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Iodine concentrations in milk

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Iodine tends to be supplemented at farm level in the expectation of increasing cow health and fertility. There is concern that such practices may result in high milk iodine, which could affect ingredients for infant formula and, thus, dairy export markets. The objective of this study was to quantify the effect of iodine fortified feed and teat disinfection practices of dairy cows on milk iodine concentration. Thirty lactating cows were fed 7 kg, 3 kg (10 mg iodine/kg) and 0 kg of concentrate feed during 3 periods of 35 days each. During the first 14 days of each period, cows were on dietary iodine treatments only; during days 15–21, one of three teat disinfection treatments (n = 10) was applied (in addition to the dietary iodine treatments): non-iodine (chlorhexidine) post-milking spray; 0.5% iodine spray post-milking; 0.5% iodine spray pre- and post-milking. Cow milk yield was 21.3 kg/day. Individual cow milk samples were analysed for iodine concentration on 2 days at the end of each treatment period. Dietary supplementation of iodine at both 30 mg and 70 mg/day, when compared to the diet with no supplement, increased milk iodine concentrations significantly ($P < 0.001$) from 449 to 1034 and 915 µg/kg, respectively. Teat disinfection both pre- and post-milking increased milk iodine concentration at each of the dietary supplementation levels of 0, 30 and 70 mg/day compared with a non-iodine teat disinfectant ($P < 0.001$). In conclusion, both dietary iodine supplementation and teat disinfection iodine increased milk iodine concentrations in an additive manner, exceeding common target values of 250 µg/kg. As both iodine treatments can occur simultaneously on farm, supplementation strategies should be monitored.

Keywords: dietary supplementation; iodine; milk; teat disinfection

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Introduction

Iodine is an essential trace element for humans and animals. Iodine is incorporated into the thyroid hormones, which have multiple functions as cell activity regulators. Iodine deficiency affects reproductive capacity, brain development and progeny as well as growth. The iodine requirement of humans depends on age and physiological state, and the recommended intake levels vary depending on the scientific group, for example, US Food and Nutrition Board (2001) and the German nutritional reference point (DACH 2000) recommend 150 µg/day and 180–200 µg/day for adults, respectively, while the corresponding recommendations for children up to 1 year old are 110–130 µg/day and 40–80 µg/day. Recommended iodine intake for animals has also been published, with the GfE German Society of Nutrition & Physiology (2006) and the US National Research Council (2001) both recommending 0.5 mg of iodine/kg dry matter (DM) intake/day, or approximately 10–12 mg of iodine/cow/day. Milk and dairy products are important sources of iodine. But excess iodine intake by cows is secreted into milk (and excreted in urine), and can be consumed by humans. This is important as the tolerable upper level of iodine is only ~3 times higher than the adult requirement (200 *vs*. 600 mg/day – European Commission 2002). Therefore, milk iodine levels need to be monitored.

Iodine is frequently used as a dietary supplement to dairy cows in the expectation of increasing cow health and fertility, and is therefore routinely added to feed concentrate at varying levels of 5–10 mg/kg. Specifically, in the Irish scenario, Rogers (2003) recommended iodine supplementation levels of 12–60 mg of iodine/cow/day to counteract previously recorded deficiencies (Rogers 1999). Teat disinfection with iodine has been shown to reduce the mastitis infection rate (Galton, Peterson and Merrill 1988) and, thus, is used as a routine practice on-farm post-milking, and in some instances is used as part of pre-milking teat preparation.

The seasonal milk production system in Ireland may exacerbate milk iodine concentrations due to cows being fed supplementary concentrate fortified with iodine, in addition to teat disinfection with iodine. This is of most concern at specific times, e.g. early and late lactation and during winter milk production. The most recent measurement of iodine in the Irish spring milk production system recorded an average of 227 µg/L (O'Brien, Mehra and Connolly 1999), but concentrations of 510 µg/kg and 180 µg/kg were recorded for December and June, respectively.

Ireland is a major producer, processor and exporter of milk (€2.2 bn in 2008). To protect this market, caution needs to be exercised with respect to milk iodine levels. Ireland is one of the leading infant formula manufacturers worldwide. The limit for iodine in milk powder as an ingredient in infant feed formula is 130 µg iodine/100 g powder; the target or preferred limit is 100 µg iodine/100 g powder. This equates to <250 µg iodine/kg fresh milk.

The objectives of this study were to quantify the effects of dietary iodine supplementation and teat disinfection with iodine on milk iodine concentrations of dairy cows, in order to give guidelines for farm practices regarding iodine usage.

