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ORIGINAL CONTRIBUTION

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Influence of dietary iodine on the iodine content of pork and the distribution of the trace element in the body

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Abstract *Background* Millions of people worldwide still suffer from iodine deficiency disorders. Besides salt iodination, iodine is added to animal feed to concentrate it in food of animal origin (milk, eggs, meat). Otherwise possible adverse effects of high supplementation should be avoided. *Aim of the study* The objective of the study was to evaluate the iodine content of pork at various feed iodine concentrations to estimate its contribution to human iodine supply. Furthermore the handling of low and high iodine dosages by the organism should be investigated using the pig as a model for the human. *Methods* Seventy pigs (live weight period 27–115 kg), divided into five groups, were fed diets supplemented with 0 (group 1), 0.5 (group 2), 1 (group 3), 2 (group 4) and 5 (group 5) mg iodine per kg diet. Iodine was determined in the thyroid and in the fractions innards/blood, bones and muscle/fat of four pigs of each group by ICP-MS. *Results* Rising iodine supplementation of feed significantly increased ($P < 0.05$) the iodine content of the muscle/fat fraction [3.9 (group 1), 6.0 (group 2), 8.5 (group 3), 10.8 (group 4) and 17.1 (group 5) μg I/kg]. Carry over (of supplemented iodine) into muscle/fat varied between 0.10 and 0.24%. The highest tested iodine dosage (5 mg I/kg diet) caused a 3.6-fold iodine concentration of the total body (calculated from the contents of the fractions), and a significantly increased thyroid weight compared to the group without supplementary iodine. Iodine supplementation increased iodine content in thyroid and bones significantly ($P < 0.05$) but not in innards/blood. On an average of the groups, the thyroid contained 80% of the body's iodine, innards/blood 14%, muscle/fat 5% and bones 1%. *Conclusions* The iodine content of pork, and consequently its contribution to human iodine supply (~1%), is very low, even at high supplementation of feed. The total body iodine content (empty body) is determined by the iodine intake. Irrespective of the iodine dosage, the thyroid contains about 4/5 of the body iodine. Bones represent a very low iodine concentration, even at a strongly increased iodine intake. The increase of the thyroid weight as an adverse effect of iodine supplementation requires further research with high dietary iodine.

Key words iodine – distribution – muscle/fat – innards/blood – bone – growing pigs

Introduction

Iodine is essential for humans and animals. The trace element is required for synthesis of the thyroid hormones triiodothyronine (T_3) and tetraiodothyronine (T_4) which regulate important metabolic processes like growth, reproduction and development of brain function. Goiter, diminished growth, mental retardation and skeletal deformations are the most apparent consequences of a deficient iodine supply [5, 18]. Iodine deficiency and the resulting disorders represent a global health problem because of their gravity and widespread incidence. Worldwide more than 800 million people still suffer from iodine deficiency [9]. Besides salt iodination, efforts are underway to increase the iodine content of food of animal origin (milk, eggs, meat) by supplementation of feeding stuffs in recent years. But iodine prophylaxis also includes a high risk of overdosing [2] because the margin between the requirement of 180–200 μg/days [4] and the upper limit (UL) of 500 μg/days [4] for adult people is narrow (~1:3). Especially in regions with previous iodine deficiency supplementation programs may lead to a transient higher incidence of iodine-induced hyperthyroidism [1, 7, 29]. The latest survey concerning the iodine supply in Germany [30], and studies recording a high iodine transfer into milk and eggs [10, 34], indicate that the problem of excessive iodine intake may come to the fore in future. The determined mean urine iodine excretion in the survey [30] shows that, following WHO references [31], Germany can no longer be considered an iodine deficient area. Since iodine content is not declared in food of animal origin, excessive iodine intake caused by such products must be avoided by controlling the maximum iodine levels in animal feed. In 2005, the European Food Safety Authority (EFSA) evaluated the benefits and risks of iodine supplementation in animal feed and established that more dose-response studies are needed to achieve secure information about the transfer of iodine from feed into food of animal origin. Furthermore, studies checking the requirements of farm animals in consideration of the higher performances are rare [8].

