system, can also be noticed in the carcass yield, which was highest in Group C ($P < 0.05$) – already noticeable after 3 weeks, and which was also recorded at the end of the experiment. Our values relating to grill processing and carcass yield differ from the findings of Vymola et al. (1995), who fed chicks with diets containing 5, 10 and 15% of rapeseed meal (00 cultivar), and no differences were found in carcass yield between the control group and the experimental groups of chicks.

The histological changes to thyroid gland observed in experimental groups, ranging from mild to medium hypertrophy of epithelium, testify to the goitrogenic effect of glucosinolates. Hypertrophy is a consequence of a raised level of TSH in the blood. Lymphoid hyperplasia of thymus, found in Group E₃, could be the consequence of a stimulating effect of prolactin on the immune system function (Rillema, 1998). Increased discharge of prolactin provokes thyrotropin-releasing hormone (TRH) within the system of the negative feedback control into hypothyroidism (Aron et al., 1997). Pathohistological findings in chicks involved in our experiment agree fully with the findings of Rotkiewicz et al. (1990), Chrappa et al. (1991), Mawson et al. (1994) and Khan et al. (1996), who fed their chicks with diets containing 10%, 15% and more, respectively, thus achieving a significant goitrogenic effect of glucosinolates contained in rapeseed meal. The results differ from those obtained by Kloss et al. (1994), who observed no pathohistological changes in internal organs of chicks fed on 10% of crambe (*Crambe abyssinica*) meal per ration.

Generally, a dietary glucosinolate concentration of 2 mmol/kg has been indicated as a no-effect level for glucosinolates in monogastric animals (Campbell and Schöne, 1998). Our results cast doubt on the above. Chickens in Group E₁, fed on rations with glucosinolate concentrations of 2.18 mmol/kg showed signs of disrupted metabolism after the 21st day as well as at the end of the experiment. Signs included a significantly increased weight of liver and decreased weight of gizzard, and significantly lower carcass yield. A false anabolic effect, manifested through values of average body weight, was also observed.

