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The effect of various iodine supplementations and two different iodine sources on performance and iodine concentrations in different tissues of broilers

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Abstract 1. The objective of this study was to determine the influence of iodine (I) supplementation of feed, within the range of the European guidelines, on the performance of broiler chickens and I transfer into different organs and tissues, especially meat. The main emphasis was to assess whether broiler meat could be enriched and used as an I source in human nutrition.

2. Two experiments were performed, one with KI and the other with Ca(IO₃)₂. For each experiment, 288 d-old broiler chicks were divided into 4 groups (72 birds/group) and fed on diets with supplementations between 0 and 5 mg I/kg feed. The birds were reared to 35 d of age under standard conditions. Six birds per group were slaughtered at 35 d and samples of blood, thyroid gland, liver, pectoral and thigh meat taken.

3. Iodine treatment did not significantly affect the growth and slaughter performance of the broiler chickens. In all investigated parameters, I concentrations increased significantly with increasing I intake of the animals. The lowest I concentrations were measured in the meat, but they were considerably higher in blood serum, liver and thyroid gland. Since the I content of meat was still low in the highest supplemented group (highest median concentration: 67.8 µg I/kg thigh meat), there is no evidence that this could substantially improve I supply in human nutrition.

INTRODUCTION

Iodine (I) deficiency is still a widely prevalent problem in human nutrition around the world, approximately 57% of the European population and 35% of the world's population having insufficient iodine intake (urinary iodine <100 µg/L; de Benoist *et al.*, 2004). I is an essential trace element for humans and animals, being required for the synthesis of the thyroid hormones triiodothyronine (T₃) and tetraiodothyronine (T₄). The appropriate I provision is important for a normal thyroid function and the associated physiological mechanisms. For an adult person, the D-A-CH (German-, Austrian- and Swiss Nutrition Society) recommends a daily intake of 180–200 µg I and a 'Tolerable Upper Intake

Level' (UL) of 500 µg I/d (D-A-CH, 2008). I is assigned to the 'high risk category' and also to 'supply category 1'. This means there is only a narrow range between recommended daily intake and UL (≈1:3) (Gaßmann, 2006). Due to adverse health effects, deficiency as well as excessive intake should be avoided. A lack in I supply impairs thyroid hormone synthesis, which results in hypothyroidism and leads to various developmental and functional disorders, summarised as 'Iodine Deficiency Disorders' (IDD; Hetzel, 1983; de Benoist *et al.*, 2004). IDD can affect the organism in all stages of life: foetus (*e.g.*, abortions, endemic cretinism), neonate (*e.g.*, endemic mental retardation), child and adolescent (*e.g.*, goitre, retarded physical development) and adult (*e.g.*, hypothyroidism, spontaneous

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hyperthyroidism in the elderly) (Hetzl, 1983; Stanbury *et al.*, 1998; Laurberg *et al.*, 2000).

To counteract I deficiency, salt iodination was implemented, but this did not solve the problem to a satisfactory extent, so further I sources were sought. Products of animal origin, such as milk or eggs, can, for example, be enriched by feed iodination. Feed iodination is limited by EU regulations to avoid I excess, the maximum allowance of feed iodination for broiler chickens being 10 ppm (EU Commission, 2005). Due to high carry-over into milk and eggs, the UL for laying hens and dairy cows was reduced from 10 ppm to 5 ppm (EU Commission, 2005). The I requirements of animals depend on species, age and physiological stage. The maximum allowance for broiler feed of 10 mg I/kg compares with a requirement of 0.35 mg I/kg feed for broilers (NRC, 1994). Thus health and performance of the animals have to be considered at the intention of feed iodination and possible elevation of the legal feed UL.

The present study encompassed two feeding experiments with broilers. A range of I supplementations and two different I sources were tested to investigate the influence on I concentrations of different tissues and the impact on growth and slaughter performance. Specifications for the I content of poultry meat are absent from the nutrition table of Souci *et al.* (2008); this shows the necessity of determining these data. The motivation for this study was a document of the European Food Safety Authority (EFSA, 2005) about the use of I in feeding stuffs, which concluded that more dose-response studies were necessary to assess the resulting I content in food of animal origin. The experiments were conducted to investigate the impact of broiler feed iodination on the I concentrations of tissues, organs and blood serum. The main interest of the study was to determine to what extent chicken meat can be enriched with I when broilers ingest feed supplied with I according to EU guidelines. As different I sources are permitted as feed additives in the EU (EU Commission, 2005), two of them were tested – potassium iodide (KI) and calcium iodate ($\text{Ca}(\text{IO}_3)_2$). Growth and slaughter performance were determined in these feeding experiments to evaluate the tolerance of the broilers towards different I concentrations.

