ORIGINAL RESEARCH ARTICLE

COACTION PUBLISHING

Higher body mass, older age and higher monounsaturated fatty acids intake reflect better quantitative ultrasound parameters in Inuit preschoolers

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Objectives. Investigate the effects of selected factors associated with quantitative ultrasound parameters among Inuit preschoolers living in Arctic communities (56° 32′–72° 40′N).

Materials and methods. Children were selected randomly in summer and early fall (n = 296). Dietary intake was assessed through the administration of a 24-h dietary recall (24-h recall) and a food frequency questionnaire (FFQ). Anthropometry was measured using standardized procedures. Plasma 25-hydroxy vitamin D (25(OH)D) and parathyroid hormone (PTH) were measured using a chemiluminescent assay (Liaison, Diasorin). Quantitative ultrasound parameters were measured using Sahara Sonometer, (Hologic Inc.).

Results. Children divided by speed of sound (SoS) and broadband ultrasound attenuation (BUA) quartiles were not different for age (years), sex (M/F), calcium (mg/d) and vitamin D intake (μg/d) and plasma 25(OH)D concentration (nmol/L). However, children in the highest BUA and SoS quartile had higher body mass index (BMI) compared to those in quartile 1. Using multivariate linear regression, higher BMI, older age and monounsaturated fatty acids (MUFA) intake were predictors of BUA while only BMI was a predictor of SoS.

Conclusions. Further investigation assessing intakes of traditional foods (TF) and nutrients affecting bone parameters along with assessment of vitamin D status of Inuit children across seasons is required.

Keywords: *speed of sound*; *broadband ultrasound attenuation*; *preschoolers*; *Inuit*; *vitamin D*

Received: 21 December 2011; Revised: 18 April 2012; Accepted: 25 May 2012; Published: 9 July 2012

Reaching the highest peak bone mass within one's genetic potential, or reducing the rate of bone loss are the main strategies for reducing the risk of osteoporosis (1). Although 60–80% of peak bone mass is determined by genetics (2), nutrition and lifestyle factors of relevance to children and development of bone mass include, body weight and composition, physical activity and nutrition (2,3).

Ethnicity is also known to have a strong impact on bone health (4–6). In adults, those who are black have higher bone mineral density (BMD) compared to Caucasians (6) and Caucasians have higher BMD than Asians (7). These differences are also apparent in newborns (8) and continue later in childhood (9). On the other hand, Aboriginal American women have higher rates of bone loss compared to their white counterparts (5), while Aboriginal Canadian women have lower BMD *z*-scores compared to their white counterparts (4). However, data from Aboriginal newborns did not show lower whole body or regional bone mineral content (10), suggesting that disparities in the postnatal environment underlies the differences observed in adults.

Several dietary and lifestyle factors may predispose Aboriginal and Inuit people to low BMD, beginning as early as childhood. Traditional food (TF) is rich in nutrients that would support bone health, including protein, polyunsaturated fatty acids (PUFA), zinc, and vitamins A, D, and C (11,12). Instead of complementing the diet with market food (MF) rich in nutrients including calcium, phosphorus, and vitamin K (3,13) that would optimize bone health, Inuit children are shifting from a nutrient dense diet to an energy dense diet, characterized by low quality MF consumption (12,14). Further, infrequent use of dietary supplements (14) and polar latitude

Citation: Int J Circumpolar Health 2012, **71**: 18999 - http://dx.doi.org/10.3402/ijch.v71i0.18999

that limits endogenous vitamin D synthesis (15) may also play a role in predisposing Inuit children to poor bone health. The quantitative ultrasound technique is being used among the paediatric population and its parameters, particularly broadband ultrasound attenuation (BUA) and speed of sound (SoS), correlate well with BMD and with bone quality measures (16).

