Influence of various iodine supplementation levels and two different iodine species on the iodine content of the milk of cows fed rapeseed meal or distillers dried grains with solubles as the protein source

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ABSTRACT

Supplementation of animal feed with iodine influences the iodine content of milk and therefore, in addition to salt iodination, provides another possibility for improving the human iodine supply. On the other hand, excessive iodine intake by humans through drinking milk must be avoided. Furthermore, the iodine content of milk varies, depending on the presence of iodine antagonists in feed (e.g., glucosinolates in rapeseed) and the applied iodine species. This study evaluated the impact of various feed iodine supplementation levels up to the permitted maximum level, the effect of applying rapeseed compared with a glucosinolate-free ration, and the impact of 2 different iodine species on the iodine content of milk. A total of 32 dairy cows were divided into 4 groups with 8 animals each. Two groups received distillers dried grains with solubles (DDGS) as the protein source, and the others received rapeseed meal (RSM, 16.5% of total diet). In each case, half the animals received feed supplemented with iodine in the form of potassium iodide, and the other half received feed supplemented with iodine in the form of calcium iodate. Iodine supplementation levels of 0, 0.5, 1, 2, 3, 4, and 5 mg/kg of dry matter (DM) were tested in consecutive periods of 21 d each. The milk iodine concentration increased with increasing iodine supplementation of the feed. Rapeseed meal in the ration (0.58 mmol of glucosinolates/kg of diet DM) diminished the milk iodine concentration by up to one-half to one-third of the concentration achieved by DDGS. At iodine supplementation levels of 2 mg/kg of DM and higher, the differences were significant. The application of iodate predominantly resulted in higher milk iodine concentrations compared with iodide, but not significantly in any period. At the highest tested iodine supplementa-

tion (5 mg/kg of DM), the milk iodine concentration increased up to 1,464 (iodide) and 1,578 µg/kg (iodate) when feeding DDGS and up to 718 (iodide) and 620 µg/kg (iodate) in the RSM groups. The carry over of iodine from feed into milk amounted to 30 to 56% when using DDGS, and 11 to 25% when using RSM. The maximum level of iodine currently allowed in the feed of dairy cows in Europe could lead to high milk iodine concentrations. As a result, the Tolerable Upper Intake Level in human nutrition could be exceeded. Therefore, this maximum level needs to be reevaluated. In addition to iodine supplementation, the application of RSM in the ration must be considered when estimating the iodine content of milk.

Key words: dairy cow, iodine, milk, rapeseed meal

INTRODUCTION

Worldwide, 13% of the population still suffers from iodine deficiency. In addition to salt iodination, the iodine supplementation of animal feed can contribute to improving the human iodine supply because iodine is one of the rare trace elements whose concentration in foods of animal origin (milk, eggs) can be altered considerably by its content in animal feed (Kaufmann and Rambeck, 1998; Flachowsky et al., 2006; Schöne et al., 2009). On the other hand, iodine in human nutrition is also characterized by a high risk of overdosing (ERNA and EHPM, 2004) because the margin between the requirement of 150 to 200 µg/d (D-A-CH, 2000; WHO, UNICEF, and ICCIDD, 2001) and the Tolerable Upper Intake Level (UL) of 500 µg/d (D-A-CH, 2000) up to 1 mg/d (WHO, 1994) is narrow for adult humans. Declaration of the iodine content of foods of animal origin seems impossible. Therefore, the enrichment of iodine in the milk of consumers should be limited by setting low maximum levels for iodine in animal feed that are still practical. In 2005, the European Food Safety Authority (EFSA) evaluated the benefits and risks of various levels of iodine supplementation in animal feed and concluded that data concerning the

Received January 12, 2009.

Accepted May 5, 2009.

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transfer of iodine from feed into foods of animal origin were insufficient (EFSA, 2005). In this regard, milk is of particular importance because, in contrast to meat (Franke et al., 2008; Meyer et al., 2008), it is characterized by a high carry over of iodine from feed into foods of animal origin (Flachowsky et al., 2006) and may therefore lead to high iodine concentrations in milk (Hillman and Curtis, 1980; Rysava et al., 2007; Schöne et al., 2009). In addition to iodine accumulating in the thyroid and other extrathyroidal tissues (salivary glands, gastric mucosa, choroid plexus, and ciliary body of the eye), it is actively accumulated in the lactating mammary gland by the sodium iodide symporter (NIS; Cavaliere, 1997; Laurberg et al., 2002). Because of the enhanced application of iodine in feedstuffs, the iodine content of milk for consumers has increased in the last 10 yr (Launer and Richter, 2005; Jahreis et al., 2007). At present, the contribution of milk and milk products to the human iodine intake in Germany is estimated at 40% (Jahreis et al., 2007). Furthermore, milk is an alternative indicator to urine for the iodine supply of the animal because of its good and fast reflectance of iodine intake (Binnerts, 1989; Herzig et al., 1996).