The carry over of iodine from feed into milk and eggs seems to be remarkable [10, 17, 26, 34]. Trials for testing iodine enrichment in pork by supplementation of feed [16, 24] showed lower transfer rates but also indicated meat iodine contents which promised to improve human iodine supply. In contrast, results of studies received by ICP-MS analysis [27, 28] indicated lower meat iodine concentrations. In the present pig experiment, the concentration of the trace element in the fraction muscle/fat should be investigated at varying feed iodine supplementation levels ranging in magnitude from the requirements up to very high levels. Furthermore, the objective of the study was to receive information about the handling of lower and higher amounts of iodine through the organism using the pig as a model for human being.

Materials and methods

Animals and diets

The experiment was carried out with 70 BHZP-hybrids (females:male-castrated in the relation of 1:1). In five groups of 14 animals each, iodine dosages of 0.5; 1; 2 and 5 mg/kg diet were tested in comparison to a control group without supplementary iodine. Iodine was added in the form of calcium iodate (anhydride). The trial covered the live weight period from 27 to 115 kg. Depending on the individual weight gain the pigs received iodine supplementation for 97–125 days. All pigs were housed in individual pens. The pigs were fed twice a day with individual rations of a grain soya-bean meal diet in accordance with a daily weight gain of >800 g. Feed composition was adapted to the changing demand with increasing weight in two phases. The composition of the basic diet in the phases P-I (27–70 kg weight) and P-II (70–115 kg weight) is shown in Table 1. The diets provided all nutrients at or above the references for pigs [12]. The amount of iodine addition did not vary between the two phases of feeding. Water was provided ad libitum.

Investigation criteria and sampling

Over the whole trial period feed intake was recorded continuously and body weight (BW) was measured once a week to calculate average daily weight gain (ADG) and feed conversion ratio (FCR).

In each group, four pigs (two females, two male-castrated) were slaughtered at the end of the study. After the removal of the thyroid, the rest of the body (except bristles, hoofs and content of the gastrointestinal tract, urinary bladder and gall bladder) was sampled into the fractions innards/blood, bones and muscle/fat. The fraction innards/blood consisted of the emptied gastrointestinal tract, liver and emptied gall bladder, kidney with leaf fat, emptied bladder, lung and heart mixed up with the collected blood at slaughter. The carcass and all fractions were weighed. The weight of the empty body was calculated from the weights of carcass, innards and blood. The fractions yielded were homogenized and representative samples were lyophilized. The fragmentation of the body allowed the estimation of the iodine content of the extrathyreoidal domain and almost the whole pig

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Table 1 Composition of diet in P-I and P-II

	P-I 27–70 kg weight	P-II 70–115 kg weight
Barley (g/kg)	300.0	350.0
Wheat (g/kg)	348.5	348.5
Maize (g/kg)	50.0	50.0
Soy bean meal (g/kg)	220.0	170.0
Soy bean oil (g/kg)	40.0	40.0
Vitamins/mineral nutrients[a] (g/kg)	30.0	30.0
L-lysine monohydrochloride (g/kg)	5.5	5.5
DL-methionine (g/kg)	3.0	3.0
L-threonine (g/kg)	2.5	2.5
L-tryptophane (g/kg)	0.5	0.5
ME[b] (MJ/kg)	14.6	14.6
Lysine[c] (g/kg)	13.5	12.3
Crude protein	20.5	19.4

[a]Content per kg premix: 250 g Ca, 60 g P, 55 g Na, 10 g Mg, 400,000 IU vitamin A, 40,000 IU vitamin D3, 1,200 mg vitamin E, 37.5 mg vitamin B1, 100 mg vitamin B2, 100 mg vitamin B6, 750 µg vitamin B12, 52.5 mg vitamin K3, 500 mg nicotinic acid, 337.5 mg Ca-pantothenate, 5,000 mg cholinchloride, 4,000 mg Fe, 1,000 mg Cu, 2,000 mg Mn, 4,000 mg Zn, 16 mg Se, 20 mg Co
[b]*ME* metabolizable energy
[c]Calulated from components of diet