Therefore, the primary objective of this study was to investigate whether anthropometric, dietary, biochemical and lifestyle risk factors are associated with bone health, as reflected by BUA and SoS among Inuit preschoolers (3–5 years) living in Arctic communities (56° 32′–72° 40′N). We hypothesize, based on the literature, that vitamin D and calcium intake, body mass index (BMI), and plasma 25-hydroxy vitamin D (25(OH)D) would be important predictors of SoS and BUA.

Materials and methods

Subjects

The sample consisted of Inuit preschool children recruited in the late summer and early fall of 2007 (August–November) and 2008 (August–September) in 16 of the 25 communities of Nunavut, representing all 3 regions of the territory (Kivalliq, Baffin, and Kitikmeot). Communities were selected based upon logistical and cost consideration and upon achieving representation of latitude, region, and community size. Inclusion criteria for participation consisted of self-identified Inuk by parents or a caregiver and 3–5 years of age. Children were recruited from both the community health centre lists of age-appropriate children and from randomly selected households with 3–5 year-olds that had participated in the International Polar Year Inuit Adult Health Survey. A randomized list of children was created from the health centre list using a random number table and parents/caregivers were contacted in the order that they appeared on the list. Caregivers were contacted either by phone, when available, or in person. Of the 537 successfully contacted households, 75 refused upon initial contact and 74 cancelled or failed to attend the study appointment; thus, the overall participation rate was 72.3% and 388 children were recruited. A more detailed description of the survey methodology is available elsewhere (17).

Ethics

The study was approved by the McGill Faculty of Medicine Institutional Review Board and by the Nunavut Research Institute. A parent or primary caregiver provided signed informed consent. A person was considered a child's primary caregiver if he/she was the person primarily responsible for the child at the time of the study. Consent forms and an information DVD were available in English and Inuit languages.

Research team and interviews

The research team consisted of bilingual interviewers who conducted face-to-face interviews. Information about household composition, living conditions, diet, supplement use, and health status were collected through interviews with the child's caregiver. Information about sun exposure was collected. Caregivers were asked an open ending question "on average, about how many hours per day, does your child play outside?"

A qualitative food frequency questionnaire (FFQ) (without assessment of portion sizes) was completed by the child's caregiver. The FFQ reflected the previous month and contained 30 commonly consumed TF items, some of which are considered good sources of vitamin D, (whitefish, arctic char, seal meat, seal liver, caribou, caribou liver, polar bear meat and walrus meat). It also contained the following MF sources of vitamin D: milk, margarine, and eggs. Furthermore, 24-hour dietary recalls (24-h recalls) were conducted with the caregiver using a multiple pass technique. Portion sizes were estimated using a 3-dimensional food model kit (Santé Quebec) to better standardize 24-h recalls. Twenty percent of the caregivers were asked to return for a repeat 24-h recall, which was conducted on a non-consecutive day to allow assessment of nutrient adequacy. Interviewers recorded if there were times when the caregiver did not know what the child ate. Daycares were called regarding daycare snacks and meals to complete dietary recall. This approach has been shown to be an accurate method of assessing intake in this age group (18). Vitamin D and calcium intakes were adjusted for the second 24-h recall and an estimation of the adjusted intake for sequence and day of week was calculated using the Iowa State Software for Intake Distribution Estimate (Iowa State University, 1996). The adjusted data were only used to compare intakes against the DRIs. Since data collection for this study was performed before the release of the 2011 guidelines of the Institute of Medicine, accordingly, vitamin D and calcium intake of participants were compared to the both the adequate intake (AI) (19) and the estimated average requirement (EAR) (20). Vitamin and/or mineral supplement use and frequency were recorded, however, due to low prevalence of supplement intake, vitamin D or calcium intake reflected only dietary intake.

