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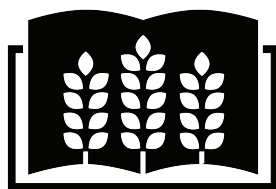
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Abstract: The objective was to determine vitamin D status of Old Order Anabaptist children in rural Southwestern, Ontario, Canada, given concerns of community healthcare professionals. Fifty-two children (2.5 months - 6.5 years) (56% female) were recruited. Finger prick blood spot (BSp) samples were analyzed for 25-hydroxy (OH) vitamins D2 & D3 (BSp25(OH)D). Three-day food records were evaluated using Dietary Reference Intakes and Canada's Food Guide (CFG) (Bush, et al. 2007). Compared to national Canadian data: mean BSp25(OH)D concentrations (78±31 nmol/L) were similar; a slightly smaller proportion (0% vs 2%) were at risk of deficiency (<30 nmol/L) or had inadequate status (4% vs 7%) (<40 nmol/L); and 10% vs 1% had BSp25(OH)D higher than 125 nmol/L. BSp25(OH)D was significantly associated ($r^2=0.358$; $p=0.001$) with



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total vitamin D intake. From food alone, vitamin D intake was 68 ± 39 IU/day, lower than the Recommended Dietary Allowance (RDA) of 600 IU/day, and intakes were all below the Estimated Average Requirement (EAR) of 400 IU. Even including supplemental vitamin D, 87% were below the EAR (total intake= 213 ± 194 IU/day). No children had vitamin D intakes greater than the Upper Limit. Servings of milk and alternates were 1.6 ± 0.8 /day (CFG= 2 /day). Unfortified farm milk was consumed by 88% of children and 89% received a vitamin D supplement. Results were comparable to recent Canadian data suggesting that most children have adequate vitamin D status. Nevertheless, these findings support the need to encourage appropriate vitamin D intake (from food and supplements) to achieve the RDA for Old Order Anabaptist children in these communities. [Abstract by authors.]

Keywords: Old Order Mennonite; Old Order Amish; dietary supplements; vitamin fortification; raw milk; public health policy; sun exposure

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Conflicts of Interest: The authors declare that they have no competing interests.

INTRODUCTION

This study assesses vitamin D status of Old Order Amish and Old Order Mennonite children in rural Southwestern Ontario, Canada. Vitamin D is an essential fat-soluble vitamin that maintains blood concentrations of calcium and phosphorus to ensure adequate bone mineralization. Vitamin D is also essential for bone growth and remodelling. In addition, vitamin D plays a hormonal role in inflammation, cell growth, immune function and glucose metabolism (Institute of Medicine 2011).

Vitamin D can be obtained from the diet, supplements, and exposure to the sun (Institute of Medicine 2011). Only a few foods such as egg yolks, fatty fish, and fish oil are good sources of naturally occurring vitamin D; just small amounts of vitamin D can be found in animal products (as vitamin D₃) and in plants (as vitamin D₂) (Roseland, et al. 2018). Foods fortified with vitamin D are the primary source of vitamin D in North America (Calvo, Whiting, and Barton 2004; Institute of Medicine 2011). For example, in both Canada and the United States, fluid milk is

fortified (mandated) with vitamin D (Government of Canada 2022; Itkonen, Erkkola, and Lamberg-Allardt 2018) and is the major source of dietary vitamin D for Canadians (Government of Canada 2022). Plant-based alternatives to milk are also typically fortified (but not mandated) with vitamin D (Government of Canada 2022). Due to the challenges of obtaining adequate vitamin D from food, vitamin D supplements are extremely important in Canada, the northern United States, and other northern countries (Itkonen, Erkkola, and Lamberg-Allardt 2018).

Vitamin D can be produced by the skin through exposure to ultraviolet rays from the sun (Bikle 2011; Holick 2007; Holick, et al. 2011; Institute of Medicine 2011). At northern latitudes, however, production of vitamin D from the skin is limited (Institute of Medicine 2011; Itkonen, Erkkola, and Lamberg-Allardt 2018). Other factors affecting production of vitamin D via sun exposure include season (with lower production in winter) and use of sunscreen (Institute of Medicine 2011). Organizations such as the Canadian Cancer Society recommend using sunscreen and limiting

sun exposure due to the risk of developing skin cancer (Canadian Cancer Society 2023).