MATERIALS AND METHODS

Animals, diets and sampling

Two experimental approaches were conducted to test the two different I sources – iodide (KI) and iodate ($\text{Ca}(\text{IO}_3)_2$). Both experiments were conducted at the same experimental station

(Friedrich-Loeffler-Institute, Institute of Animal Welfare and Animal Husbandry, Celle, Germany) under identical experimental conditions. The experimental approach testing KI was conducted in April/May 2007; the approach testing $\text{Ca}(\text{IO}_3)_2$ was carried out in March/April 2008.

Each of the two experimental trials was performed as follows. Ten Ross 308 broiler breeder hatching eggs which had been produced under standard management conditions were used to determine the initial I concentration. On the day of hatch, 288 male chicks were divided into 4 groups (72 per group) and a further 10 chicks killed for I determination. The birds of each group were allocated to 6 cages (1.28×0.82 m) with 12 birds (11.4 chickens/ m^2) per cage and reared to 35 d under standard management conditions. Feed and water were provided *ad libitum*. The feed composition is detailed in Table 1. Feed intake (FI) was recorded by cage weekly and body weight (BW) measured on a cage basis at 7 d and individually at 14, 21 and 35 d, and the data used to calculate the feed conversion ratio (FCR).

KI and $\text{Ca}(\text{IO}_3)_2$ were tested as different I sources in a range of concentrations (0 (control), 1, 2.5 and 5 mg I/kg feed). The whole feed was mixed prior to each experiment. An I-free mineral premix was used for the composition of the feed mixtures. Pre-mixtures were produced to achieve a homologous dispersion of I in the feed. In a first step, the calculated I amounts were applied to 70 g ground wheat, then 70 g were mixed into 2 kg ground wheat, and finally this pre-mixture was mixed into the 300-kg feed mixture. The control group were given the same basic diet as the test groups but without additional I; these birds only received naturally occurring I from the feed ingredients. Only I concentrations of up to 5 mg I/kg were tested, despite 10 mg I/kg being allowed for growing birds, because other experiments conducted with laying hens used 5 mg I/kg.

On d 35, one bird from each cage with a BW close to the mean of the respective cage-group was slaughtered by electrical stunning and severance of the carotid artery and jugular vein according to welfare regulations (EU Commission, 2008). The blood was collected into test tubes [10 mL, SARSTEDT, No./REF 26.323] containing a clot activator to retrieve blood serum. Afterwards, samples were taken from the thyroid gland, liver, pectoral and thigh meat of the carcass for weighing and I determination. Heart, liver, ventriculus (gizzard), spleen, breast skin, breast meat, thigh, abdominal fat and empty body (empty carcass, without head, innards and tibiotarsi) were weighed to determine the carcass composition of the birds.

Table 1. Composition of the diets for KI and Ca(IO₃)₂ supplemented groups, without I supplementations

Composition of the feed mixture (g/kg)		
Wheat	200.0	
Maize	336.0	
Soybean oil	42.9	
Soybean meal	196.7	
Dicalcium phosphate	22.0	
Calcium carbonate	4.6	
Salt	3.2	
DL-Methionine	2.9	
L-Lysine-HCl	1.7	
Soy beans	180.0	
Mineral/vitamin premix ¹	10.0	
<i>Analysed and calculated composition (g/kg)</i>		
	KI	Ca(IO ₃) ₂
Dry matter	902.4	893.6
Crude protein (CP)	196.6	190.3
Crude fat (EE)	95.0	102.7
Starch (ST)	364.3	352.2
Sugar (S)	47.8	38.7
Lysine ²	12.5	12.5
Methionine + cysteine ²	9.6	9.6
Methionine ²	6.0	6.0
Metabolisable Energy (MJ/kg) ³	13.01	12.86

¹Content per kg premix: 360 mg vitamin A, 75 mg cholecalciferol, 3000 mg 2-*ambo*- α -tocopherol, 200 mg thiamin, 480 mg riboflavin, 360 mg pyridoxine, 1.5 mg cyanocobalamin, 300 mg menadione, 2700 mg nicotinic acid, 900 mg Ca-pantothenic acid, 90 mg folic acid, 5 mg biotin, 80 000 mg choline chloride, 5000 mg Fe, 1500 mg Cu, 12 000 mg Mn, 8000 mg Zn, 40 mg Se, 55 mg Co.

²Calculated from amino acid contents of feeds.

³Calculated *via* the energy estimation formula, of the WPSA (1984).

Analysis of feed composition

Feed analyses were performed of the feed mixtures from each experimental group. Dry matter, crude fibre, crude ash, crude protein (CP), crude fat (EE), starch (ST), sucrose (S), NDF, ADF and Ca were analysed according to the guidelines of "Verband Deutscher Landwirtschaftlicher Untersuchungs- und Forschungsanstalten" (VDLUFA, 1976).