In addition to the primary impact of iodine in the feed on the iodine concentration of milk, there are further influencing factors, such as goitrogens in the feed, the cow breed, the iodine species applied, the stage of lactation of the cow, the use of iodized udder disinfecting solutions, the use of iodine-containing drugs, and possibly the milk yield of the cow (Iwarsson, 1973; Hemken, 1979; Papas et al., 1979; Franke et al., 1983; Swanson et al., 1990; Flachowsky et al., 2007). Because of the expected increase in by-product availability from bio-energy manufacture, the use of rapeseed meal (RSM) and rapeseed press cake in animal feed is gaining importance. The degradation product thiocyanate of the contained glucosinolates (GSL) competitively inhibits iodine uptake by the NIS into the thyroid (Brown-Grant, 1957) and also seems to reduce the transfer of iodine into milk (Papas et al., 1979; Laurberg et al., 2002; Schöne et al., 2006). In the European Union, sodium iodide, potassium iodide, calcium iodate hexahydrate, and anhydrous calcium iodate are approved for the iodination of feedstuffs (European Union, 2005). Because iodine is characterized by instability in the presence of oxygen, storage losses may also result in lower iodine contents in milk.

In the present cow study, milk iodine concentrations at various feed iodine supplementation levels up to the permitted maximum level in feed (5 mg/kg; European Union, 2005) were investigated. Furthermore, the impact of a high percentage of RSM in the ration and of the applied iodine species (iodide, iodate) on the iodine content of milk was evaluated.

MATERIALS AND METHODS

Experimental Design and Conditions

The trial was carried out at the experiment station of the Institute of Animal Nutrition (FLI), Braunschweig, Germany. Thirty-two dairy cows of the German Holstein breed in the first stage of lactation were divided equally into 4 groups of 8 animals each, considering the milk yield, age, and days in lactation. At the beginning of the trial, on average the cows were 48 ± 25 DIM and featured an average number of lactations of 2.1 ± 1.2 , a BW of 582 ± 74 kg, and a daily milk yield of 31.3 ± 6.3 kg. The cows were fed a TMR that was formulated to meet the nutritional requirements of dairy cows stated by the German Society of Nutrition Physiology (GfE, 2001) and which, on a DM basis, consisted of 50% concentrate, 25% corn (*Zea mays*) silage, and 25% grass silage. The precise composition is presented in Table 1. Feed and water were provided ad libitum. In 2 groups, RSM containing $3.5 \mu\text{mol}$ of GSL/g of DM was applied as protein source (16.5% of TMR); in the other 2 groups, the cows received distillers dried grains with solubles (DDGS) made from 90% wheat and 10% barley. One group from each protein source received iodine in the form of potassium iodide (KI), whereas the other group received iodine in the form of anhydrous calcium iodate [$\text{Ca}(\text{IO}_3)_2$]. Below, the groups are mentioned in the way they are shown in Table 1. In 7 periods of 21 d each, iodine supplementation levels of 0, 0.5, 1, 2, 3, 4, and 5 mg/kg of DM were tested (equally in all groups).

Iodine addition was realized by making a premix of ground wheat and the appropriate iodine amount for each iodine species and supplementation level. The adequate amount of wheat in the concentrate was exchanged for this premix. The feed was mixed a few days before the beginning of each period. The feed intake was recorded continuously for each cow by self-feeding stations (Type RIC, Insentec B.V., Marknesse, the Netherlands) and ear transponders. According to their feeding groups, the cows were housed in group pens that were equipped with a slatted floor and cubicles covered with rubber mats and straw dust. Milking took place twice a day at 0530 and 1530 h, at which time milk yield was recorded with automatic milk counters. Teats were dipped with an iodine-free teat dip (Nattidine, Hypred GmbH, Bornheim-Sechtern, Germany) after each milking.