Vitamin D is important for young children due to its role in bone growth; deficiency can result in rickets where bone mineralization is affected and may result in skeletal abnormalities and soft bones (Institute of Medicine 2011). In Canada, the incidence rate of rickets reported from 2002-2004 from 2325 pediatricians was 2.9/100,000; most cases were in breastfed infants (Ward, et al. 2007).

In Canada, a supplement of 400 IU of vitamin D is currently recommended for all healthy term infants (exclusively breastfed, partially breastfed and formula-fed) (Government of Canada 2022); these recommendations are currently under revision. For children 1-18 years, a supplement of 400 IU of vitamin D is recommended in addition to 200 IU of vitamin D from food; children who do not consume cow's milk, infant formula, or appropriate fortified plant-based beverages may need additional vitamin D (Alberta Health Services 2023). The Tolerable Upper Limit (UL) for vitamin D for children 1-3 years of age is 2500 IU/day and for children 4-8 years of age is 3000 IU/day (Institute of Medicine 2011). The Dietary Reference Intakes (DRIs) do not specify a UL for infants (0-12 months of age) (Institute of Medicine 2011). The UL for vitamin D is based on hypercalcemia of hypervitaminosis D (Institute of Medicine 2011).

The vitamin D status for young Old Order Anabaptists in Southwestern Ontario is undocumented. The Old Order Amish and the Old Order Mennonites, who use horses and buggies for transportation, are two of several groups of Old Order Anabaptists in rural Southwestern Ontario, Canada (Perth District Health Unit 2012). The Old Order Anabaptist lifestyle differs considerably from that of society's dominant culture, in terms of style of dress, avoidance of modern conveniences, education, and health-related practices (Perth District Health Unit 2012). Within these communities, transportation by horse and buggy limits the distance of travel, which may impact access to healthcare. Other risk factors that may impact health for these communities include living on low incomes, lack of access to a phone and other technology, lack of hydro and refrigeration, nonparticipation in social welfare services such as Ontario Health Insurance, limited access to family

physicians, and low health literacy (Perth District Health Unit 2012).

Huron Perth Public Health (formerly Perth District Health Unit) in Southwestern Ontario, has supported public health initiatives, involving healthcare practitioners including nurses, midwives, dietitians and nurse practitioners, in the Old Order Anabaptist communities in its catchment area for more than 20 years. These include a home visiting program and nurse practitioner-led Mother and Young Child Clinics, supported in part through the Canada Prenatal Nutrition Program (Government of Canada 2023; personal communication, NM). Concerns about the nutritional status, particularly vitamin D, of these Old Order Anabaptist communities have been expressed by these healthcare professionals.

Specific risk factors for vitamin D deficiency include prolonged breastfeeding and, for older children, consumption of farm milk that is not fortified with vitamin D, and low consumption of foods, such as fatty fish, with naturally occurring vitamin D (personal communication NM). Further, Old Order Anabaptist children wear clothing with long sleeves, long pants and dresses, thus limiting skin exposure to the sun (Perth District Health Unit, 2012). There was a case of rickets several years ago in the study community (personal communication, CS). Given the small size of the pediatric population (~150-200 births per year (personal communication CS)), the past occurrence of even one case of rickets was part of the rationale for conducting this study.

The biomarker most widely used to assess vitamin D status is serum 25 hydroxy (OH) vitamin D (25(OH)D), with the interpretation depending on the cut-off point used (Weiler, et al. 2023). The cut-off point for 25(OH)D associated with dietary intake of the Recommended Dietary Allowance (RDA) (that meets the needs of 97.5% of the population) is 50 nmol/L (Institute of Medicine 2011) whereas the suggested cut-off point from the Canadian Pediatric Society is 75 nmol/L (Godel, et al. 2007). Cut-off points of 30 nmol/L and 40 nmol/L are associated with risk of deficiency and population inadequacy, respectively (Institute of Medicine 2011). Concentrations of 25(OH)D >125 nmol/L may present increased risk of adverse effects such as retarded linear growth in infants (Institute of Medicine 2011).

Given our concerns about the vitamin D status of children in these Old Order Anabaptist communities in Southwestern Ontario, our primary objective was to assess vitamin D status based on blood spot total 25 hydroxy vitamin D (vitamin D2 + vitamin D3) (Blood Spot 25(OH)D) and dietary assessment. The relationship of vitamin D status to growth and other relevant factors was also explored.