I analyses

The I contents were measured using Inductively Coupled Plasma-Mass Spectrometry (ICP-MS) [Agilent 7500C]. The measuring solutions were prepared in an alkaline medium in order to avoid vaporisation of I (Wagner, 2007). Feed samples (5 g) were boiled for 30 min with ammonia solution (0.59 mol/L). After cooling, samples were filled with ammonia solution up to 1000 mL, and then filtered with a fluted filter. Prior to use, filters were flooded twice with boiling distilled deionised water and then dried in an oven to remove possible I and avoid contamination. After filtration with a syringe filter (Minisart 0.45 μ m), 10 mL of the filtrate were taken for measurements. Blood serum

was diluted 1:10 with 0.11 mol/L Tetramethylammonium hydroxide solution [TMAH, Alfa Aesar GmbH & Co KG Karlsruhe, 25% w/w aq.soln., Electronic Grade, 99.9%] before measuring. Meat, liver, 1-d-old chicks and egg samples were each homogenised with a mixer [ESGE-ZauberStab[®]] prior to digestion, with the exception of the thyroid gland, which was taken in whole due to its small size. Therefore, the thyroid weight was included into the calculation for its I concentration. After mechanical homogenisation, meat, liver, eggs and 1-d-old chicks were digested chemically with TMAH (Fecher *et al.*, 1998) and a 1-g sample heated for 3 h at 90°C in a gas-tight Erlenmeyer flask, with 5 mL distilled deionised water and 1 mL TMAH. After cooling, the solution was filled up to 25 mL with distilled deionised water. 10 mL of the solution were taken for measurement using ICP-MS, 0.5 mL tellurium oxide solution [100 μ g/L; tellurium (IV) oxide, Alfa Aesar GmbH & Co KG Karlsruhe, 99.9%] as the internal standard. The masses of 127 for I and 125 for Te were used. During calculation of I concentrations, the standard addition method was applied during calibration.

Statistical analyses and calculations

The carry-over was calculated as the percentage of I content of the sample from the total amount of I which was ingested during the whole experiment (35 d). Nitrogen-corrected apparent metabolisable energy, was calculated with the energy estimation formula: $AME_N [MJ/kg] = (15.51 \text{ g CP/kg} + 34.31 \text{ g EE/kg} + 16.69 \text{ g ST/kg} + 13.01 \text{ g S/kg}) / 1000$ (WPSA, 1984 in GFE, 1999).

The statistical analyses were carried out with SAS 9.1 (© by SAS Institute Inc., Cary, NC, USA) and STATISTICA 8.0 (StatSoft, Inc. 2007). Differences were recognised as significant at $P < 0.05$. Since the two experiments were carried out at two different points in time, the effects of iodate and iodide were not compared with each other. Outliers were detected with the ROBUSTREG procedure and then excluded from subsequent analysis. Afterwards, data were tested for normality with the procedure UNIVARIATE, using the Kolmogorov-Smirnov test ($P = 0.01$). Performance and slaughter data were normally distributed and thus analysed parametrically with the Tukey test using the GLM procedure. The data of I concentrations showed significant differences from the normal distribution. These not normally distributed data were analysed non-parametrically with the Kruskal-Wallis test using STATISTICA.

RESULTS

Feed I concentrations

The naturally occurring I concentration of the feed mixtures was 0.5 mg I/kg in the control diet of both experiments. The I concentrations of the groups supplemented with 1, 2.5 and 5 mg I/kg approximated to the desired concentrations of 0.6, 2.4 and 5.5 mg I/kg respectively in KI groups and 0.9, 2.7 and 4.7 mg I/kg in Ca(IO₃)₂ groups.

Rearing performance

The I supplementation had no significant effect on FI and BW gain (BWG; Tables 2 and 3). The mean FI of all groups and total experimental period was 98.2 ± 3.3 g/d in KI groups and 97.2 ± 3.6 g/d in Ca(IO₃)₂ groups. The mean BWG was 65.1 ± 2.2 g/d in KI groups and 67.3 ± 2.4 g/d in Ca(IO₃)₂ groups.

The FCR of KI-birds was significantly improved between 21 and 35 d, where the 5 mg I/kg supplemented group had a significantly superior FCR to the control group (1.61 kg/kg versus 1.68 kg/kg). There was no significant effect on FCR during the other phases (Table 2). When testing FCR through to 35 d, the FCR of the 5 mg I/kg (1.49 kg/kg) was also significantly higher than in the control (1.53 kg/

kg; $P=0.0071$). In the Ca(IO₃)₂ broilers no significant differences were found between the different groups, the average FCR of all Ca(IO₃)₂ groups was 1.45 ± 0.04 kg/kg. In contrast to KI groups the highest supplemented group had in tendency a lower FCR than the control group. The significant positive effect in FCR, found in the 5 mg I/kg KI group was caused by negligibly low standard deviation and was not considered to have biological relevance.