Sample Collection, Preparation, and Analysis

Representative concentrate samples were taken once a week, and silage samples were taken twice a week. Before analysis, the feed samples were dried, ground,

Table 1. Composition of the concentrates and the TMR in the experimental groups¹

Item	DDGS/iodide	DDGS/iodate	RSM/iodide	RSM/iodate
Calculated composition of concentrate				
DDGS (%)	33.0	33.0	—	—
RSM (%)	—	—	33.0	33.0
Wheat (%)	25.0	25.0	25.0	25.0
Corn (%)	25.0	25.0	25.0	25.0
Dried sugar beet pulp (%)	13.5	13.5	13.5	13.5
Calcium carbonate (%)	0.5	0.5	0.5	0.5
Soybean oil (%)	1.0	1.0	1.0	1.0
Mineral feed ² (%)	2.0	2.0	2.0	2.0
Analyzed composition of TMR				
DM (g/kg)	618	618	611	612
Crude ash (g/kg of DM)	63	63	67	67
CP (g/kg of DM)	164	164	162	162
Crude fat (g/kg of DM)	42	42	37	38
Crude fiber (g/kg of DM)	150	150	159	160
ADF (g/kg of DM)	179	181	187	186
NDF (g/kg of DM)	353	358	353	355
ME (MJ/kg of DM)	11.5	11.5	11.3	11.3
NE _L (MJ/kg of DM)	7.03	7.03	6.92	6.92
Glucosinolate content (mmol/kg of DM)	ND ³	ND	0.58	0.58

¹DDGS = distillers dried grains with solubles; RSM = rapeseed meal.

²Without iodine supplementation, per kilogram of mineral feed: 140 g of Ca, 120 g of Na, 70 g of P, 40 g of Mg, 6 g of Zn; 5.4 g of Mn; 1 g of Cu; 40 mg of Se; 25 mg of Co; 1,000,000 IU of vitamin A; 100,000 IU of vitamin D₃; 1,500 mg of vitamin E.

³Not detected.

and homogenized. For disintegration, the silages were spiked with a 1% ammonia solution at a ratio of 1:100 and the concentrates were spiked at a ratio of 1:200 up to 1:1,000, depending on the expected iodine content (as in Franke et al., 2001). The dilutions were boiled for 30 min and filtered afterward. After addition of 0.5 mL of tellurium (100 µg/L, Alfa Aesar, Johnson Matthey GmbH, Karlsruhe, Germany) as the internal standard to 10 mL of filtrate, the samples were analyzed by inductively coupled plasma-mass spectrometry (model 7500c, Agilent, Waldbronn, Germany). The GSL content of the RSM was analyzed by HPLC according to the international standard DIN EN ISO (1995). Milk samples were taken on d 1, 4, 11, 19, 20, and 21 of each period to evaluate the achievement of a steady state after the change in iodine supply. Samples were stored at −20°C. Before analysis, the milk samples were heated to 50°C and homogenized by UltraTurrax (Type T25, Ika Werke GmbH, Staufen, Germany). The appropriate morning and evening milks were mixed according to the milk yields. The milk was diluted (1:10 or 1:25, depending on feed iodine supplementation) with a solution of tetramethylammonium hydroxide (0.07 mol/L, Alfa Aesar GmbH Co. KG, Karlsruhe, Germany) according to the method of Fecher et al. (1998). After addition of 0.5 mL of tellurium as an internal standard to 10 mL of the dilution, the samples were analyzed by inductively coupled plasma-mass spectrometry according to the method of Sturup and Buchert (1996).

Calculations and Statistics

The iodine concentration of the TMR was established from the analyzed concentrations in the single feed components [concentrate, corn silage, grass silage] with regard to the proportion of the component of the TMR. In each period, the day was identified on which a steady state of milk iodine concentration was reached (breakpoint, BP), using the following model:

$$\begin{aligned} \text{Milk iodine} = & (a + b \times \text{day}) \times (\text{day} \leq \text{BP}) \\ & + (a + b \times \text{BP}) \times (\text{day} > \text{BP}), \end{aligned}$$

where *a* and *b* are regression parameters: *a* = intercept with the ordinate and *b* = slope of first part of function. The established breakpoints showed that at sampling d 19, 20, and 21, a steady state of milk iodine concentration was definitely reached, independently of the period. Therefore, only the values of those days were used for calculations and statistical evaluation.

The carry over represents the total amount of iodine in the milk per day (milk yield multiplied by milk iodine concentration) related to the respective daily iodine intake (feed intake multiplied by feed iodine concentration), given as a percentage. Milk iodine reduction per millimole of ingested GSL was calculated by dividing the differences in milk iodine concentration between DDGS and RSM by the mean daily GSL intake.

