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Iodine deficiency is uncommon in North America (18). In fact, the issue may be that part of the population is consuming iodine in excess. High levels of iodine in the diet may inhibit the function of the thyroid gland and produce symptoms of iodine deficiency (9). Normal diets are unlikely to supply more than 1,000 µg of iodine per day, and most adults are tolerant to a high iodine intake (1,100 µg/day) from food. However, the upper tolerance limit for iodine consumption is lower for children aged 1 year (200 µg/day) to 8 years (300 µg/day) (18).

Cow's milk has become one of the most important sources of nutritional iodine in several countries (8, 24, 30). However, the tolerable iodine intake level could easily be exceeded with high iodine concentrations in milk (11, 12, 31). In 2004 and 2005, a study was conducted in Canada on a total of 411 retail milk samples from nine provinces and 34 brand names. The samples were collected and analyzed by Health Canada (28). The report concluded that the average iodine content of Canadian retail milk was high, at 393 ± 150 µg/kg.

One of the main factors that determine the iodine content in cow's milk is the iodine consumed by the animal (25). Several studies found that, depending on the level of iodine intake, the carryover effect from feed to milk ranged from 7 to 27% (20, 26, 27). The use of iodized teat dips and sanitizers are also factors reported to increase the level of iodine in milk (12, 16). Direct contamination from and absorption through teat skin were reported to increase milk iodine when iodophors were used in pre- and postmilking management (7).

The objectives of the present study were to evaluate the current iodine content in milk, before processing, in all provinces of Canada, and to determine the factors in feeding and milking management associated with high levels of iodine in milk.

MATERIALS AND METHODS

Sampling. Milk samples were collected from the bulk tanks of farms in all provinces of Canada. The sampling frame consisted of farms that had direct or indirect contact with the various institutions that collaborated on the sampling process for the project (dairy farmers' associations, provincial dairy boards, or provincial departments of agriculture). The number of samples collected was as follows: 200 in the province of Quebec, 100 in Ontario, 35 in New Brunswick, 50 in Nova Scotia, 50 in Prince Edward Island, 5 in Newfoundland and Labrador, 20 in Manitoba, 8 in Saskatchewan, 12 in Alberta, and 20 in British Columbia. With a view to characterizing the farms' feeding programs and milking management practices, a questionnaire was completed at each farm. The questions were based on a questionnaire used previously by Brander (5).

Analysis. The milk samples were analyzed at Health Canada's accredited laboratory in Longueuil, Quebec, Canada. Total iodine concentration (organic and inorganic) was determined by the method of Benkhedda et al. (2) by means of inductively coupled plasma mass spectrometry (7500 series model, Agilent Technologies) optimized for raw milk samples. Prior to analysis, the samples were digested in a closed microwave system using a mixture of perchloric and nitric acids. The detection limit was 12 ng/g for a 0.5-g sample, with precisions of 4.0 and 2.2% obtained for 10 replicate measurements of 50- and 1,000-ng/g standards.

Statistical analysis. An initial step consisted of performing descriptive statistics, regression analysis, and analysis of variance followed by looking at group differences using Student's t tests with the Data Analysis Toolpak of Microsoft Office Excel. All statistical testing was at the 5% level. The farms were grouped according to their characteristics for each of the variables, to determine if the differences in iodine concentrations between groups were statistically significant. Based on these results, further analyses were restricted to 15 variables selected from the questionnaire (29). A general linear model was fitted, with iodine levels in the milk as the response variable and all main effects and two-way interaction effects as the explanatory variables. These response variables are listed in Table 1 and included three constructed variables, SYST, HOW_DISIN, and NEW_PL.?

The SYST variable was constructed from the variables "milk system" (MLKSYS) and "stall configuration" (PIPCONF); these two variables overlap and are structurally correlated, given that, when MLKSYS had a value of "pari" (parlor), then PIPCONF had a value of "nr" (no response). Conversely, if MLKSYS had a value of "pipeline," then PIPCONF had a numerical value. The constructed variable had a numerical value if PIPCONF did but had a value of "pari" if MLKSYS had a value of "pari" and a value of "other" in all other cases. This variable is treated as a categorical variable.