Slaughtering performance

The data retrieved at slaughtering are listed in Tables 4 and 5. The final BW was not significantly affected by the various I concentrations, neither with KI supplementation (2230 ± 87 g) nor with Ca(IO₃)₂ supplementation (2303 ± 58 g) birds.

No significant differences were found in the mass of the different organs and fractions or in their proportions of final BW as influenced by I supplementation. Considering the masses of samples gained from KI- and Ca(IO₃)₂-supplemented birds, the mean weight of pectoral meat was 340.1 g and 371.1 g, of breast skin 26.2 g and 29.7 g and thigh mass with bone and skin 440.5 g and 472.0 g respectively. In the other fractions and organs, mean weights were: heart 10.6 g and 11.5 g, liver 47.0 g and 44.9 g, spleen 1.93 g and 2.19 g, ventriculus 24.7 g and 27.7 g and

Table 2. Mean ± SD feed intake (FI), body weight gain (BWG) and feed conversion ratio (FCR) of broilers fed diets supplemented with KI to give I concentrations of 0, 1, 2.5 or 5 mg/kg (n = 6)

Iodine supplementation (mg I/kg feed)	FI (g/d)	BWG (g/d)	FCR (kg/kg)
0-14 d			
0	39.3 ± 1.5	30.9 ± 1.7	1.25 ± 0.02
1.0	37.8 ± 1.2	30.4 ± 1.6	1.24 ± 0.03
2.5	39.3 ± 0.5	31.2 ± 0.7	1.26 ± 0.02
5.0	38.1 ± 1.2	30.5 ± 1.6	1.25 ± 0.03
14-21 d			
0	99.0 ± 3.6	72.0 ± 2.2	1.37 ± 0.04
1.0	97.5 ± 3.0	71.7 ± 2.1	1.36 ± 0.01
2.5	101.0 ± 2.8	72.9 ± 3.0	1.39 ± 0.06
5.0	97.9 ± 2.9	71.2 ± 2.2	1.38 ± 0.01
21-35 d			
0	156.9 ± 5.6	93.3 ± 3.9	1.68 ^a ± 0.03
1.0	154.9 ± 5.6	94.3 ± 4.6	1.64 ^{ab} ± 0.04
2.5	160.5 ± 7.4	98.8 ± 5.2	1.63 ^{ab} ± 0.04
5.0	157.4 ± 6.0	97.7 ± 3.2	1.61 ^b ± 0.05
0-35 d			
0	98.3 ± 3.4	64.1 ± 2.3	1.53 ^a ± 0.02
1.0	96.5 ± 2.6	64.2 ± 2.1	1.50 ^{ab} ± 0.01
2.5	100.1 ± 3.5	66.6 ± 2.3	1.50 ^{ab} ± 0.02
5.0	97.7 ± 3.2	65.5 ± 1.7	1.49 ^b ± 0.02

^{a,b}Within a column and time period, means without a common superscript are significantly different at $P < 0.05$.

Table 3. Mean ± SD feed intake (FI), body weight gain (BWG) and feed conversion ratio (FCR) of broilers fed diets supplemented with Ca(IO₃)₂ to give I concentrations of 0, 1, 2.5 or 5 mg/kg (n = 6)

Iodine supplementation (mg I/kg feed)	FI (g/d)	BWG (g/d)	FCR (kg/kg)
0-14 d			
0	36.9 ± 1.0	29.8 ± 0.9	1.24 ± 0.01
1.0	36.9 ± 1.5	30.0 ± 1.5	1.23 ± 0.03
2.5	37.2 ± 1.7	30.3 ± 1.5	1.23 ± 0.02
5.0	36.9 ± 0.8	30.1 ± 0.7	1.23 ± 0.01
14-21 d			
0	94.2 ± 4.0	70.6 ± 3.2	1.33 ± 0.04
1.0	95.4 ± 4.3	70.2 ± 3.7	1.36 ± 0.02
2.5	94.4 ± 5.0	69.7 ± 4.0	1.35 ± 0.03
5.0	93.6 ± 2.1	67.9 ± 2.3	1.38 ± 0.02
21-35 d			
0	163.6 ± 6.2	107.2 ± 4.0	1.53 ± 0.04
1.0	159.6 ± 7.3	102.8 ± 5.6	1.55 ± 0.06
2.5	160.0 ± 5.5	102.7 ± 2.8	1.56 ± 0.05
5.0	158.0 ± 5.9	100.6 ± 5.2	1.57 ± 0.04
0-35 d			
0	98.1 ± 4.7	68.9 ± 1.9	1.42 ± 0.04
1.0	97.3 ± 3.8	67.2 ± 2.9	1.45 ± 0.04
2.5	97.4 ± 3.6	67.1 ± 2.0	1.45 ± 0.03
5.0	95.9 ± 2.8	65.8 ± 2.1	1.46 ± 0.02