Table 2. Analyzed iodine concentrations of the TMR and daily iodine intakes (means $\pm$ SD, $n = 8$) in the experimental groups¹ at the various iodine supplementation levels

Iodine supplementation	DDGS/iodide	DDGS/iodate	RSM/iodide	RSM/iodate
Iodine concentration of TMR (mg/kg of DM)				
0	0.18	0.18	0.16	0.16
0.5	0.61	0.71	0.70	0.61
1	1.37	1.37	1.38	1.38
2	1.98	1.84	1.92	1.97
3	2.69	2.98	2.65	3.00
4	3.93	3.58	3.37	3.92
5	4.81	4.78	5.17	5.38
Iodine intake (mg/d)				
0	3.7 $\pm$ 0.5	3.9 $\pm$ 0.5	3.7 $\pm$ 0.4	3.2 $\pm$ 0.4
0.5	12.1 $\pm$ 1.7	13.6 $\pm$ 2.5	15.9 $\pm$ 1.7	12.2 $\pm$ 0.9
1	26.3 $\pm$ 3.5	25.8 $\pm$ 3.5	26.6 $\pm$ 5.2	26.3 $\pm$ 3.6
2	40.9 $\pm$ 4.2	38.5 $\pm$ 4.3	43.4 $\pm$ 4.8	43.0 $\pm$ 3.5
3	55.4 $\pm$ 7.6	63.5 $\pm$ 7.5	62.5 $\pm$ 5.4	62.7 $\pm$ 4.7
4	84.8 $\pm$ 9.6	81.6 $\pm$ 8.6	79.9 $\pm$ 7.7	87.4 $\pm$ 6.1
5	102.9 $\pm$ 3.5	108.7 $\pm$ 9.6	119.7 $\pm$ 0.4	119.1 $\pm$ 8.9

¹DDGS = distillers dried grains with solubles; RSM = rapeseed meal.

Statistical analysis was performed with the SAS software package (version 9.1, SAS Institute, Cary, NC). The individual cow was the experimental unit. The normal distribution was tested by the Kolmogorov-Smirnov test. The tested variables milk iodine concentration, milk iodine amount, and carry over were subjected to ANOVA by applying the GLM procedure with a 3-factorial design. Fixed effects were supplementation (0, 0.5, 1, 2, 3, 4, and 5 mg of iodine/kg of DM), protein source (DDGS, RSM), and iodine species (iodide, iodate). As a covariable, the lactation number (first lactation or heifer = 1; 2 or more lactations = 2) was taken into account for all 3 variables for milk iodine concentration auxiliary to the milk yield. The Tukey-Kramer procedure was used for multiple comparisons of the least squares means of the experimental groups. Least squares means and standard errors of the mean of the milk iodine concentration and milk iodine amount are reported. Differences were considered significant at $P < 0.05$.

RESULTS AND DISCUSSION

Iodine and GSL Content of Feed

The analyzed iodine concentrations of the TMR corresponded closely with the intended feed iodine supplementation levels (Table 2). The iodine concentrations in the period without supplementation were below the recommendations of 0.5 mg/kg of DM for dairy cows (GfE, 2001). Iodine concentrations of the concentrates in the groups with iodide were not consistently lower than in the groups with iodate. Hence, the analyzed iodine contents of the feed showed that no larger losses of iodine occurred when adding potassium iodide com-

pared with calcium iodate at the storage time of 3 to 4 wk. In table salt, iodine losses of approximately 30% were investigated at 60% relative air humidity and unlimited air access after 30 d of storage (Waszkowiak and Szymandera-Buska, 2007), whereas when light, humidity, and high temperatures were excluded, most of the potassium iodide added was conserved during storage for several months (Voudouris, 1975).

Consequently, in the present study possible differences in the tested variables attributable to iodide and iodate application could not be ascribed to iodine losses in feed but could be traced back to differences in metabolism. The RSM used showed a low GSL concentration of 3.5 mmol/kg of DM. The resulting GSL content of the TMR of 0.58 mmol/kg of DM (Table 1) led to mean daily GSL intakes of 11.0 to 13.7 mmol.

Performance

The cows had an average milk yield of 34.2 ± 5.7 kg/d at a mean DMI of 21.3 ± 2.5 kg/d and a mean BW of 633 ± 70 kg. The cows in the different experimental groups exhibited similar performance (K. Franke, U. Meyer, and G. Flachowsky; unpublished data).

Milk Iodine Concentration

Milk iodine concentration adapted to the changed iodine supply within a few days (breakpoints between d 3 and 11). Consequently, the milk iodine concentrations on d 19, 20, and 21 of each period were independent of the iodine supplementation of the previous period. Figure 1 shows the course of the milk iodine concentration in the periods with increasing iodine supplementation of feed.