METHODS

This was a descriptive cross-sectional study with a convenience sample. Ethics approval was from the Research Ethics Board (REB) at the University of Guelph (REB#17-07-022). The target population included children (0-6 years) of ~150 women who had given birth between January 2018 and January 2019 and who were clients of Countryside Midwifery Services or the Mother and Young Child Clinics in Perth County in Ontario, Canada. The mothers of the children in this study, who were invited to participate in person at the clinics, participated in a parallel study (Randall Simpson, et al. 2023). Mothers provided informed written consent on behalf of their children.

All data collection took place in participants' homes. Basic demographic information (age, gender, birth weight, general health information, season, breastfeeding status, supplement use, sun exposure, sunscreen use, etc.) was collected by means of a paper questionnaire based on one used previously in the OOA community (Randall Simpson, et al. 2004). The questionnaires were completed by the registered dietitian (RD) based on responses from the mothers; steps to ensure confidentiality, as described in an information letter (approved by the REB), were discussed with participants by the RD.

Finger prick blood samples were drawn by the project RD and deposited onto a blood spot test card. The dried blood samples were analyzed for vitamins D2 and D3 (Blood Spot 25(OH)D) by ZRT Laboratory in Beaverton, OR, USA (ZRT Laboratory 2008). The blood spot test method has been demonstrated to be highly correlated with serum 25(OH)D, considered to be the main marker for vitamin D status (Weiler, et al. 2023) in previous measurements (Larkin, et al. 2011; Newman, et al. 2009; ZRT Laboratory 2008).

Dietary intakes were determined based on three-day (two weekdays and one weekend day) estimated food records (including food plus supplements) that were completed by mothers on behalf of their children. Food records were entered into the ESHA Food Processor (version 11.6.441, 2018, Food Processor, Salem, OR, USA) program, based on the Canadian Nutrient File (Government of Canada 2015a), at the University of Guelph. Intake of dietary vitamin D was adjusted for intra-individual variation (Nusser, et al. 1996). Information on vitamin D content of supplements was determined from label information and online searches. Adjusted intake of vitamin D was compared to the Dietary Reference Intakes (DRIs) for each child, depending on the child's age and gender (Institute of Medicine 2000). The DRI reference values that were used were the Estimated Average Requirement (EAR) (value estimated to meet requirements of 50% of healthy people) and the Tolerable Upper Intake Limit (UL) (highest daily intake not likely to pose risk) (Institute of Medicine 2000). Use of eaTracker, available on the Dietitians of Canada website at the time (Dietitians of Canada 2015), was used to determine food group servings according to the 2007 Canada's Food Guide (CFG) (Bush, et al. 2007).

Growth measurements were taken by the RD. Weight, in light clothing and no shoes, was measured in triplicate using a calibrated scale (accurate to 50 g), with a removable weighing tray for younger children (Tanita Digital Baby Scale (1584), Arlington Heights, IL, USA). Height (to 0.1 cm) for children ≥ 24 months of age was measured in triplicate using a portable stadiometer (Road Rod (214), SECA, Chino, CA, USA). Length (to 0.1 cm) was measured in triplicate using a portable measuring mat for toddlers < 24 months of age (Starters Pediatric Measure Mat, Slater & Frith, Norwich, UK). Growth measurements for the children were assessed using the 2010 World Health Organization growth standards adapted for Canada (Dietitians of Canada 2014). Body Mass Index (BMI) and associated percentiles for children ≥ 2 years of age were calculated using the British Columbia Children's Hospitals Tools and Calculators (British Columbia 2023); classification of BMI into categories was based on the World Health Organization 2007 growth standards (de Onis, et al. 2007).

Data were entered (in duplicate) into EXCEL; data were analyzed in the Statistical Package for the Social Sciences (SPSS v. 28, IBM, Chicago, IL, USA). Student t-tests were used to assess differences in Blood Spot 25(OH)D concentrations. Multiple regression was used to predict Blood Spot 25(OH)D from total dietary intake of vitamin D, gender, and season. Categorical data are presented as percentages; continuous data are presented as mean \pm SD (standard deviation).