The other reason for constructing variables was to remove cells with very few (five or fewer) observations, combining them with an adjacent cell; this was the case with the variable "number of milking units used" (PIPUNTT). The new variable (NEW_PIP) took cells with a low observed number of farms and combined these cells with an adjacent cell. For example, there were four farms that had two milking units (no farms had just one); those four farms were combined with the 21 farms with three milking units. Similarly, the farms with more than nine milking units were combined with the farms with nine milking units. As well, the farms that reported "nr" when MLKSYS indicated that the operation was a parlor were assigned to the cell that had zero milking units. The HOW-DISIN variable was constructed from the variables PREDIP (whether premilking teat disinfection is practiced) and PREHOW (how premilking teat disinfection is applied).

During model building, several interaction terms had 0 df because of the structure of the data and hence could not be tested. A process of model reduction was used to reduce the number of effects into a final model. This process consisted of removing all two-way and main effects that tested not significant according to analysis of variance. Then, a test of the reduction of the sums of squares attributed to the removal of these variables was done and found to be statistically not significant, with a P value of 0.16.

Least-squares means and the difference between effects were not calculated, because some terms were not estimable in these data. For presentation purposes,

however, "equal cell means" were calculated by obtaining the mean for each cell, summing across all the cells for that effect, and dividing by the number of cells that had observations. This step was done to minimize the dominance of cells that had a large number of observations. Each cell was treated equally regardless of the number of observations in it.

RESULTS AND DISCUSSION

The mean value for iodine concentrations in milk in Canada (501 farms) was found to be 304 ± 8.4 pg/kg with a median of 265 pg/kg. There was a wide range of iodine concentrations (54 to 1,902 pg/kg), with a high coefficient of variation (61.7%). Grouped frequency distribution shows that 44% of the farms presented concentrations of iodine between 151 and 300 $\mu\text{g}/\text{kg}$ (Fig. 1) and that 85% of the milk samples contained more than 150 μg of iodine per kg.

In view of the recommendations of Canada's Food Guide stating that children should consume 500 to 750 ml of milk daily (17) and some may consume 1 liter/day, it is estimated that on the average the level of iodine in milk should be maintained around 200 $\mu\text{g}/\text{kg}$, assuming that milk is the main source of iodine for young children. In Canada, the iodine levels observed in bulk tank milk were in the same range as those observed previously in retail milk (28), suggesting that most milk iodine is already present at the farm gate.

In the various provinces sampled, mean values for iodine ranged between 180 and 358 $\mu\text{g}/\text{kg}$ (Table 2). Differences between provinces were significant ($P < 0.001$), with higher values for Central and Atlantic Canada compared to the Prairies and Western Canada. Previous studies in Canada were done locally in the various provinces in different years, with different analytical methods used to determine iodine in milk. Previous sampling of 1,516 Ontario dairy herds in 1998 revealed a lower milk iodine content, 285 ± 7 pg/kg, compared with the level found in this study (325 pg/kg) (23). Values for Quebec were found to be lower than previous sampling values, i.e., 329 versus 490 pg/kg in 1979 (21). For Saskatchewan, values were found to be similar to previously determined values,

i.e., 270 versus 300 µg/kg (6). Between 1965 and 1980, a positive association between milk iodine concentration and the addition of organic iodine in the diet and medication was found. The implementation of a ban on the use of organic iodine (ethylenediamine dihydriodide) in medication in Canada did not, however, result in a significant lowering of iodine levels in dairy products (10, 22). In 2004 and 2005, the average iodine content in Canadian retail milk was 393 ± 150 µg/kg, compared with 302 ± 165 µg/kg in 1992.

The variables selected in the modeling process to explain the possible factors associated with actual milk iodine concentrations in Canada are presented in Table 3. The regression coefficient (R^2) was 36.4%, implying that the model explains only 36% of the variation in the data; this value is considered low. Equal cell means for the variables along with the number of farms that had each response are shown in Tables 4 to 8. Since the aim of the study was to ascertain farm practices that influence iodine levels in milk, equal cell means gave an indication of the direction of the effects for a specific variable. They should not be considered "cause-effect" variables but rather ones that have a statistical association with the iodine levels in bulk tank milk. The interaction effect between ration and type of wipe used ($RATION \times DWIPTYPE$) should have had 15 df but had only 8 df. Empty cells created problems (as did several cells with only one to three observations), and hence only certain combinations could be tested.

The farms with automatic tank wash systems presented higher levels of iodine in milk (Table 4). It is possible that sanitizer residues in the automatic systems (pipeline units and tank) contaminate the milk. However, conclusions should not be drawn without further investigation. Previous studies on automatic systems involving mainly automatic dipping (14) found no associations with iodine in milk.