Table 3. Iodine concentration of milk (LSM $\pm$ SEM of values from d 19, 20, and 21, $n = 24$)¹

Iodine supplementation (mg/kg of DM)	DDGS/iodide ($\mu\text{g/kg}$)	DDGS/iodate ($\mu\text{g/kg}$)	RSM/iodide ($\mu\text{g/kg}$)	RSM/iodate ($\mu\text{g/kg}$)
0	83 ^D $\pm$ 5	72 ^D $\pm$ 3	18 ^C $\pm$ 2	27 ^C $\pm$ 2
0.5	158 ^D $\pm$ 8	188 ^D $\pm$ 9	51 ^{AB} $\pm$ 4	59 ^C $\pm$ 5
1	214 ^D $\pm$ 12	231 ^D $\pm$ 14	82 ^{AB} $\pm$ 7	73 ^C $\pm$ 6
2	550 ^{a,C} $\pm$ 28	584 ^{a,C} $\pm$ 30	186 ^{b,AB} $\pm$ 11	234 ^{b,BC} $\pm$ 15
3	638 ^{a,C} $\pm$ 39	930 ^{a,B} $\pm$ 44	249 ^{b,AB} $\pm$ 17	262 ^{b,BC} $\pm$ 17
4	1,085 ^{a,B} $\pm$ 54	1,188 ^{a,B} $\pm$ 50	321 ^{b,B} $\pm$ 24	393 ^{b,AB} $\pm$ 27
5	1,464 ^{a,A} $\pm$ 67	1,578 ^{a,A} $\pm$ 52	718 ^{b,A} $\pm$ 46	620 ^{b,A} $\pm$ 29

^{a,b}Values with different superscripts within a row are significantly different ($P < 0.05$).

^{A-D}Values with different superscripts within a column are significantly different ($P < 0.05$).

¹DDGS = distillers dried grains with solubles; RSM = rapeseed meal.

The supplementation and the protein source showed a significant influence ($P < 0.05$) on the iodine concentration of milk (Tables 3 and 4). Increasing iodine supplementation levels in the feed caused increased milk iodine concentrations in all groups (Table 3). Feeding RSM led to strongly diminished milk iodine concentrations compared with feeding DDGS. On the one hand, the significant interaction between supplementation level and protein source illustrated that differences between the least squares means of the RSM and DDGS groups were significant only at iodine supplementation levels of 2 mg/kg of DM and higher. Conversely, significant differences between supplementation levels were first investigated at feed iodine supplementation levels of 2 mg/kg of DM and higher in the groups with DDGS, and were investigated only at iodine supplementation levels of 4 and 5 mg/kg of DM in the RSM groups. Less significant differences in the case of RSM may be

explained by generally lower iodine concentrations at a relatively high individual cow variation.

In almost all periods, milk iodine concentrations were slightly higher in the iodate groups compared with the appropriate iodide groups (Table 3). Analysis of variance (Table 4) showed a significant influence of the iodine species on milk iodine concentration, but no significant differences were observed in any period when the respective groups were compared (Table 3).

The iodine concentration in milk showed a linear dependence on the feed iodine supplementation (Figure 2). In the region of the highest permitted feed iodine concentration (5 mg/kg), the milk iodine concentration with application of DDGS increased to 1,464 and 1,578 $\mu\text{g/kg}$ when iodide and iodate, respectively, were applied. This lack of a steady state, which would limit the attainable iodine content of milk, was found in previous studies (Swanson et al., 1990; Schöne et al., 2009).

Figure 1. Milk iodine concentration within periods of increasing iodine supplementation of the different experimental groups ($\blacktriangle$, DDGS/iodide; $\triangle$, DDGS/iodate; $\blacksquare$, RSM/iodide; $\square$, RSM/iodate). The 5-part graduation within each period represents the means of the sampling d 4, 11, 19, 20, and 21 ($n = 8$). DDGS = distillers dried grains with solubles; RSM = rapeseed meal.

Table 4. *P*-values for the influence of iodine supplementation, protein source, iodine species in the ration, and their interaction, as well as for the impact of the covariables lactation and milk yield on different milk parameters

Item	Iodine concentration	Iodine amount	Carry over rate
Supplementation	<0.001	<0.001	<0.001
Protein source	<0.001	<0.001	<0.001
Iodine species	0.027	0.350	0.588
Supplementation × protein source	<0.001	<0.001	0.014
Supplementation × iodine species	0.306	0.495	0.869
Protein source × iodine species	0.053	0.423	0.431
Supplementation × protein source × iodine species	0.249	0.219	0.713
Lactation number	0.375	0.011	0.274
Milk yield	0.758	—	—

Mean milk iodine concentrations of 2,762 and approximately 2,160 µg/kg were detected, with a maximum of approximately 6,200 µg/kg, (Hillman and Curtis, 1980; Schöne et al., 2009).