(empty body). The iodine concentration of the extrathyreoidal domain was calculated from the concentrations and the weights of the fractions innards/blood, muscle/fat and bones. The empty body additionally included the thyroid. To calculate the carry over of iodine from feed into the muscle/fat fraction, the amount of iodine, stored in this fraction over the trial period was established by subtracting an assessed initial iodine amount in muscle/fat from the amount at the end of the trial. The initial amount of iodine in muscle/fat was estimated from the investigated iodine concentration of the fraction at a feed iodine dosage of 1 mg/kg diet, which nearly corresponds to the iodine content of pig feed in practice [14], and the initial weight of the muscle/fat fraction. The initial weight was evaluated from the proportion of the fraction weight to the total BW postulating a similar proportion at the beginning and at the end of the trial. Finally the carry over was calculated as the percentage of the iodine amount, stored in muscle/fat during the fattening period, of the calculated intake of supplemented iodine.

Iodine determination

Iodine content was analyzed by inductively coupled plasma-mass spectrometry (ICP-MS SCIEX ELAN® DRC-e, Perkin Elmer) after alkaline digestion. An ammonia solution was used for disintegration of the feed samples [11]. The lyophilized body samples were treated with Tetramethylammonium hydroxide (TMAH, Appli Chem, Darmstadt, Germany). One milliliter TMAH and 5 ml distilled deionized water were added to 50 mg finely ground, lyophilized thyroid or to 500 mg finely ground, lyophilized sample of the other fractions. Iodine was extracted from the samples in gastight vessels for 3 h at 90°C. After cooling to room temperature and the addition of 14 ml of distilled deionized water, samples were centrifuged for 15 min at 4,000 rpm. 0.5 ml of the supernatant was mixed with 4.5 ml distilled deionized water and 1 ml tellurium dilution was added as internal standard (200 µg/l, Merck, Darmstadt, Germany). For determination of iodine concentration, the method of standard addition calibration was applied. Four vessels with 0.5 ml sample each were spiked with three different amounts of iodine as KI (Merck, Darmstadt, Germany) according to the expected range of the sample's iodine content, and all vessels were filled up to 5 ml with distilled deionized water. The fourth, not spiked, sample was measured after the highest calibration sample, and acted as a signal for the software to start the analysis. Control of results was carried out by the certified standard BCR 185 R (Community Bureau of Reference, Brussels, Belgium).

Statistics

The data are presented in terms of mean and standard deviation. Statistical analysis was performed using SPSS 13.0 (SPSS Inc., Chicago, IL, USA). Analysis of variance (ANOVA) was used to test the influence of iodine supplementation on the performance parameters and the iodine content of the body samples. In the case of significance ($P < 0.05$) the Student–Newman–Keuls procedure was used to compare the distinctive group means.

Results and discussion

The analyzed iodine contents of the feed agreed satisfactorily with the added quantities of the trace element (Table 2). The iodine content of the feed in the group without supplementation nearly met the requirements of growing pigs [12, 21].

On average of the groups, ADG amounted to 837 ± 83 g/days at an average daily feed intake (ADFI) of 2.24 ± 0.11 kg/days resulting in a FCR of 2.70 ± 0.23 kg/kg. The various iodine dosages had no significant effect ($P < 0.05$) on the performance parameters (Table 3) or on the empty BW (Table 4).