Clinical assessments

Venous blood (3 mL) was collected into heparin coated vacutainers followed by centrifugation and storage of plasma at −20°C and was then transported on ice packs in coolers to McGill University and stored at −80°C until analysis. Height was measured to the nearest 0.1 cm using a portable stadiometer (Road

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Rod214 Portable Stadiometer, Seca) and weight was measured to the nearest 0.1 kg using an electronic scale. BMI-for-age *z*-score (BAZ), height-for-age *z*-score (HAZ) and weight-for-age *z*-score (WAZ) were calculated and interpreted using the World health organization (WHO) Child Growth Charts for children (21). The non-dominant calcaneus was measured using ultrasound Sahara Sonometer (Hologic, Bedford, MA, USA). Measurement of the os calcis consisted of BUA (expressed in decibels per megahertz, Δdb/ΔMHz) and SoS (expressed in meters per second, m/s). The quantitative ultrasound device measures BUA and SoS and the results are combined to estimate BMD. Ultrasound bone densitometry was shown to be sensitive enough to detect physiological bone development in childhood (22) and compromised bone health in diseased children (16). At the start of each clinical day, the sonometer was quality checked according to the manufacturer's instructions. Briefly, the machine was checked using a control bone also known as a "phantom". These assessments were recorded in a quality control log and tracked to ensure the machine was working consistently.

Laboratory analysis

Plasma Alkaline Phosphatase (ALP) and total calcium were measured by Beckman Coulter D × C 800 (Beckman Coulter Inc., Mississauga, Ontario, Canada) at the Royal Victoria Hospital. The Laboratory participated in external validation and received a proficiency certificate for 2009–2010 from DigitalPT. High sensitivity C-reactive protein (CRP) was measured in the serum using Enzyme Linked Immunosorbance Assay. Plasma 25(OH)D and parathyroid hormone (PTH) concentrations were measured using LIAISON total 25(OH)D and N-tact PTH assays (DiaSorin Inc., Stillwater, MN, USA) at McGill University. The inter-assay and the intra-assay coefficient of variation (CV) were 4.5 and 11.1% for the low 25(OH)D control (38.2 nmol/L) and 6.2 and 5.3% for the high 25(OH)D control (127.2 nmol/L); the accuracy using the mid-range of the manufacturer's specifications was 95%. For the PTH low control, the inter-assay CV% was 19.1 (5.2 pmol/L) and 8.7 for the high PTH control (52.1 pmol/L). The accuracy using the mid-range of manufacture specifications was 86.7%. The laboratory (HW) that measured 25(OH)D participated in the Vitamin D External Quality Assessment Scheme program and obtained a certificate of proficiency for 2009–2010, which reflects that ≥80% of the reported results fell within 30% of the All-Laboratory Trimmed Mean.

Statistical analysis

Questionnaires and clinical information were entered into a Microsoft Access Database and 24-h dietary recall data were entered using CANDAT (Godin London). For outliers, all dietary variables exceeding mean + 3 standard deviations (SD) were substituted with mean + 3 SD. For all other variables, values exceeding mean + 3 SD were excluded. All variables were tested for normality prior to statistical analysis using Shapiro–Wilk test. Since variables were skewed, spearman correlations were used to identify correlations between BUA or SoS and different continuous characteristics. ANOVA, followed by Bonferroni post-hoc tests whenever appropriate, were used to determine whether dietary, anthropometric, biochemical and lifestyle factors were different by BUA or SoS quartiles when variables were continuous and Chi-square was used when variables were categorical. Prior to ANOVA, all skewed variables were log transformed or square rooted. Multivariable linear regression was used to identify predictors of SoS or BUA. SoS and BUA measurements were available for 285 children and full multivariable analysis was possible for 211 children, because data on food intake, accurate anthropometry measures, the number of hours spent outside or 25(OH)D concentration were missing. Various models were constructed. Normality of residuals was ensured using the Kolmogorov–Smirnov test along with visual examination of residual-normal quantile plot, and heteroskedasticity was evaluated using the Breusch–Pagan/Cook–Weisberg test. Variance inflation factors were examined post-regression to ensure the absence of multicollinearity. For all tests, $p < 0.05$ was considered significant. Values in the text are percent (95% CI), means ± SD, or medians with interquartile ranges (IQR). All statistical analyses were completed using Stata 10 (Stata Corp).