In contrast, at the highest supplementation level, milk iodine concentrations in the RSM groups amounted to only 718 and 620 µg/kg for iodide and iodate, respectively. The distribution of all values for this supplementation level after reaching the steady state showed that, in the DDGS groups, no sample contained less than the recommended maximum value for milk of 500 µg/kg (Hamann and Heesch, 1982), whereas in the RSM groups, 32% of the values were below this concentration. In the period without iodine supplementation, milk iodine concentrations (Table 3) in the DDGS groups were far above the critical concentration of 20 or 25 µg/L, indicating a deficient supply in the cow (Alderman and

Stranks, 1967; Miller et al., 1975b), whereas in the RSM groups, concentrations were barely above this critical level for iodate and slightly below this level for iodide. With regard to the various impact factors on milk iodine concentration mentioned in the introduction and the resulting test conditions of the various trials (Hemken, 1979; Franke et al., 1983; Flachowsky et al., 2007), comparison of the results of different studies appears to be difficult. The high milk iodine concentrations at the highest tested supplementation of feed (5 mg/kg of DM) in the magnitude correspond to the results found in a preliminary investigation at our institute (Schöne et al., 2009) in which, at a feed iodine supplementation level of 5.5 mg/kg of DM, a milk iodine content of 1,215 µg/kg was determined. The slightly lower milk iodine contents in the previous trial resulted from lower feed intakes, and therefore lower iodine intakes, of those ani-

Figure 2. Dependence of the milk iodine concentration (LSM; $n = 8$) on feed iodine supplementation in the experimental groups (▲, DDGS/iodide; △, DDGS/iodate; ■, RSM/iodide; □, RSM/iodate). DDGS = distillers dried grains with solubles; RSM = rapeseed meal.

imals. At comparable iodine intakes and without RSM in the ration, Swanson et al. (1990) and Herzig et al. (1999) investigated milk iodine concentrations in the magnitude of those determined in the present study in the DDGS groups, whereas Kaufmann and Rambeck (1998) determined slightly lower concentrations than in the RSM groups. No information was given on feed composition in this trial. A study by Hillman and Curtis (1980) at comparable iodine intakes showed higher milk iodine concentrations, possibly because of the use of ethylenediamine dihydroiodide as the iodine species, which seemed to allow higher carry over of feed iodine into milk compared with potassium iodide (Swanson et al., 1990). The method for analyzing iodine may also be partly responsible for the differences found (Leiterer et al., 2001).

Former studies have also shown a reducing effect of rapeseed in the ration on iodine content of the milk, even at low GSL contents (Piironen and Virtanen, 1963; Iwarsson, 1973; Papas et al., 1979; Schöne et al., 2006). Papas et al. (1979) and Schöne et al. (2006) investigated reductions in milk iodine concentrations of 78 and 54% at GSL intakes of 78.7 and 9.2 mmol/d, respectively. In the present study, average milk iodine concentrations in the groups fed RSM were reduced by 51 and up to 78% compared with the appropriate DDGS groups, showing no dependency on the level of iodine supplementation. This percentage corresponded to a reduction in milk iodine concentration by 45 to 958 $\mu\text{g/kg}$ or 4 to 75 $\mu\text{g/kg}$ per mmol of ingested GSL, respectively, at the applied type and amount of RSM (3.5 μmol of GSL/g, 16.5% of ration DM), and a linear increase with increasing iodine supplementation level in the feed. The effect of further GSL contents in the feed could not be derived from this trial. High iodine supplementation levels with application of RSM increased the iodine concentration of milk but could not completely abolish the inhibition of NIS. Like the thyroid, the mammary gland responds to GSL exposure with a reduced iodine uptake. Therefore, milk iodine better reflects the availability of the ingested iodine to the animal than does urinary iodine, which may be elevated at GSL intake (Miller et al., 1975a). The German Society of Nutrition Physiology (GfE, 2001) suggests that iodine supplementation should be twice the recommendation if the ration contains RSM. At the recommended allowance for dairy cows of 0.5 mg/kg of DM (GfE, 2001), in the DDGS groups, a milk iodine concentration of 85.2 $\mu\text{g/kg}$ would be expected for iodide and a concentration of 98 $\mu\text{g/kg}$ would be expected for iodate (calculated from the regression equation; Figure 2). Thus, milk iodine concentrations of this magnitude seem to signify a sufficient iodine supply for the animal. When applying RSM, similar milk iodine contents would be reached at feed iodine concentrations

of 1.1 mg/kg of DM both in iodide and in iodate, which corresponds quite well with the recommended doubling of the iodine supplementation when applying RSM.

Differences in iodine content of milk attributable to the iodine species used were shown for potassium iodide and ethylenediamine dihydroiodide (Swanson et al., 1990). Studies comparing the effect of potassium iodide and calcium iodate on milk iodine showed controversial results. Whereas Leskova (1969) found a greater increase in milk iodine when using iodate compared with iodide, Lengemann (1969) and Bretthauer et al. (1972) found no significant difference with daily oral iodine application. Analysis of variance of the present study indicates that higher iodine concentrations in milk may be reached with calcium iodate application compared with potassium iodide. However, the nearly significant interaction between protein source and iodine species and the following comparison of the single-group least squares means do not support this finding.