A significant effect ($P < 0.05$) of the iodine supplementation was shown on the thyroid weight (Table 4). The organ weight increase caused by the highest tested iodine addition (5 mg/kg diet) amounted to 54% in comparison to the control. The goitrogenic effect of a chronically high iodine intake

Table 2 Results of iodine analysis in the diets and derived means for calculation of iodine intake (mg/kg)

Iodine supplementation	0	0.5	1.0	2.0	5.0
P-I	0.25	0.31	1.24	1.98	4.19
P-II	0.09	0.51	0.73	2.43	4.56
Mean	0.17	0.41	0.99	2.20	4.38

was already observed in former studies with pigs, bulls and also humans [20, 22, 23, 25, 35]. Thus an enlargement of the thyroid is not only a sign of iodine deficiency [5, 18], but also can be an indication for excessive iodine exposure [19, 22, 23, 25, 35]. Studies show that in a healthy organism, excessive iodine intake leads to acute inhibition of organification of iodine (Wolff–Chaikoff effect), followed by a so-called "escape" phenomenon [19, 33]. Thereby normal thyroid hormone synthesis returns and iodine uptake into the thyroid is decreased by down-regulation of sodium iodide symporter (NIS). The absence of the escape from the acute Wolff–Chaikoff effect and the enhancement of thyroid autoimmunity or both together are discussed as reasons for the iodine induced goiter and hypothyroidism. Therby the role of the individual susceptibility also remains unclear [6, 19, 29, 35]. An unchanged performance and the absence of an enlargement of the thyroid in the group without iodine supplementation shows that the existing references [12, 21] for growing pigs are also sufficient at higher performances.

The thyroid represented by far the highest iodine concentration of the investigated fractions of the pig body (Table 5). Rising iodine dosages up to 2 mg/kg diet increased the thyroid iodine content significantly ($P < 0.05$). A still higher iodine supplementation of 5 mg/kg diet did not increase it further, which indicates that a plateau of thyroid iodine concentration is reached at high iodine intake. The highest iodine application caused a 2.7-fold iodine concentration of the thyroid compared to the control group. The total amount of iodine in the thyroid also rose due to the described weight increase. Since iodine uptake into the thyroid seems not to be diminished, the main reason for the enlargement of the thyroid in growing pigs at iodine dosages between 2.2 and 4.4 mg/kg diet may be the inhibition of the thyroid hormone formation [19].

Compared to the thyroid, a very small amount of iodine is stored in the extrathyreoidal fractions (Table 5). The innards/blood as the fraction with the highest extrathyreoidal iodine content, represented less than 0.1 per mille of the iodine concentration of the thyroid. Differences in the iodine concentration among the groups of the innards/blood showed no significance ($P > 0.05$).

The muscle/fat tissue showed the lowest iodine concentration of the investigated fractions (Table 5). Rising iodine supplementation of feed increased the iodine content significantly ($P < 0.05$) up to 17.1 µg/kg at the highest iodine dosage. The calculated carry over of supplemented iodine into the muscle/fat fraction varied between 0.10 and 0.24% and tended to decrease with the rising iodine content of feed (Table 6). Compared to the carry over into milk and eggs [10, 26, 34] the transfer into pork is marginal. In general, former pig experiments using the Sandell and Kolthoff method for iodine detection [16, 24] provided higher values than the present study (Table 5).

Table 3 Comparison of performance parameters among the experimental groups (mean ± SD, $n = 14$)

Iodine supplementation	0	0.5	1.0	2.0	5.0
Initial BW (kg)	27.2 ± 1.9	27.4 ± 1.9	26.5 ± 1.6	27.0 ± 1.9	27.1 ± 1.7
Final BW (kg)	119.0 ± 3.5	116.5 ± 6.1	116.5 ± 3.2	119.4 ± 5.0	118.7 ± 2.8
ADG (g/d)	837 ± 70	819 ± 99	811 ± 93	851 ± 84	867 ± 63
ADFI (kg/d)	2.26 ± 0.10	2.24 ± 0.13	2.21 ± 0.10	2.24 ± 0.13	2.26 ± 0.10
FCR (kg/kg)	2.72 ± 0.21	2.77 ± 0.29	2.74 ± 0.28	2.65 ± 0.18	2.62 ± 0.16