Table 4. Mean \pm SD organ and fraction weights of broilers fed diets supplemented with KI to give I concentrations of 0, 1, 2.5 or 5 mg/kg (n = 6)

	Iodine supplementation (mg/kg)			
	0	1.0	2.5	5.0
Final body weight (g)	2211.2 \pm 120.4	2184.7 \pm 53.4	2226.8 \pm 49.2	2296.0 \pm 82.3
Pectoral meat (g)	349.5 \pm 40.6	323.0 \pm 16.2	341.8 \pm 30.1	347.1 \pm 13.6
Breast skin (g)	28.5 \pm 6.4	25.4 \pm 3.3	25.7 \pm 3.7	25.1 \pm 3.8
Thigh (g)	437.9 \pm 18.7	439.1 \pm 11.8	439.9 \pm 33.9	445.6 \pm 13.6
Heart (g)	11.1 \pm 2.3	11.0 \pm 1.3	9.6 \pm 0.3	10.8 \pm 1.4
Liver (g)	46.7 \pm 3.7	44.4 \pm 2.6	47.2 \pm 1.2	49.5 \pm 4.5
Spleen (g)	1.95 \pm 0.48	1.74 \pm 0.11	1.95 \pm 0.32	2.07 \pm 0.33
Ventriculus (g)	28.8 \pm 7.2	24.2 \pm 4.2	22.2 \pm 4.7	23.7 \pm 4.5
Abdominal fat pad (g)	27.0 \pm 11.3	32.9 \pm 6.9	34.3 \pm 6.9	38.7 \pm 6.2
Thyroid gland (g)	0.14 \pm 0.03	0.15 \pm 0.03	0.17 \pm 0.06	0.16 \pm 0.05

Table 5. Mean \pm SD organ and fraction weights of broilers fed diets supplemented with Ca(IO₃)₂ to give I concentrations of 0, 1, 2.5 or 5 mg/kg (n = 6)

	Iodine supplementation (mg/kg)			
	0	1.0	2.5	5.0
Final body weight (g)	2285.8 \pm 39.8	2340.5 \pm 82.8	2302.5 \pm 55.8	2284.3 \pm 37.7
Pectoral meat (g)	348.5 \pm 34.2	377.6 \pm 44.6	383.6 \pm 45.5	374.9 \pm 34.4
Breast skin (g)	27.9 \pm 2.9	29.2 \pm 8.3	31.8 \pm 5.7	30.2 \pm 5.4
Thigh (g)	471.2 \pm 12.8	469.7 \pm 27.8	472.4 \pm 17.0	474.5 \pm 28.3
Heart (g)	12.4 \pm 1.3	11.4 \pm 1.4	10.8 \pm 1.3	11.4 \pm 1.5
Liver (g)	44.7 \pm 2.4	45.3 \pm 3.9	44.2 \pm 3.5	45.5 \pm 2.7
Spleen (g)	2.08 \pm 0.17	2.40 \pm 0.49	2.03 \pm 0.34	2.20 \pm 0.30
Ventriculus (g)	25.0 \pm 3.7	30.2 \pm 6.0	28.2 \pm 6.6	27.5 \pm 4.3
Abdominal fat pad (g)	32.4 \pm 8.8	27.3 \pm 4.5	32.3 \pm 6.4	28.1 \pm 5.1
Thyroid gland (g)	0.23 \pm 0.13	0.17 \pm 0.04	0.18 \pm 0.07	0.17 \pm 0.04

abdominal fat pad 33.5 g and 30.0 g respectively for KI and Ca(IO₃)₂ supplementation. The thyroid gland had a mean weight of 0.15 \pm 0.04 g in KI and 0.19 \pm 0.08 g in Ca(IO₃)₂ broilers.

I concentrations

The hatching eggs ($n = 10$) had I concentrations of 687 \pm 170 μ g I/kg in the KI experiment and 484 \pm 82 μ g I/kg in the Ca(IO₃)₂ experiment. The 1-d-old chicks had concentrations of 749 \pm 107 μ g I/kg in the KI experiment and 565 \pm 148 μ g I/kg in the Ca(IO₃)₂ experiment. I content was less than 1% of the final I concentration found in the slaughtered birds at the end of the experiment, and so the initial I content was ignored in carry-over calculations.