Materials and Methods

Pre-experimental cow selection

A group of 40 Holstein Friesian lactating dairy cows were segregated as a herd and offered 7 kg concentrate (containing 10 mg iodine/kg) and silage. Teat preparation during this 12-day period consisted of washing and drying cows pre-milking. These cows were teat disinfected post-milking by spraying with a non-iodine containing product (chlorohexidine). After 11 days, each individual cow was milk sampled at morning and evening milking of the same 24 h period (am + pm). The evening and morning milk samples were proportionally mixed so as to represent milk from that cow over a 24 h period. This procedure was repeated for a further 24 h period. Each of the 80 samples was split into duplicate samples. All samples were frozen immediately and analysed for iodine content within 1 week. These data were used to select 30 cows that reacted similarly to the treatment applied and had similar milk iodine concentrations.

Pre-experimental iodine depletion period and randomisation of cows

The 30 lactating cows selected had a mean calving date of 21 October, mean days in milk (DIM) of 104, and an average milk yield of 21.6 kg/cow/day at the start of the trial, and were managed as one group for the duration of this study. During a pre-experimental period of 12 days, cows received a diet of grass silage *ad lib* + 7 kg fresh concentrate/cow/day, which contained no added iodine (Table 1). The concentrate was offered at milking time: 4 kg at morning and 3 kg at evening milking. During the last 2 days of the pre-experimental period (days 11 and 12), individual cow milk samples were collected and analysed for iodine concentration. The 30 cows were blocked into 10 blocks with 3 cows each (one cow from each block was then randomly assigned to 1 of 3 treatment groups) based on milk iodine concentration and DIM.

Experimental treatments

The cows were offered 7 kg and 3 kg (both including 10 mg iodine/kg) and 0 kg fresh concentrate during 3 periods of 35 days each. During the first 14 days of each period, cows were on one of the dietary iodine treatments only; during days 15–21, 1 of 3 teat disinfection treatments was applied (in addition to the dietary iodine treatments) to each of 3 cow groups (n = 10): non-iodine (chlorhexidine) post-milking; 0.5% iodine post-milking; 0.5% iodine pre- and post-milking. Cows received 12 (Period 1) and 16 kg (Period 2) DM silage/day during the first 2 treatment periods and 18 kg DM grass/cow/day during the third treatment period. Teat disinfectant was applied by spray and when pre-milking disinfectant was applied, teats were dried with individual paper towels prior to cluster attachment. Dietary supplementation and depletion periods of 14 days were allowed, based on data reported in Flachowsky *et al*. (2007) and Schöne *et al.* (2006).

Individual cow milk samples were collected on the last two days of each of the three trial periods (n = 360). Day milk samples represented aliquots of am and pm milks where the am and pm milks were added proportionally according to yield. Samples were then frozen at –20 °C. Milks were analysed for iodine concentration by inductively coupled plasma mass spectrometry within approximately 3 months of sample collection.

Statistical analysis of the data was undertaken using SAS (SAS 2011). As the dietary treatments were applied in turn in different periods, the dietary iodine effect was confounded with any other changes from one period to the next. The other constituent portions of the diet besides concentrate were grass and silage. But the levels of iodine in grass and silage are

Table 1. Schedule of dietary iodine and teat disinfection treatments

Period	Days	Treatments		
Cow selection	1–12	7 kg concentrate (10 mg/kg I) (70 mg I) + wash and dry cows pre-milking and spray cows post-milking with 'non-iodine'		
Pre-experimental iodine depletion and cow randomisation	1–12	7 kg concentrate (0 mg I/day) + wash and dry cows and spray cows post-milking with 'non-iodine'		
Experimental treatments Period 1	1–14 Dietary iodine	7 kg concentrate (10 mg/kg I) (70 mg I) + wash and dry cows pre-milking and spray cows post-milking with 'non-iodine'		
	15–21 Teat disinfection iodine	Group 1 (10 cows) 70 mg I + PRE Iodine + POST Iodine	Group 2 (10 cows) 70 mg I + POST Iodine	Group 3 (10 cows) 70 mg I + 'non-iodine'
	22–35 Iodine depletion	3 kg concentrate (0 mg I/day) + wash and dry cows and spray cows post-milking with 'non-iodine'		
Experimental treatments Period 2	1–14 Dietary iodine	3 kg concentrate (10 mg/kg I) (30 mg I) + wash and dry cows pre-milking and spray cows post-milking with 'non-iodine'		
	15–21 Teat disinfection iodine	Group 1 (10 cows) 30 mg I + 'non-iodine'	Group 2 (10 cows) 30 mg I + PRE Iodine + POST Iodine	Group 3 (10 cows) 30 mg I + POST Iodine
	22–35 Iodine depletion	0 kg concentrate (0 mg I/day) + wash and dry cows pre-milking and spray cows post-milking with 'non-iodine'		
Experimental treatments Period 3	1–14 Dietary iodine	0 kg concentrate (0 mg I/day) + wash and dry cows pre-milking and spray cows post-milking with 'non-iodine'		
	15–21 Teat disinfection iodine	Group 1 (10 cows) 0 mg I + POST Iodine	Group 2 (10 cows) 0 mg I + 'non-iodine'	Group 3 (10 cows) 0 mg I + PRE Iodine + POST Iodine
	22–35 Iodine depletion	0 kg concentrate (0 mg I/day) + wash and dry cows pre-milking and spray cows post-milking with 'non-iodine'		