RESULTS

Demographic and other characteristics of the 52 children who were recruited for the study are shown in Table 1. The age range of the children was 2-80 months. Ninety-six % were breastfed as infants; only 7% were introduced to solid food before 5-6 months of age. Most children consumed farm milk (mostly cow, but some goat) that was neither fortified with vitamin D nor pasteurized. It was reported that 67% of all the children in the study received a vitamin D supplement when they were <1 year of age; this was a daily supplement for 50%. Two of the 4 mothers of children < 1 year of age in the study reported giving a vitamin D supplement at least 5-7 times/week. Eighty-nine percent of mothers of children \geq 1 year of age reported giving

TABLE 1: CHARACTERISTICS OF 52 OLD ORDER ANABAPTIST CHILDREN IN SOUTHWESTERN ONTARIO

Characteristic		Children (n)
Age	<12 months	04 (08%)
	1-2 years	10 (19%)
	2-3 years	17 (33%)
	>3 years	21 (40%)
Gender	Male	23 (44%)
	Female	29 (56%)
Type of Milk	Farm Milk	46 (88%)
	Grocery Milk	06 (12%)
Use of Sunscreen	Rarely or never	42 (81%)
	Sometimes	10 (19%)
Type of Buggy	Open	32 (62%)
	Closed	20 (38%)
Time Outdoors	\geq 3 hours/day	39 (75%)
	< 3 hours/day	13 (25%)

their children a vitamin D supplement. Of children receiving a vitamin D supplement, the frequency of use was daily for 47%, 5-6 times a week for 20%, and 3-4 times a week for 20%. Sixty-four % of children were given additional vitamin/mineral supplements.

Blood Spot 25(OH)D concentrations were available for all 52 children. Mean Blood Spot

TABLE 2: CATEGORIZATION OF VITAMIN D STATUS FOR 52 OLD ORDER ANABAPTIST CHILDREN

Marker ^a	Cut-off points ^b	Number of Children (%)				
		0-6 mo ^c (n=3)	7-12 mo (n=1)	1-3 y (n=27)	4-6 y (n=21)	Total ^c (n=52)
Blood Spot 25-Hydroxy vitamin D	< 30 nmol/L	0	0	0	0	00 (00%)
	30-39 nmol/L	0	0	1	1	02 (04%)
	40 - 49 nmol/L	0	0	1	4	05 (10%)
	50-74 nmol/L	1	0	12	6	19 (37%)
	75 - 125 nmol/L	1	1	10	9	21 (40%)
	>125 nmol/L	1	0	3	1	05 (10%)

^a Serum/plasma 25 hydroxy vitamin D (25(OH)D) is the main marker for assessment of vitamin D status. In this study, total vitamin D (vitamins D2 and D3) were measured by blood spot vs serum; the blood spot test has been demonstrated to be highly correlated with serum 25(OH)D.

^b Cut-off points are based on: Institute of Medicine (2011) and Godel, et al. (2007).

^c mo = months; y = years; percentages may not add up to 100 due to rounding

TABLE 3. DIETARY INTAKE OF VITAMIN D FOR 50 OLD ORDER ANABAPTIST CHILDREN

Age Category	Vitamin D Intake from Food (IU/d) (mean ± SD)	Vitamin D Intake from Supplements (IU/d) (mean ± SD)	Vitamin D Intake from Food and Supplements (IU/d) (mean ± SD)
< 12 months	24 ± 36 (n=4)	133 ± 154 (n=4)	158 ± 153 (n=4)
1 to 3 years	62 ± 55 (n=25)	116 ± 167 (n=18)	178 ± 184 (n=25)
4 to 6 years	95 ± 70 (n=21)	164 ± 209 (n=15)	266 ± 210 (n=21)
All 1 to 6 year olds	68 ± 39 (n=46)	137 ± 182 (n=33)	213 ± 194 (n=46)

Note: IU/d = International Units per day; SD = standard deviation.