Total Milk Iodine Output

Differences in the amounts of iodine (Table 5) between the experimental groups and supplementation levels were comparable with the iodine concentrations (Table 3). The impact of the supplementation and the protein source (Tables 4 and 5) was significant, whereas the iodine species showed no significant influence. The DDGS and RSM groups were significantly different at iodine supplementation levels of 2 mg/kg of DM and higher. Significant differences between feed supplementation levels were observed at levels of 1 mg/kg of DM and higher for DDGS and beginning at 2 mg/kg of DM for RSM (Table 5). Comparable with the milk iodine concentration, the daily amount of iodine excreted with milk showed a linear dependence on the daily iodine intake of the cow.

Carry Over

Carry over was significantly influenced by the supplementation and the protein source, but not by the iodine species (Tables 4 and 6). The carry over showed no constant increase or decrease with iodine supplementation. In all groups, the lowest carry over was observed at a feed iodine supplementation level of 1 mg/kg of DM. Differences between DDGS and RSM were significant at all supplementation levels (Table 6). In cows with low milk yields fed a GSL-free ration, Schöne et al. (2009) found a comparable carry over of supplemented iodine of 35 to 47%. Compared with meat and eggs (Flachowsky et al., 2006; Franke et al., 2008; Meyer et al., 2008; Röttger et al., 2008), milk shows the highest carry over of iodine from feed into foods of animal origin.

Table 5. Total iodine amounts in milk (LSM $\pm$ SEM of values from d 19, 20, and 21, n = 24)¹

Iodine supplementation (mg/kg of DM)	DDGS/iodide (mg/d)	DDGS/iodate (mg/d)	RSM/iodide (mg/d)	RSM/iodate (mg/d)
0	1.90 ^E $\pm$ 0.19	2.03 ^F $\pm$ 0.08	0.73 ^E $\pm$ 0.07	0.87 ^D $\pm$ 0.08
0.5	5.80 ^{DE} $\pm$ 0.35	6.09 ^{EF} $\pm$ 0.26	1.99 ^{DE} $\pm$ 0.17	2.17 ^D $\pm$ 0.21
1	8.51 ^D $\pm$ 0.57	9.00 ^E $\pm$ 0.45	3.31 ^{CDE} $\pm$ 0.31	3.13 ^{CD} $\pm$ 0.20
2	21.9 ^{a,C} $\pm$ 0.95	21.1 ^{a,D} $\pm$ 0.90	7.29 ^{b,BCD} $\pm$ 0.61	8.42 ^{b,BC} $\pm$ 0.49
3	25.5 ^{a,C} $\pm$ 1.43	30.1 ^{a,C} $\pm$ 1.60	9.39 ^{b,BC} $\pm$ 0.96	8.91 ^{b,BC} $\pm$ 0.66
4	41.1 ^{a,B} $\pm$ 2.33	41.3 ^{a,B} $\pm$ 1.67	11.5 ^{b,B} $\pm$ 1.28	14.2 ^{b,AB} $\pm$ 1.10
5	47.5 ^{a,A} $\pm$ 3.01	47.8 ^{a,A} $\pm$ 1.72	21.9 ^{b,A} $\pm$ 2.18	18.8 ^{b,A} $\pm$ 0.85

^{a,b}Values with different superscripts within a row are significantly different ($P < 0.05$).

^{A-F}Values with different superscripts within a column are significantly different ($P < 0.05$).

¹DDGS = distillers dried grains with solubles; RSM = rapeseed meal.

Milk Iodine Regarding the Consumers' Need

Considering the per capita consumption of milk in 2006 by consumers in Germany (62.6 kg/yr or 172 g/d; ZMP, 2008), milk produced with the highest feed iodine supplementation presently permitted (5 mg/kg) and with DDGS as the protein source would lead to an iodine intake in the range of 250 to 270 μ g/d. This amount corresponds to 126 to 180% of the recommendation and 25 to 54% of the UL for adults (WHO, 1994; D-A-CH, 2000; WHO, UNICEF, and ICCIDD, 2001). Drinking more milk, or even connecting with iodine intake from other food sources (e.g., eggs, milk products, bread, or sausage produced with iodized salt, seafood, oceanic algae, and iodized salt in the household), may lead to exceeding the UL in human nutrition.