Increasing I supplementation significantly raised the I concentration in all investigated tissues and blood serum of the birds (Tables 6 and 7). The order of I concentrations in the different investigated samples was meat < liver < serum < thyroid gland.

I concentration of the thyroid was independent of feed I supplementation, the highest of the various samples, and increased significantly with rising feed I concentration. In KI groups, it ranged from 2317 to 5053 μ g I/g and in Ca(IO₃)₂ groups from 2323 to 3652 μ g I/g. Calculated on the weight of the thyroid gland, the I content of one thyroid gland of the highest KI supplemented group had a mean I content of 825 \pm 373 μ g and in the highest Ca(IO₃)₂ groups the I content of one thyroid gland was 601 \pm 197 μ g. The carry-over of I into the thyroid was higher than the other samples. Regarding both I sources, the carry-over decreased with increasing feed I concentration, the median amount in the control group of KI birds was 18.0% and 21.8% in Ca(IO₃)₂ birds. In the highest KI group, the median carry-over into the thyroid was 4.2% and, in the highest Ca(IO₃)₂ group, 3.5%.

In comparison with liver and meat samples, the I concentration of blood serum was relatively high. The I concentration in the control groups

Table 6. I concentration of samples collected from broilers fed diets supplemented with KI to give I concentrations of 0, 1, 2.5 or 5 mg/kg (median, Q25, Q75; n = 6). Median values shown in bold

	Iodine supplementation (mg/kg)											
	0			1.0			2.5			5.0		
Pectoral meat (µg/kg)	5.8^c	5.0	7.0	10.3^{bc}	9.1	10.4	39.3^{ab}	37.1	42.9	58.0^a	52.3	59.5
Thigh meat (µg/kg)	5.9^b	5.5	7.3	6.9^b	6.1	9.4	38.6^{ab}	32.3	44.8	63.1^a	58.2	66.1
Liver (µg/kg)	22.4^c	20.0	24.7	31.0^{bc}	27.2	35.4	103.6^{ab}	97.8	131.9	173.4^a	163.8	197.1
Blood serum (µg/L)	29.1^c	27.2	30.3	50.0^{bc}	41.9	68.5	226.4^{ab}	221.5	305.6	382.4^a	366.3	430.8
Thyroid gland (µg/g)	2317^b	1991	2385	3315^{ab}	3071	3448	4599^a	4304	4812	5053^a	4446	5899

^{a,b,c}Within a row, means without a common superscript are significantly different at $P < 0.05$.

Table 7. I concentration of samples collected from broilers fed diets supplemented with $\text{Ca}(\text{IO}_3)_2$ to give I concentrations of 0, 1, 2.5 or 5 mg/kg (median, Q25, Q75; n = 6). Median values shown in bold

	Iodine supplementation (mg/kg)											
	0			1.0			2.5			5.0		
Pectoral meat (µg/kg)	7.0^c	6.3	8.7	11.5^{bc}	10.6	12.7	27.9^{ab}	25.1	30.8	52.1^a	48.5	54.9
Thigh meat (µg/kg)	12.2^b	11.3	16.7	22.9^b	12.8	28.8	37.7^{ab}	35.6	41.1	67.8^a	62.9	78.4
Liver (µg/kg)	28.1^c	27.5	32.1	44.1^{bc}	41.9	47.6	104.5^{ab}	97.7	112.0	181.3^a	175.0	198.7
Blood serum (µg/L)	28.2^c	23.8	34.1	50.5^{bc}	48.0	68.9	179.4^{ab}	172.9	198.9	349.9^a	318.5	386.5
Thyroid gland (µg/g)	2323^b	1927	2723	3170^{ab}	2960	3517	3117^{ab}	2740	4206	3652^a	3154	3840

^{a,b,c}Within a row, means without a common superscript are significantly different at $P < 0.05$.

was 29 µg I/L (KI groups) and 28 µg I/L ($\text{Ca}(\text{IO}_3)_2$ groups) and increased to 382 µg I/L (KI groups) and 350 µg I/L ($\text{Ca}(\text{IO}_3)_2$ groups) in the highest supplemented groups.

The results show that the I concentration of liver was at a higher level than meat and also increased significantly with increasing feed I concentrations. I concentrations of liver ranged from 22.4 to 173.4 µg I/kg in KI groups and from 28.1 to 181.3 µg I/kg in $\text{Ca}(\text{IO}_3)_2$ groups.