TABLE 4. DRI COMPARISON FOR DIETARY VITAMIN D INTAKE FOR 46 OLD ORDER ANABAPTIST CHILDREN

Vitamin D	1 to 3 y (n=25)		4 to 6 y (n=21)		Total (n=46)	
	Food Only	Food & Supplements	Food Only	Food & Supplements	Food Only	Food & Supplements
# < EAR	25	22	21	15	46 (100%)	37 (80%)
# > UL	0	0	0	0	0	0

Note: y = years; DRI = Dietary Reference Intake; EAR = Estimated Average Requirement; UL = Tolerable Upper Intake Limit

Note: No children < 12 months had intakes > UL

25(OH)D was significantly higher for female children (82±33) compared to male children (74±30) ($F=5.534$, $t=0.942$, $df 50$, $p=0.023$); similar results for blood 25(OH)D were reported in a nationally representative Canadian sample (Weiler, et al. 2023). There was no significant difference by season (classified into two categories, fall/winter and spring/summer); Weiler et al. (2023) reported lower vitamin D status in the winter months. Results for categorization of blood 25(OH)D by various cut-off points are shown in Table 2.

Food records were available for 50 children. Results for intake of vitamin D from food and supplements are shown in Table 3. Comparison of dietary intakes with the Estimated Average Requirement (EAR) is suitable for children ≥ 12 months (Institute of Medicine, 2000). Results of the comparison of dietary intakes with the DRIs for 46 children are presented in Table 4. DRI age

categories are for children 1 to 3 years and 4 to 8 years of age (Institute of Medicine, 2000).

Comparison of food group intakes with 2007 Canada's Food Guide is suitable for children ≥ 2 years (Bush, et al. 2007); results of the comparison of food group intakes with Canada's Food Guide for 36 children ≥ 2 years of age are presented in Table 5.

Growth measurements were available for 48 children. Categorization of BMI status is suitable for children ≥ 2 years of age (Dietitians of Canada 2014) with data available for 37 children: 2 children were classified as underweight; 26 were classified as healthy BMI; 5 were classified as overweight; and 4 were classified as obese using the World Health Organization (2007) criteria (de Onis, et al. 2007). There was no significant difference in blood vitamin D between BMI classified into two categories (underweight + normal [$n=28$] vs overweight + obese [$n=9$]), unlike findings of

TABLE 5: FOOD GROUP INTAKE FOR 36 OLD ORDER ANABAPTIST CHILDREN

	Grains	Fruits and Vegetables	Milk and Alternates	Meat and Alternates
CFG Recommendations for age 2 to 3 years	3	4	2	1
Food Group Intake for ages 2 to 3 years (n=15)	2.6 ± 1.1	3.7 ± 1.7	1.5 ± 0.6	1.2 ± 0.3
Number < CFG Recommendations	10 (67%)	9 (60%)	12 (80%)	2 (13%)
CFG Recommendations for ages 4 to 8 years	4	5	2	1
Food Group Intake for ages 4 to 6 years (n=21)	3.1 ± 1.1	4.2 ± 1.6	1.8 ± 0.9	1.6 ± 0.6
Number < CFG recommendations	17 (81%)	16 (76%)	11 (52%)	5 (24%)
Total < CFG recommendations	27 (75%)	25 (69%)	23 (64%)	7 (19%)

Note: CFG = Canada's Food Guide (Bush, et al. 2007)

TABLE 6: FOOD GROUP INTAKE FOR 36 OLD ORDER ANABAPTIST CHILDREN

Blood 25(OH)D	B	95% CI for B		SE B	β	R ²	ΔR^2
		LL	UL				
Model						.358	.316*
Constant	48.746*	24.878	72.614	11.858			
Vitamin D Intake	.084*	.047	.121	.018	.548*		
Gender	6.501	-7.761	20.764	7.086	.109		
Season	9.518	-5.642	24.678	7.531	.150		

Note: Model = "Enter" method in SPSS Statistics' B = unstandardized regression coefficient; CI = confidence interval; LL = lower limit; UL = upper limit; SE B = standard error of the coefficient; β = standardized coefficient; R² = coefficient of determination; ΔR^2 = adjusted R²

*= p value < 0.001

other researchers (Greene-Finestone, et al. 2017; Weiler, et al. 2023).

The multiple regression model statistically significantly predicted Blood Spot 25(OH)D, $F(3, 36) = 8.546$, $p < .001$, adj. $R^2 = .358$. Only total vitamin D intake, but not gender or season, added significantly to the model. Regression coefficients and standard errors can be found in Table 6.

DISCUSSION

Results of this study indicate that vitamin D status of children in these Old Order Anabaptist communities in Southwestern Ontario, while generally good, still presents some challenges despite over two decades of public health initiatives.