When grouped by milking system (parlor or pipeline) and automatic take-off, the farms presented numerically different milk iodine values, but these differences were not statistically significant. Type of housing (free or tie-stall) presented no

relationship with iodine levels. In the work of Leslie et al. (23), herds kept in free-stall housing and other housing systems (combined) had significantly higher iodine contents than tie-stall-housed herds. In the present study, farms grouped by parlor type (herringbone, parallel, etc.), configuration (double or single), or number of stalls presented no differences in terms of milk iodine levels. The effect of the variables "pipeline configuration," "number of milking units," or "people involved in milking" was not statistically significant.

One of the main factors that determines the iodine content in cow's milk is the iodine consumed by the animal (25). Some reports showed that the transfer from the amount of iodine fed to the amount of iodine in milk could range from 7 to 27% depending on how much the animal consumes (20, 26, 27). Caution is necessary in diet formulation for lactating dairy cows, because feeding iodine in excess will immediately result in higher concentrations of iodine in milk. In the present study, component feeding (feeding forages and concentrates separately) was associated with lower iodine levels in milk when compared with total mixed rations (TMR) (Table 5). One possible explanation is that the use of iodized mineral mixtures, which are important sources of iodine in dairy diets, may be more frequent in farms feeding TMR.

According to Underwood and Suttle (33), iodine concentration in the basal diet is influenced by forage and crop species and variety, as well as by soil and marine deposition of iodine, which decreases with distance from the sea. The effects of factors such as goitrogens in the feed and iodine species (e.g., iodide versus iodate) have also been shown to influence the amount of iodine in milk (13). Nevertheless, the main source of iodine in dairy rations is the intensified use of iodized mineral mixtures (1, 19). No information on the iodine concentrations of the diets or feeds was supplied in the questionnaire, however. In the present study, the variables "use of mineral supplementation" and "form of mineral supplementation" (complete ration, supplemented concentrate, free-choice block, or mineral mixture) showed no significant association with the levels of iodine in milk. Further investigations are required to determine the effect on iodine

concentration of different dietary ingredients and the amounts used, in order to control the effect of dietary supply on milk iodine.

Although the farms that reported prewashing of the teats before milking presented numerically lower concentrations of iodine in milk, this variable was not statistically significant. The farms using predipping before milking presented higher levels of iodine in milk. Blowey and Collis (4) found no effect of predipping on milk iodine, but Galton et al. (14-16) and Flachowsky et al. (12) reported that pre- and postdipping practices increase milk iodine levels. In 1984, Galton et al. (16) found a combination of pre- and postdipping effects with significantly higher values of milk iodine than in the control. The reported increase in milk iodine ranged from 28.6 to 1,067 µg/liter, with larger effects for treatments that included no cleaning and drying of the teats before milking. A more recent study by Galton (14) found that the effect of postdipping (with a 0.5% iodine product) was significant and increased milk iodine by 27.0 to 31.8 µg/liter on average. The effect disappeared when the practice of postdipping was stopped. Flachowsky et al. (12) found that dipping with iodophors significantly increased the mean iodine concentration, from 100 + 23 to 154 + 42 µg/kg of milk, when compared with the levels observed after 18 days without dipping.

Pre- and postdipping using sprays (hand or in-line application) were associated with higher levels of iodine in milk than those observed with the dip-cup procedure ($P < 0.001$; Tables 6 and 7). In general, spraying tends to use more disinfectant than dipping (15 versus 10 ml) (32), and the technique is not effective unless the teats are totally covered. In-line spraying is more effective in covering the teats, but the rest of the mammary gland is also partially sprayed (3). Direct absorption of iodine through the skin has been demonstrated by Conrad and Hemken (7). Indeed, they reported increasing milk iodine values from both halves of the udder even though teat dip was used only on one side. Therefore, increasing the area of skin covered is likely to influence the amount of iodine absorbed and its concentration in milk.

The farms that used treatment with an iodine ointment for early signs of mastitis (19 of 452 farms) presented higher levels of iodine in milk (Table 8). Again, iodine rubbed onto the teats may be absorbed through the skin and cause an increase in milk iodine concentration.

The results of this study confirm that actions should be taken to reduce iodine levels in Canadian milk. Ration formulation and dipping practices at milking appear to be the main determinants of the amount of iodine in milk. Controlled studies are necessary to quantify the relationship between iodine intake, the presence of goitrogens, and milk iodine. This information could then be used to provide precise recommendations for iodine levels in diet formulation. In addition, controlled experiments to study milking management practices, such as the use of iodophors for teat dipping or spraying, should be considered to confirm the presence of a cause-effect relationship and assess its contribution to milk iodine levels.

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