BW body weight, *ADFI* average daily feed intake, *ADG* daily weight gain, *FCR* feed conversion ratio

Table 4 Weights of the investigated fractions and the empty body (mean ± SD, $n = 4$)

	Iodine supplementation in mg/kg diet				
	0	0.5	1.0	2.0	5.0
Thyroid (g)	8.2 ± 1.5^{a}	10.1 ± 2.3^{ab}	9.8 ± 2.1^{ab}	8.3 ± 1.8^{a}	12.9 ± 2.3^{b}
Innards/blood (kg)	17.0 ± 0.5	16.3 ± 1.2	16.7 ± 1.0	16.4 ± 0.8	17.0 ± 1.0
Bones (kg)	9.7 ± 0.6	10.0 ± 0.3	9.1 ± 0.6	10.0 ± 0.4	9.8 ± 0.5
Muscle/fat (kg)	85.9 ± 2.7	83.0 ± 4.1	81.8 ± 4.1	86.3 ± 3.1	84.9 ± 2.8
Empty body[1] (kg)	115.3 ± 3.7	111.9 ± 5.4	109.9 ± 4.6	114.9 ± 3.3	114.0 ± 2.9

[1]The empty body represents the total slaughtered animal without bristles and hoofs and the content of the gastrointestinal tract, urinary bladder and gall bladder

abcd values with various superscripts in a row differ significantly ($P < 0.05$)

Table 5 Iodine concentration in the thyroid and in the body fractions innards/blood, bones and muscle/fat (mean ± SD, $n = 4$)

	Iodine supplementation in mg/kg diet				
	0	0.5	1.0	2.0	5.0
Thyroid (μg/g)	620 ± 71^{c}	1054 ± 280^{b}	1154 ± 191^{b}	1699 ± 184^{a}	1645 ± 159^{a}
Innards/blood (μg/kg)	94 ± 61	63 ± 40	138 ± 73	230 ± 145	126 ± 38
Bones (μg/kg)	15 ± 5^{b}	19 ± 10^{b}	17 ± 4^{b}	18 ± 2^{b}	37 ± 4^{a}
Muscle/fat (μg/kg)	3.9 ± 0.6^{c}	6.0 ± 1.9^{c}	8.5 ± 1.9^{b}	10.8 ± 1.2^{b}	17.1 ± 1.5^{a}
Extrathyreoidal (μg/kg)	19 ± 9^{b}	16 ± 6^{b}	30 ± 12^{ab}	43 ± 19^{a}	35 ± 6^{ab}
Empty body[1] (μg/kg)	63 ± 6^{d}	112 ± 27^{c}	137 ± 40^{bc}	168 ± 15^{b}	226 ± 48^{a}

[1]The empty body represents the total slaughtered animal without bristles and hoofs and the content of the gastrointestinal tract, urinary bladder and gall bladder
abcd values with various superscripts in a row differ significantly ($P < 0.05$)

Applying a feed iodine dosage of 5 mg/kg, He et al. [16] found an iodine concentration in muscle of 38.5 μg/kg and in adipose tissue of 33.2 μg/kg. Without adding iodine to the feed, He et al. [16] and Rambeck et al. [24], investigated muscle iodine contents of 32 and 23 μg/kg, respectively. Studies utilizing ICP-MS for iodine analysis [27, 28] show similarly low meat iodine contents as in the present study (Table 5).

The present study shows that in Germany (with an average pork and pork product consumption of 38.9 kg per head and year [3]), pork and its products, produced with 5 mg I/kg diet supplementation and without the application of iodized salt, could feature an iodine intake of just 1.8 μg/days per person. With regard to the references of the German Nutrition Society (DGE), pork and its products could just contribute about 1% to the recommended iodine consumption [4] at the tested feed iodine dosages. In Germany, feeding stuffs currently used for pigs contain an average of about 1.51 mg I/kg diet [14]. Iodine enrichment of meat can be achieved more effectively by the use of iodized salt in the manufacture of pork products [32].