References

- Alberts, B., Bray, D., Jonson, A., Lewis, J., Raff, M., Roberts, K. and Walter, P. (1998): *Essential Cell Biology*. Garland Publishing, Inc. New York and London.
- Allen, R. (1993): Ingredient analysis table. *Feedstuffs Issue* **65**, 24–37.
- A.O.A.C. (1984): *Official Methods of Analysis* (14th edition). Association of Official Analytical Chemists, Arlington, VA, USA.
- Aron, D. C., Finding, W. J. and Tyrrell, J. B. (1997): Hypothalamus and Pituitary. In: Greenspan, F. S. and Strewler, G. J. (eds) *Basic and Clinical Endocrinology*, 5th edition. Prentice-Hall International, London. pp. 95–156.
- Campbell, L. D. and Schöne, F. (1998): Effects of antinutritional factors in rapeseed. In: Jansman, A. J. M., Hill, G. D., Huisman, J. and van der Poel, A. F. B. (eds) *Recent Advances of Research in Antinutritional Factors in Legume Seed and Rapeseed*. EAAP Publication No. 93. Wageningen Pers., Wageningen. pp. 185–198.
- Chrappa, V., Stražnicka, H., Abelova, H. and Sabo, V. (1991): Effects of feeding with rapeseed 'OO' on efficiency of broiler chicks. *Živočišna Vyroba* **36**, 437–448.
- Foster, D. W. and McGarry, J. D. (2000): Glucose, Lipid, and Protein Metabolism. In: Griffin, J. E. and Ojeda, S. R. (eds) *Textbook of Endocrine Physiology*. Oxford University Press, Oxford–New York. pp. 393–419.
- Fritz, Z., Lipstein, B., Kinal, S., Šplitek, M. and Pašmik, M. (1993): Effect of pre-pressed rapeseed in broiler diets. *Archiv für Geflügelkunde* **57**, 175–180.
- Ganong, W. F. (2001): The Thyroid Gland. In: *Review of Medical Physiology*. 20th edition. Lange Medical Books/McGraw-Hill, New York. pp. 307–321.
- Gobec, S. and Ljubić, I. (1970): Slaughterhouse research of the Nichols-Lohman hybrid line of broilers (in Croatian). *Tehnologija mesa* **11**, 306–310.
- Grala, W., Buraczewska, L., Gdala, J. and Pastuszewska, B. (1994): Effect of the thermal processing on the protein value of double low rapeseed products. *Int. J. Anim. Feed Sci.* **3**, 33–42.
- Grbeša, D., Černy, T. and Homen, B. (1994): Chemical composition and nutritive values of fodder for ruminants in Croatia (in Croatian). *Stočarstvo* **48**, 1–2.
- Greenspan, F. S. (1997): The Thyroid Gland. In: Griffin, J. E. and Ojeda, S. R. (eds) *Textbook of Endocrine Physiology*. Oxford University Press, Oxford–New York. pp. 192–262.
- Griffin, J. E. (2000): The Thyroid. In: Griffin, J. E. and Ojeda, S. R. (eds) *Textbook of Endocrine Physiology*. Oxford University Press, Oxford–New York. pp. 303–327.
- Haščik, P., Kovač, M. and Hanzlik, K. (1994): Substitution of rapeseed oilcake for soyabean meal during the second feeding phase of broilers. *Živočišna Vyroba* **39**, 1041–1047.
- ISO-9167-1-1992: Rapeseed determination of glucosinolates content. Part 1: Method using high-performance liquid chromatography. First edition, 1992. Geneva, Switzerland.
- Jamroz, D. (1995): Use of rape and its by-products in the feed of poultry (in Croatian). *Krmiva* **37**, 175–190.
- Karunajeewa, H., Ljagbuji, E. G. and Reece, R. L. (1990): Effect of dietary levels of rapeseed meal and polyethylene glycol on the performance of male broiler chicks. *Br. Poultry Sci.* **31**, 545–555.
- Khan, M. Z., Sarwar, M., Mahmood, S. and Frebaz, A. (1996): Effect of various levels of rapeseed meal on the performance of broilers. *Pakistan Vet. J.* **16**, 192–195.
- Kloss, P., Jeffery, E., Wallig, M., Tumbleson, M. and Parsons, C. (1994): Efficacy of feeding glucosinolate-extracted crumble meal to broiler chicks. *Poultry Sci.* **73**, 1542–1551.
- Kolodziej, J. (1995): Rape '00' in feeding domestic animals in Poland (ruminants, pigs, poultry) (in Croatian). *Krmiva* **37**, 191–219.
- Koncicki, A., Krasnodebska-Depta, A., Faruga, A., Mikulski, D., Kozłowski, M., Kozłowska, H., Janowska, I. and Rotkiewicz, D. (1991): Effect of complete feed diets containing various rapeseed meals on selected haematological and biochemical indices in broiler chicks. *Zeszyty Naukowe Akademii Rolniczej we Wrocławiu, Weterynaria* **48**, 97–105.