The most widely used biochemical marker to assess vitamin D status is blood/plasma/serum 25(OH)D (Institute of Medicine 2011; Weiler, et al. 2023) whereas we used Blood Spot 25(OH)

D as our marker. Comparisons of our results for mean concentrations of Blood Spot 25(OH)D with other studies (using blood/serum/plasma 25(OH)D) are shown in Table 7.

The use of different 25(OH)D cut-offs for determining vitamin D status is challenging for comparing data from both large national and smaller studies. There were no children in this study who were at risk of deficiency (<30 nmol/L); 4% had inadequate status (<40 nmol/L). As shown in Table 7, these proportions were slightly lower than national Canadian data from the Canadian Health Measures Survey (CHMS) 2012-2019 (Weiler, et al. 2023). However, 5 children (10%) in our study had blood 25(OH)D >125 nmol/L, considerably higher than the 1.1% reported from the CHMS (Weiler, et al. 2023). Vitamin D toxicity would be considered at concentrations of 25(OH)D of 250-375 nmol/L (Vogiatzi, et al. 2014), well above the highest value of 152 nmol/L found in this study.

TABLE 7: COMPARISON OF RESULTS FOR 25 HYDROXY VITAMIN D FOR 52 OLD ORDER ANABAPTIST CHILDREN WITH OTHER STUDIES

Study	Location	Children		Dietary vitamin D intake (IU)	Blood/Blood Spot 25 Hydroxy Vitamin D (nmol/l)			
		Sample size	Age (years)		<30 (%)	<40 (%)	<50 (%)	<75 (%)
Randall Simpson, et al. 2023 (current study)	Ontario, Canada	82	0-6	79 ± 30	0	4	14	50
National Data								
Kumar, et al. 2009	U.S.A.	6275	1-21	-	-	-	9	61
Langlois, et al. 2010	Canada	903	6-11	75	-	4	-	52
Greene-Finestone, et al. 2017	Canada	3381	6-17	-	6	15	30	-
Herrick, et al. 2019	U.S.A.	1438	1-5	-	0.5	7	-	-
Weiler, et al. 2023	Canada	3525	3-8	-	2.1	7.4	-	-
Canadian Studies								
Maguire, et al. 2011	Toronto	91	2	-	-	-	32	82
El Hayek, et al. 2013	Montreal	508	2-5	74	0.6	10	12	51
Gallo, et al. 2016	Montreal	87	3	74	0	1	10	62
Brett, et al. 2016	Montreal	24	2-8	59	4	4	29	-
Brooks, et al. 2022	Ottawa	57	6-19	58	-	9	-	-

^a Blood vitamin D status is based on Blood Spot 25(OH)D for this study and on serum/plasma 25(OH)D for the other studies.

Until the most recent report on CHMS data, there were no national Canadian data for children < 6 years of age (El Hayek, et al. 2013; Greene Finestone, et al. 2017; Weiler, et al. 2023). To address this, a unique 2013 study conducted in Montreal, QC, Canada assessed the vitamin D status of 508 preschoolers (ages 2-5 years) (El Hayek, et al. 2013). Our results are similar to this large Montreal sample (Table 7). There are other reports of vitamin D status in young Canadian children from smaller studies with which to compare our results as shown in Table 7.

Another indicator of vitamin D status is dietary intake from food and supplements. Total dietary intake of vitamin D was significantly and positively associated with Blood Spot 25(OH)D. Other researchers have found associations between 25(OH)D and dietary vitamin D intake (Brett, et al. 2016; Herrick, et al. 2019; Larkin et al. 2011; Mortensen, et al. 2016). Relationships between milk intake and blood vitamin D status

have also been reported (Langlois, et al. 2010; Maguire, et al. 2011).

Dietary intakes of vitamin D were very low, with 100% of OOA children < EAR for food only and 80% < EAR considering both food and supplements. This is not surprising, given that more than 80% of Anabaptist families in the study communities reported consuming unfortified farm milk (cow, goat, sheep). The primary source of vitamin D in Canada is fortified fluid cow milk (Janz and Pearson 2013) given mandatory vitamin D fortification, in part to protect against rickets (Janz and Pearson 2013).

Our results for comparison to the DRIs are like other Canadian data for very young children. For example, of 77 children aged 2-8 years in Montreal, none met the EAR at baseline from food alone (Brett, et al. 2016). For 508 preschoolers and for 25 3-year-old children in Montreal, just 5% met the EAR from food alone (El Hayek, et al. 2016; Gallo, et al. 2016).