At present in Germany, very high feed iodine supplementation levels (<3 mg/kg of DM) are not applied in

practice. In mixed feed for dairy cows, Grünewald et al. (2006) detected a mean iodine content (minimum to maximum) of 0.79 mg/kg of DM (0.45 to 3.04 mg/kg of DM). The expectable iodine concentration in milk consumed in Germany was estimated from the regression (Figure 2). When DDGS was fed, it amounted to 184 μ g/kg (76 to 905 μ g/kg), and for RSM, it was 52 μ g/kg (10 to 332 μ g/kg). The means are in the range of milk iodine concentrations currently found in the milk consumed (Jahreis et al., 2007; Rysava et al., 2007). Regarding those mean milk iodine concentrations and the per-person consumption, the milk consumed presently contributes approximately 16 to 22% to the human iodine demand when feeding DDGS, and 5 to 6% with the tested RSM in the ration. Taking into account that milk from different producers is mixed in practice, occurrence of the estimated maximum concentrations in milk in Germany seems improbable. Nevertheless,

Table 6. Carry over¹ of iodine from feed into milk (LSM $\pm$ SEM of values from d 19, 20, and 21, n = 24, as well as overall LSM for the protein source² and iodine species³)

Iodine supplementation (mg/kg of DM)	DDGS/iodide (%)	DDGS/iodate (%)	RSM/iodide (%)	RSM/iodate (%)
0	48.2 ^{a,AB} ± 4.7	48.6 ^{a,AB} ± 2.9	18.6 ^b ± 2.7	24.6 ^b ± 3.2
0.5	51.6 ^{a,A} ± 5.7	45.1 ^{a,AB} ± 3.3	13.1 ^b ± 1.7	18.3 ^b ± 2.2
1	30.4 ^{a,B} ± 2.4	34.3 ^{a,B} ± 2.8	11.8 ^b ± 1.4	10.7 ^b ± 0.7
2	55.6 ^{a,A} ± 4.8	55.5 ^{a,A} ± 4.9	18.1 ^b ± 2.2	19.9 ^b ± 2.3
3	44.4 ^{a,AB} ± 4.4	45.2 ^{a,AB} ± 4.4	14.0 ^b ± 2.0	13.8 ^b ± 1.9
4	51.0 ^{a,A} ± 5.1	54.2 ^{a,A} ± 4.4	14.1 ^b ± 2.4	16.8 ^b ± 2.4
5	47.9 ^{a,AB} ± 4.5	44.1 ^{a,AB} ± 3.3	18.5 ^b ± 2.9	16.0 ^b ± 1.3
	DDGS		RSM	
0 to 5	46.9 ^a ± 1.3		16.3 ^b ± 0.6	
	Iodide		Iodate	
0 to 5	31.2 ± 1.8		31.9 ± 1.7	

^{a,b}Values with different superscripts within a row are significantly different ($P < 0.05$).

^{A,B}Values with different superscripts within a column are significantly different ($P < 0.05$).

¹The carry over represents the total amount of iodine in the milk per day related to the respective daily iodine intake, given as a percentage.

²Protein sources: DDGS = distillers dried grains with solubles; RSM = rapeseed meal.

³Iodine species: iodide and iodate.

an analysis of milk consumed in the Czech Republic (Kursa et al., 2004; Rysava et al., 2007) showed that much higher iodine concentrations than the mean of 184 µg/kg could occur because of regional or country-specific differences in feed iodine supplementation or manufacturing practices.

Consequences for the Maximum Level of Iodine in Feed

Regarding the cited maximum level of 500 µg of iodine/kg of milk (Hamann and Heeschen, 1982) and the regression of milk iodine on iodine in the DDGS ration (Figure 2), a maximum level of iodine in dairy cow feed below 2 mg/kg of DM would have to be recommended (see Table 3). On the other hand, the detected milk iodine concentrations confirm the estimation of the GfE (2001) that in dairy cow feed containing RSM as the protein source, iodine supplementation of at least 1 mg/kg of DM is recommended. Such a narrow range between the recommended and maximum levels does not seem to be practical because different feeding practices for dairy cows might easily result in exceeding the maximum level. Hence, with regard to the greater requirements with the application of rapeseed, the maximum level should not be decreased below 3 mg/kg.

CONCLUSIONS

Iodine supplementation of feed can considerably increase milk iodine and may therefore contribute to improving the human iodine supply. On the other hand, the high transfer of iodine from feed into milk may cause the UL in human nutrition to be exceeded at the maximum level of iodine currently permitted in feed in Europe (5 mg/kg). Therefore, the maximum level of iodine in dairy cow feed needs to be reevaluated. Because the iodine supplementation levels presently applied in practice in Germany are far below this maximum level, no acute risk of overdose exists at the moment. When the iodine content of milk is estimated from the iodine concentration of feed, the use of RSM in the ration must be considered. Milk iodine concentrations may vary with application of potassium iodide or calcium iodate because of metabolic differences. The impact of storage losses could not be investigated in this trial.

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