similar and relatively low. Rogers (2005) indicated the iodine levels in grass and silage to be 0.26 and 0.27 mg/kg DM and the change in proportions as the season advanced should not have an important impact on iodine measurements. Adding grass to the diet and increasing the total herbage (grass and silage) intake from say 10 kg/cow/day to approximately 18–19 kg/cow/day, would mean an increase in intake of approximately 2.25 mg iodine/cow/day.

The disinfection was totally controlled as a non-iodine disinfection was applied at all times, except when the pre- or post- or both disinfection treatments with iodine were required by the trial.

On this basis, a factorial structure in the treatments (3 levels of dietary iodine and 3 disinfection treatments) was used as the basis for the analysis. Main effects of dietary addition are to be interpreted in the same way as observational data, where other confounding factors cannot be completely discounted. Within-animal correlations were included in the analysis using the mixed model procedures in SAS. Baseline data was tested for inclusion in the final analysis model. Residual checks were made to ensure that the assumptions of the analysis were valid.

Results and Discussion

It is critical that milk iodine concentration meets market requirements for ingredients to infant feed formula. The influence of dietary and teat disinfection iodine within a seasonal milk production system on milk iodine concentrations will indicate the necessity to modify cow iodine supplementation practices.

There was no significant interaction ($P = 0.11$) of dietary supplementation and disinfection treatment. Dietary supplementation of iodine at both 30 mg and 70 mg/day, when compared to the diet with no supplement, increased milk iodine concentrations significantly ($P < 0.001$) from 449 μg/kg to 1034 and 915 μg/kg, respectively (Table 2). There were significant differences between the disinfection methods (Table 3). The lowest level of milk iodine concentration was found for the non-iodine product (574 μg/kg) and both the post-milking teat disinfection with iodine (753 μg/kg) and the pre- and post-milking teat disinfection with iodine (1071 μg/kg) showed significant increases ($P < 0.001$) relative to the non-iodine product. The observed milk iodine concentrations for each treatment combination are shown in Table 4.

Regressing on the levels of dietary iodine addition was used to assess trend and

Table 2. Effect of two different levels of dietary iodine supplementation on milk iodine levels

	0 mg iodine/day	30 mg iodine/day	70 mg iodine/day	Standard error	Significance
Milk iodine (μg/kg)	449[a]	1,034[b]	915[b]	39.9	***

[a,b]Superscripts indicate a significant difference of $P < 0.001$.

Table 3. Effect of teat disinfection practices with iodine on milk iodine level

	Non-iodine teat dipping	Post-milking teat dipping with iodine	Pre- and post-milking teat dipping with iodine	Standard error	Significance
Milk iodine (μg/kg)	574[a]	753[b]	1,071[c]	39.9	***

[a,b,c]Superscripts indicate a significant difference of $P < 0.01$.

Table 4. Effect of teat disinfection practises with iodine on milk iodine level (µg/kg) at three different levels of dietary supplementation

	Non-iodine teat dipping	Post-milking teat dipping with iodine	Pre- and post-milking teat dipping with iodine	Average standard error
70 mg iodine/day	813	817	1,115	39.4
30 mg iodine/day	692	982	1,429	96.4
0 mg iodine/day	217	461	670	45.5

non-linearity in the relationship between those levels and iodine concentrations in milk, adjusting for block and disinfection treatment. There was a positive trend with increasing levels of dietary addition ($P < 0.001$) and evidence of curvature (quadratic term, $P < 0.001$). Baseline values were tested as a covariate in the analysis and the relationship was found to be non-significant.

These results indicate very significant increases in milk iodine concentrations as a consequence of both diet and disinfection treatments, showing an increase from 217 µg/kg when dietary and disinfection iodine was absent to >1,000 µg/kg when both pre- and post-disinfection with iodine and either 30 or 70 µg/day were applied.