Even though the proportion of children with vitamin D intakes < EAR from food alone is similar between our study and the Montreal studies, the amount of vitamin D from food alone was considerably lower for children in our study. For example, the dietary intake of vitamin D in the Old Order Anabaptist children (1-6 years) was 68 IU/day compared to 236 IU/day for the large cohort of Montreal preschoolers and 247 IU/day and 198 IU/day in the Gallo, et al. (2016) and Brett et al. (2016) studies, respectively. Other Canadian researchers have reported similar results (El Hayek, Egeland, and Weiler, 2010; Roth, et al. 2005). The lower amounts of vitamin D from food in our study for very young children is most likely attributable to the lack of vitamin D fortification of farm milk.

Even though vitamin D intake was much lower for children in this study compared to other studies, the vitamin D status was not remarkably different. This suggests that parents in the OOA communities may be more adherent to supplementation regimes than other Canadian parents. In the large Montreal study, just 28% of parents reported that their children took a vitamin D supplement compared to 89% in this study. Based on the 2015 Canadian Community Health Survey, 45% of Canadian children ages 1-8 years took vitamin/mineral supplements (Government of Canada 2015). Most mothers (90%) in this study who reported giving their children a vitamin D supplement did so at least 3-4 times per week with 50% giving a vitamin D supplement daily. Further, a large proportion of mothers in our study (64%) reported giving other supplements, that may have included additional vitamin D, in addition to vitamin D supplements. This may account for the fact that 10% of children in this study had Blood Spot 25(OH)D concentrations higher than 125 nmol/L, the level at which there is a risk of adverse effects (Institute of Medicine 2011). However, no children had dietary intakes, during the 3 days of data collection, of vitamin D greater than the Upper Limit.

The finding that 64% of children in this study had servings of milk and alternatives less than the recommended number of servings in the 2007 CFG (Bush, et al. 2007) and that the mean number of servings was less than the recommendation for their age groups is concerning with regards to bone health (Bush, et al. 2007). For children in other Canadian studies on vitamin D status, the number

of servings of milk and alternatives were more than recommended (Brett, et al. 2016; El Hayek, et al. 2013). Low intake of milk and alternates also affects calcium status. Indeed, more than a third of children in this study had calcium intakes <EAR (data not shown), a higher proportion than for Montreal pre-schoolers (4.8% for <3.9 y and 25.9% for > 4 y) (El Hayek, et al. 2013).

Another factor that may have contributed to the generally good vitamin D status for these Old Order Anabaptist children could be that daily time outdoors was considerably more (75% with ≥ 3 hours/day) than the ~ 30 min/day, 3 times/week that has been reported to maintain vitamin D status (Holick 2007; Raman 2023).

Despite our findings that the status of vitamin D in this sample of Old Order Anabaptist children was comparable to other young Canadian children, our findings should not lead to complacency surrounding vitamin D status and, in particular, the role of vitamin D supplementation. It is not likely that families in these communities would purchase vitamin D fortified milk as it is not part of their traditional diet and also due to financial constraints, travel and lack of refrigeration. While Health Canada has plans to increase the level of vitamin D fortification in milk and to supplement other milk products with vitamin D (Government of Canada, 2021), this will have little effect on vitamin D intakes in the Old Order Anabaptist communities for the aforementioned reasons.

Adherence to daily supplementation regimens for vitamin D is important in these Old Order Anabaptist communities. While reported adherence in this study is higher than that reported by other researchers, there is still room for improvement. Just half of our study participants reported supplementing their children with vitamin D daily. Adherence to vitamin D supplementation recommendations for infants is reportedly higher in Canada (Crocker, et al. 2011; Loyal and Cameron 2020) than in the US, likely due to a heightened awareness of the importance of vitamin D supplementation in Canada due to lower cutaneous production of vitamin D in northern latitudes (Loyal and Cameron 2020). Adherence to recommendations for vitamin D supplementation in US infants 0-11 months from the 2009-2016 NHANES has recently been addressed by Simon and Ahrens (2020). For all children, fewer than 40% of infants in all demographic subgroups met the vitamin D

intake guidelines and there was no difference between data collected in 2009-2010 and 2015-2016 (Simon and Ahrens 2020). Research in the UK on adherence to messaging on vitamin D health in young children found that parents felt they need clear, simple, easy to read information on the importance of vitamin D that is delivered regularly (Day, et al. 2019).