- Korol, W., Jaskiewicz, T., Bartuzi, G., Bogusz, G., Niescior, H., Grabowski, C. and Mojek, E. (1994): Chemical composition of rape seed from low glucosinolate varieties grown in Poland. *J. Anim. Feed Sci.* **3**, 57–64.
- Mawson, R., Heaney, R. K., Zdunczyk, Z. and Kozłowska, H. (1993): Rapeseed meal-glucosinolates and their anti-nutritional effects. 2. Flavour and palatability. *Die Nahrung* **37**, 336–344.
- Mawson, R., Heaney, K., Zdunczyk, Z. and Kozłowska, H. (1994): Rapeseed meal glucosinolates and their antinutritional effects. Part 4. Goitrogenicity and internal organs abnormalities in animals. *Die Nahrung* **38**, 178–191.
- Mendelson, C. R. (2000): Mechanics of Hormone Action. In: Griffin, J. E. and Ojeda, S. R. (eds) *Textbook of Endocrine Physiology*. Oxford University Press, Oxford–New York. pp. 51–88.
- Mustapić, Z. and Pospišil, M. (1995): Quality of oil and rapeseed meal of new '00' rapeseed cultivars (in Coatian). 11th International Symposium of Technologies for Drying and Storing. *Zbornik radova*. pp. 66–73.
- Pastuszevska, B. (1992): Rapeseed in the Nutrition of Animals (in Polish). Omintech Press, Warsaw.
- Richter, G., Lemser, A., Lüdke, H. and Carlsohn, H. (1996): The use of rapeseed and rapeseed meal in chicken and pullet mixtures. *Wirtschaftseigene Futter* **42**, 67–82.
- Rillema, J. A. (1998): Action of Prolactin. In: Knobil, E. and Neill, J. D. (eds) *Encyclopedia of Reproduction*. Volume 4. Academic Press, San Diego, pp. 39–43.
- Rojas-Ramirez, E., Gonzales, E. A. and Tirado, A. J. (1985): Nutritive value of rapeseed oil meal and its effect on performance of broiler chicks and laying hens. *Tecnica Pecuaria en Mexico* **49**, 135–142.
- Rotkiewicz, T., Ruta, A., Kozłowski, M., Faruga, A., Mikulski, D., Rotkiewicz, D. and Kozłowska, H. (1990): Pathomorphology of thyroid glands and liver of broiler chicks fed diets containing rapeseed oil meal of various cultivars. *Acta Academiae Agriculturae et Technicae Olstenensis. Veterinaria* **19**, 159–174.
- SAS Institute, Inc. (1996): SAS/STAT® Software. Changes and Enhancements through Release 6.11, Cary, NC: SAS Institute Inc.
- Slominski, B. A., Campbell, L. D. and Stanger, N. E. (1988): Extent of hydrolysis in the intestinal tract and potential absorption of intact glucosinolates in the laying hens. *J. Sci. Food Agric.* **42**, 305–314.
- Trefny, D., Sova, Z., Petkov, S., Fukal, L., Fučíkova, A., Vrabec, P., Vit, M. and Žak, P. (1989): Physiological aspects of using rapeseed oil meal in fattening broiler chicks. *Živočišna Vyroba* **51**, 17–30.
- Vymola, J., Kodes, A. and Obadalek, J. (1995): Rapeseed cake in diets of broiler chicks. *Živočišna Vyroba* **40**, 407–409.
- Wetscherek, W., Lettner, F. and Würzner, H. (1991): Rapeseed meal in broiler diets. *Archiv für Geflügelkunde* **54**, 57–61.
- Wetscherek, W., Lettner, F., Steinwieder, A. and Lorenz, T. (1993): Rapeseed by-products in poultry finisher diets. *Forderungsdienst* **41**, 320–325.
- Würzner, H., Wetscherek, W. and Lettner, F. (1989): Rapeseed meal in rations for broilers. *Archiv für Geflügelkunde* **53**, 6–12.
- Yu, B. C., Funan, X., Chunrong, S., Wenfeng, S. and Jouping, S. (1995): Histological changes in livers, kidneys and thyroid glands of broiler chicks fed on different rapeseed meals. *Jiangsu J. Agric. Sci.* **11**, 23–28.
- Zavodsky, B., Klecker, D. and Voda, M. (1990): Use of low-glucosinolate rapeseed oil meal in fattening broilers. *Sbornik Vedeckych Praci* **23**, 75–81.
- Zeb, A., Satter, U. and Meulen, U. Ter. (1999): Effect of feeding different levels of rapeseed meal on the performance of broiler chicks. *Archiv für Geflügelkunde* **63**, 77–81.