Norouzian and Azizi (2013) indicated that when iodine intake is at the recommended feeding amount of 0.5 mg/kg DM (US National Research Council 2001), milk iodine will be approximately 160 µg/kg. When supplemental iodine was added to a low iodine diet (i.e. increased from 11 to 62 mg/cow/day), the milk iodine concentration was increased 3.3 fold. However, when supplemental iodine was increased from 62 to 165 mg/cow/day, the milk iodine increased by only 12%. This suggests that milk iodine response curves are not linear. The data also show some evidence of nonlinearity. However, the exact mechanisms for this reduction in response at higher concentrations of iodine supplementation are not clear; a possible explanation might be that the mammary gland of the dairy cow may act as bio-regulator for iodine excretion (Norouzian 2011).

In a study by Borucki Castro *et al*. (2010), the milk iodine concentration from 501 farms across Canada varied considerably and appeared to be influenced by feeding practices. More than 85% of the farms tested were feeding more iodine than recommended (0.5 mg iodine/kg DM). But the authors considered that variations in the iodine concentrations of forages were unlikely to cause iodine overfeeding. Similarly, in the current study, it is unlikely that silage or grass contributed significant levels of iodine to the cow diet at different stages of the lactation; Rogers (2005) reported iodine levels of 0.26 and 0.27 mg/kg DM in Irish pasture and silage, respectively, while Wichtel *et al.* 1996 and Grace and Waghorn (2005) recorded herbage iodine levels of 0.18 to 0.27 mg/kg DM.

In the current study, cows were supplemented with approximately 4 mg/kg DM (when calculated for the complete grass and concentrate diet of the cow), which is eight times the animal requirement according to the GfE (2006) and the US National Research Council (2001). The resultant milk iodine concentrations were considerably greater than the target desired for processing of the milk into milk powder for infant formula and potentially other products also. However, that supplementation was within the maximum dietary levels of the present European Union legislation (European Commission 2005) (5 mg iodine/kg DM of cow feed), (maximum iodine concentration in cow feed reduced from 10 to 5 mg/kg in 2005). This European Union legislation was influenced by studies that indicated a 30–40% transfer of ingested supplemental iodine to milk (Schöne *et al*. 2006; Flachowsky *et al*. 2007). Additionally, there are no reports (known to these authors) indicating beneficial effects of iodine overfeeding on reproduction, animal growth or fattening (Wichtel *et al*. 1996; Meyer *et al*. 2008). As milk iodine concentration is critical for some dairy products, it may be prudent to measure milk iodine routinely. Additionally, from a herd health perspective, measurement of iodine may be necessary as an indicator of the iodine status of the herd.

The variable increase in milk iodine associated with the different teat disinfection practices was not unexpected. As teat preparation (washing of teats and drying with paper towel) was undertaken pre-milking, the contribution of post-disinfection iodine to milk iodine was probably largely due to absorption through skin. Considerable variation has been reported in trials investigating the effect of different teat disinfection practices on milk iodine between 1980 and 2007, but much of it was due to differences in iodine strength which varied between 0.1% and 0.8%, formulation, analytical methods, housing conditions and teat dipping versus spraying. A study by Flachowsky *et al.* (2007) reported an increase of 54 µg iodine/kg of milk when cows were teat dipped with a 0.3% iodine solution post-milking. Pre-milking disinfection can pose a substantial risk of iodine transfer to milk, as it is dependent on the degree of removal from the teats prior to cluster attachment; if not sufficiently removed the iodine may enter the milk directly during milk removal or may be absorbed through the teat skin. Galton *et al*. (1984) and Rasmussen, Galton and Peterson (1991) reported that pre-dipping with 0.5% iodine solutions followed by complete drying of the teat did not significantly increase milk iodine concentrations. However, pre-dipping with 1% iodine solution increased the milk iodine content (Rasmussen *et al*. 1991). Galton *et al*. (1984) also reported that attaching the cluster without drying the teats after dipping in 1% iodine sanitiser increased the milk iodine by 600 µg/kg above the concentration reported when teats were dried before milking.

Conclusion

In conclusion, both dietary iodine supplementation and teat disinfection using iodine resulted in milk iodine concentrations exceeding common target values of 250 µg/kg. With milk iodine levels of approximately 200 µg/kg in the absence of dietary and disinfection iodine, considered in association with a target level of 250 µg/kg, there is minimal flexibility for iodine supplementation, if target milk iodine concentrations are to be achieved.

In the immediate future, both dietary iodine supplementation of the lactating cow and teat disinfection with iodine should be undertaken with extreme caution. In the longer term, it is necessary to investigate the complete iodine budget in the lactating cow, quantifying total dietary iodine intake, blood iodine concentration and excess iodine released in milk and urine. Such a study would allow optimum cow iodine concentrations to be established, thus allowing adequate cow diet and management practices to be developed.

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