The iodine content in the bones was slightly higher than in the muscle/fat fraction (Table 5) and was elevated significantly only at an iodine dosage of 5 mg/kg diet ($P < 0.05$). In accordance with existing studies with goats and pigs [13, 28], the present results show that excessive absorbed iodine is stored in bones to a minor degree.

Table 6 Carry over of supplemented iodine from feed into muscle/fat of growing pigs (mean ± SD, $n = 4$)

Iodine supplementation (mg/kg)	Carry over[a] (%)
0	–
0.5	0.24 ± 0.12
1	0.21 ± 0.06
2	0.15 ± 0.02
5	0.10 ± 0.01

[a]Percentage of supplemented iodine intake

Calculated from the iodine contents of the fractions, the overall concentrations of the extrathyreoidal domain and the empty body were significantly ($P < 0.05$) increased by the iodine supplementation. Comparing the highest tested iodine supplementation (5 mg/kg) with the unsupplemented group, the extrathyreoidal iodine concentration was nearly doubled, and the content of the pig body (empty body) was 3.6 times higher (Table 5).

Looking at the iodine amounts, a human study estimated a total body iodine content of 14.6 mg at an iodine intake that conforms to the threefold requirement [15]. The present trial investigated at an adequate iodine dosage (0.5 mg/kg diet) a total iodine content of the same magnitude (12.3 mg).

With regard to the distribution of iodine in the organism in the present study, the thyroid contained, on average of the groups, 80% of the total body iodine (empty body), innards/blood 14%, muscle/fat 5% and the bones 1%. The distribution of iodine in the body remained widely unaffected by the varying iodine dosages (Fig. 1). While the human study [15] recorded that in the overall extrathyreoidal domain

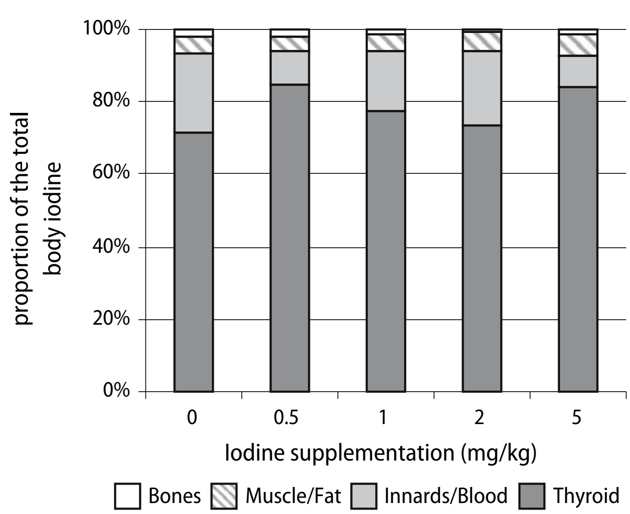

Fig. 1 Iodine distribution in the pig body at various iodine supplementation of feed

nearly 30% of the total body iodine is stored, the proportion of extrathyreoidal iodine in the present trial ranged between 14.9 and 28.7%.

Conclusion

Iodine dosages of up to 5 mg/kg feed allow no effective iodine enrichment of pork. Compared to the transfer rates of iodine into eggs and milk, the carry over into muscle and fat of pigs is inconsiderable. The contribution of pork to daily human iodine demand is marginal (about 1%) even at a high iodine supplementation of feed (5 mg/kg). Iodine contents analyzed by ICP-MS seem to be continuously lower than values obtained with older methods. Since nutrition tables are still based on the results of older studies, data certainly require revision. The total body iodine concentration is determined by the iodine intake. Considering the iodine distribution, the thyroid contains, irrespective of the iodine dosage, about 4/5 of the empty body iodine. Bones represent a low iodine concentration, even at a strongly increased iodine intake. The references of the scientific committees [12, 21] concerning the iodine requirements of growing pigs are also adequate for pigs with higher performances.

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