The transfer into the meat (pectoral) was below $0.17 \pm 0.04\%$. Hence the total I content of meat was low compared with the other tissues investigated. The I concentrations of pectoral and thigh meat were similar and increased significantly with increasing I content of the feed. In KI groups, the I concentrations of pectoral meat ranged from 5.8 µg I/kg for the control to 58.0 µg I/kg for the highest group and from 5.9 µg I/kg to 63.1 µg I/kg respectively in thigh meat. Breast meat of $\text{Ca}(\text{IO}_3)_2$ groups contained between 7.0 µg I/kg in the control and 52.1 µg I/kg in the highest supplemented group, and correspondingly 12.2 µg I/kg to 67.8 µg I/kg in thigh meat.

DISCUSSION

Feed I concentrations

The control feed met the I requirements of the broiler chickens (NRC, 1994). Since an I-free

premix was used this relatively high naturally occurring concentration must have been due to feed ingredients.

Rearing performance

The results regarding rearing performance agree with previously published studies, which found no significant impact on FI, BWG or FCR when similar feed I concentrations were supplied (Perry *et al.*, 1989; Kaufmann and Rambeck, 1998; Maroufyan and Kermanshahi, 2006). These results show that the I concentrations used in the current study met the requirements and did not exceed a critical level for the broilers.

Slaughtering performance

The results of the slaughtering performance are in accordance with previous studies, which investigated the impacts of I supply at concentrations up to 10 mg I/kg and found no significant effect (Kaufmann and Rambeck, 1998; Maroufyan and Kermanshahi, 2006). The effect on thyroid weight observed in present study confirms the results of Travnicek *et al.* (1999) who investigated the influence of dietary I concentrations between 0.3 (control) and 15 mg I/kg on histometric parameters and thyroid gland weight in layers over a 74-d period. They found a decrease in the height of follicular cell-thyreoocytes with increasing feed I content, but no significant impact on

thyroid weight. The present experiment showed that I applications at moderate amounts below EU-regulations did not significantly affect the weight of the thyroid gland. Further studies of the impact of I on thyroid glands of growing chickens, histological investigations should be carried out to identify possible hyperplasia or hypertrophy of the thyroidal follicle cells.

I concentrations

According to previous studies on egg I contents, the I content of the hatching eggs corresponded with concentrations of eggs from laying hens provided with I concentrations of approximately 1.6 mg I/kg (Roettger *et al.*, 2008).

The relatively high I concentrations in the blood serum can be explained by the intense absorption of iodide in the small intestines, which occurs after ingestion and the reduction of free I and iodate into iodide (Lewis, 2004). Subsequently, iodide enters the extracellular fluid which includes blood plasma and interstitial fluid.

From the extracellular compartment, the sodium/iodide-symporter (NIS) actively transports I into the thyroid gland. The concentration gradient between plasma and thyroid can be 1 to 100 or more. The high I concentration measured in thyroid samples is enabled by the active NIS-transport and is necessary for the thyroid function as an I reservoir and endocrine gland for the synthesis of thyroid hormones. The lower carry-over in high supplemented groups is caused by saturation, which occurs above a certain point of provision. There are no data for I concentration in practical broiler diets. Grünewald *et al.* (2006) investigated the actual I concentration of laying hen feed, which meets a mean I concentration of 1.09 mg/kg (min/max: 0.5/2.16 mg I/kg); this corresponds with the I content supplied in group 2 (1 mg I/kg) of the present study. Based on the data from this study, and at typical dietary I inclusions, the I content of a thyroid gland would amount to $492 \pm 127 \mu\text{g}$ with KI supplementation and $562 \pm 395 \mu\text{g}$ with $\text{Ca}(\text{IO}_3)_2$ supplementation. This exceeds the recommended 200-mg daily I intake and agrees with the UL for an adult human suggested by the D-A-CH. However, the risk for an oversupply through a consumption of the thyroid gland remains minimal, because the thyroid is usually removed with oesophagus and trachea during processing.

Thyroid hormones and I enter the liver *via* the *venae portae*. The liver is an important location for deiodination of T_4 . If the outer ring is deiodinated, T_3 is generated, and deiodination of the inner ring rT_3 is produced (Visser *et al.*, 1988). Because of this, a greater accumulation of I was assumed for that tissue

compared with meat. Groppe *et al.* (1991) and Kaufmann and Rambeck (1998) investigated the influence of KI and KIO_3 supplementation on the I content in tissues of male broiler chickens. In liver samples of the 1 mg I/kg supplemented group they found $71.0 \mu\text{g I/kg}$ dry matter, assuming a 70.3% water content for the liver (Souci *et al.*, 2008), which amounts to $21.1 \mu\text{g I/kg}$ fresh matter. In the 0.9 mg I/kg supplemented $\text{Ca}(\text{IO}_3)_2$ group of the present study, approximately double this concentration ($44.1 \mu\text{g I/kg}$) was measured. Furthermore, in 10 mg I/kg-groups, Souci *et al.* (1991) and Kaufmann and Rambeck *et al.*, (1998). found the same or only the 1.5 fold amount of the concentration found in this study at 5 mg supplementation, but this difference partly lay within the range of standard deviation. However, Stibilj and Holcman (2002) detected $31.7 \mu\text{g I/kg}$ with a supplementation of 0.66 mg I/kg laying-hen feed. This agrees well with the data gained in this study; in comparable groups, KI birds showed 31.0 and $\text{Ca}(\text{IO}_3)_2$ birds $28.1 \mu\text{g I/kg}$.