The healthcare professionals working in these OOA communities promote the importance of vitamin D supplementation for breast fed infants and children over the age of one. The organizations involved also reduce barriers to supplementation through the distribution of vitamin D drops at no cost, a practice that may enhance uptake of supplementation in these Old Order communities. Health professionals working with these communities should counsel clients on the importance of daily supplementation and offer strategies to improve consistency, which mothers often cite as a challenge in busy Old Order households. Examples of strategies that have been helpful to some families with consistency in use of supplements include keeping the supplement bottles in a visible location (out of reach of small children), habit stacking (pairing the desired behaviour with another well-established daily habit) or enlisting the help of older siblings to provide daily reminders (personal communication with co-author Natalee Miller [NM]). Anecdotally, some Old Order Anabaptist families take supplements only in the fall and winter months, including months that contain the letter 'r' (personal communication with NM). Given the low vitamin D intakes from food, it is warranted for professionals working with these communities to continue to promote year-round daily supplementation in order to ensure continuing adequate status of Vitamin D.

Vitamin D status is not regularly monitored as a routine part of well-child visits and 25 (OH) D testing is not covered by provincial health insurance plans. Although widespread deficiency was not observed in this study, it is reasonable for healthcare practitioners to continue to recommend daily supplementation at the level of the RDA to help fill gaps in vitamin D intake from diet with the goal of maintaining normal serum vitamin D levels. Data on safety of high vitamin D intake are limited (Vogiatzi, et al. 2014). While excess vitamin D supplementation is rare, it is important to ensure that infants and young children do not

exceed the UL; if this is the case, monitoring of serum 25(OH)D is recommended (Vogiatzi, et al. 2014).

Women in these Old Order Anabaptist communities are receptive to taking dietary supplements; for example, we have reported high intake and high blood concentrations of folic acid and vitamin B12 in the mothers of the children in this study (Randall Simpson, et al. 2023). Practitioners, including nurses, dietitians, and nurse practitioners have observed frequent herbal and vitamin/mineral supplement use among clients in these groups (personal communication, NM). To our knowledge, there are no published data on the usage of natural health products in Canadian Old Order communities; researchers in the US reported that 77% of a sample of 2372 Old Order Amish adults reported supplement use (Reed, et al. 2015).

Also, of importance for public health professionals working with Old Order Anabaptists is ensuring they use supplements in appropriate amounts. The advice received should include vitamin D intakes at both ends of the spectrum. It is clear from this study, that, while most OOA children receive appropriate vitamin D supplementation, over supplementation should be addressed.

CONCLUSION

To the best of our knowledge, this study is unique in this population of children where low intakes of vitamin D are expected. This study provides evidence for ongoing public health education in the community. A strength of this study included three-day dietary records (checked 3 times by research assistants) that enabled the use of the technique to adjust intakes for intra individual variation (Nusser, et al. 1996) thereby allowing for comparison to the DRIs. Limitations include a relatively small sample size with a fairly large range of ages of the children; this limited the strength of the regression analysis. While the most recognized biochemical marker of vitamin D status is serum 25(OH)D, our study relied on finger prick blood samples necessitating the use of a dried blood spot analysis that has been validated against the more common serum analysis (Larkin, et al. 2011; Newman, et al. 2009; ZRT Laboratory 2008).

This research is relevant to the OOA involved. Results have been shared within the community by

written communication via newsletters, by health-care professionals, and with the community leaders; recommendations for appropriate vitamin D supplementation are already in place. While these results may not be generalizable to the general population, they will be of interest to public health professionals serving other Old Order Anabaptist populations in both Canada and the US where milk that is not fortified with vitamin D is consumed. It is estimated that there were approximately 10,000 horse and buggy Anabaptists in Ontario in 2022 (Daynard 2022).

In conclusion, in this sample of 52 Old Order Anabaptist children from Southwestern Ontario, most had adequate vitamin D status. Of concern is the number of children with relatively high values of 25(OH)D suggesting over supplementation. These findings support the need to encourage adequate dietary intake of Vitamin D and complementary appropriate supplementation to consistently achieve the RDA of 600 IU/day, but not over supplement, for Old Order Anabaptist children.

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