There are very few studies on the I concentration of fowl meat, in response to moderate I supplementation. In studies of I concentration in the tissues of male broiler chickens, Groppe *et al.* (1991) and Kaufmann and Rambeck (1998) found $73 \mu\text{g I/kg}$ dry matter in breast meat in response to a dietary I concentration of 1 mg I/kg. Converted to fresh matter and assuming 70% as water content of the meat (Souci *et al.*, 2008), this would be $22 \mu\text{g I/kg}$ fresh matter, which is in good agreement with the $22.9 \mu\text{g I/kg}$ found in thigh meat of the 1 mg I/kg $\text{Ca}(\text{IO}_3)_2$ group of the present study. The I concentration of meat found in a group given a 10 mg I/kg-feed ($115 \mu\text{g I/kg}$ meat) was approximately double that found in the present study in the 5 mg I/kg supplemented groups (50 to $60 \mu\text{g I/kg}$). At an I dosage of 0.66 mg I/kg laying-hen feed, Stibilj and Holcman (2002) reported an I content of $6.8 \pm 1.2 \mu\text{g/kg}$ muscle tissue. At comparable feed I concentrations, similar meat I concentrations were found in the present study (Tables 6 and 7).

In studies investigating the influence of I supplementation on meat I concentration of other livestock, it was concluded that the impact was rather minor. Franke *et al.* (2008) and Schöne *et al.* (2006) investigated the carry-over of I in pigs; Meyer *et al.* (2008) studied this issue on fattening bulls. At I supplementations between 0.17 and 4.38 mg I/kg feed, Franke *et al.* (2008) measured between 3.9 and $17.1 \mu\text{g I/kg}$ in the muscle/fat fraction, Schöne *et al.* (2006) detected up to $14.6 \mu\text{g I/kg}$ meat at a supplementation of 3 mg I/kg feed and Meyer *et al.* (2008) found a significant influence of feed I concentration on meat I concentration, but these were

still at a rather low level: 0.79 to 8.31 mg I/kg feed produced 16 to 147 µg I/kg in muscle.

The German annual per capita consumption of chicken meat in 2007 was 18 kg (Damme and Möbius, 2009). At the usual feed I dosages of 1 mg I/kg (Grünwald *et al.*, 2006) and a median measured I concentration of 10.6 µg I/kg in broiler meat (thigh and pectoral), this meat would contribute with 0.5 µg I/d to the daily I uptake in consumers. If the feed was enriched with 5 mg I/kg (median concentration of pectoral and thigh meat: 60.2 µg I/kg), the daily contribution would amount to 3.0 µg I/d. In comparison with the recommended daily I intake of 200 µg for adults, it becomes obvious that meat of broiler chickens fed on EU guidelines is not a considerable I source. However, the European guidelines should not be elevated above 10 mg I/kg feed, because the meat I concentration will not be remarkably elevated therewith, but high I doses could have possible adverse effects on broiler health and performance. A reduction of feed UL is not recommended as well because, at current levels, there is no risk for consumers due to the low carry-over into the broiler meat, and furthermore the UL for feed iodisation is not exhausted in practice (Grünwald *et al.*, 2006). It can be positively reviewed that the consumption of chicken meat would not hold the risk for an over-dosage, if thyroid tissue is excluded from comestible production.

CONCLUSION

The study showed that neither of the two I sources at the applied concentrations had a significant impact on growing and slaughtering performance. The investigations show that the carry-over and accumulation into meat is marginal; thus, the contribution to the I supply of consumers is considered as minor. The thyroid gland should be removed at slaughtering to prevent over-consumption. However, the European guidelines on UL for broiler feed should be retained because feed I concentration does not greatly influence the carry-over into meat. An elevation could lead to possible adverse effects on health and performance of the birds, but a reduction is also not necessary because in practice the UL for feed iodisation is not exhausted.

Detailed studies concerning I metabolism and I distribution in different tissues in chicken are still lacking. Therefore more holistic studies with regard to carry-over into eggs, together with meat, blood serum, organs (especially thyroid gland) and the loss *via* excrements, as well as

studies on long term effects of I supply, are necessary.

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