Results

Although 388 children participated in the health survey, not all children had BMD measures. Children with available SoS and BUA measures (n = 285) were similar to those with unavailable values (n = 103) for age, gender, BMI percentile, number of hours spent outside, amount of TF consumed the previous day, milk intake, calcium intake, vitamin D intake, carbohydrates intake, protein intake, fat intake, PUFA intake, monounsaturated fatty acids (MUFA) intake, saturated fatty acids (SFA) intake, plus concentrations of CRP, ALP, total calcium, PTH, and 25(OH)D. Thus, results are presented for only the 285 children with available BMD measures (Table I).

The average latitude of communities participating in the study was 65.2 [95% CI: 64.8–65.6°N]. Median plasma total calcium, ALP, CRP, and PTH concentrations of Inuit preschoolers fell in the normal range. However, median 25(OH)D concentration was below 50 nmol/L (Table I) with 57.5% [95% CI: 50.9–64.1%] of the children below this cut off and 82.2% were below 75 nmol/L. Of Inuit preschoolers, 60.9% met the EAR of 800 mg/d for calcium while 10.5% met the EAR of 10 μg/d for vitamin D.

Since correlation coefficients were similar between boys and girls, all participants were analyzed together.

Table I. Selected characteristics of Inuit preschool children: Nunavut Inuit Child Health Survey, 2007–2008[a]

Characteristics	n	
Demographics		
Age (years)	285	4.4 ± 0.8
Boys (%)	132	46.3
Anthropometry		
BMI (kg/m^2)	284	18.2 [17.1–19.3]
BMI percentile (%)[b]	284	96.8 [88.1–99.3]
BMI-for-age *z*-score	284	1.85 [1.18–2.48]
Weight-for-age *z*-score	284	1.03 [0.43–1.72]
Height-for-age *z*-score	285	−0.39 [−0.93 to 0.25]
BUA (Δdb/ΔMHz)	285	40.6 [36.2–47.8]
SoS (m/s)	285	1541.7 [1532.7–1549.1]
Dietary intake		
Energy (Kcal/d)	276	1762 [1336–2234]
Carbohydrate (g/d)	276	239.6 [186.1–311.3]
Protein (g/d)	276	66.2 [45.6–90.9]
Fat (g/d)	276	54.3 [38.6–75.8]
SFA (g/d)	276	18.6 [12.0–25.5]
MUFA (g/d)	276	19.6 [12.8–28.8]
PUFA (g/d)	276	7.8 [5.0–12.2]
Vitamin D (μg/d)	276	4.9 [2.7–8.1]
Calcium (mg/d)	276	812 [522–1111]
Supplemental intake		
Vitamin D (% yes)	13	4.6
Multivitamins including vitamin D (% yes)	47	16.8
Biochemistry		
25(OH)D (nmol/L)	219	46.7 [30.2–68.9]
PTH (pmol/L)	205	2.33 [1.68–3.29]
Calcium (mmol/L)	168	2.34 [2.29–2.39][c]
ALP (U/L)	169	183.0 [160.0–207.5][d]
CRP (ng/mL)	222	0.7 [0.2–2.3][e]

[a]Values are percent, mean ± SD, or median [IQR].
[b]According to World Health Organization (23).
[c]Normal range: [2.0–2.4] mmol/L.
[d]Normal range: [111–356] U/L.
[e]CRP values ≥8 ng/mL indicating active infection (40) were excluded (n = 8).

Anthropometric variables, including WAZ, HAZ and BAZ correlated significantly ($p < 0.01$) with SoS ($r^2 = 0.21$, 0.19 and 0.19, respectively). Anthropometric variables, including WAZ, HAZ and BAZ correlated significantly ($p < 0.01$) with BUA ($r^2 = 0.39$, 0.28 and 0.32, respectively). None of the dietary variables, including energy (Kcal/d), carbohydrates (g/d), fat (g/d), SFA (g/d), MUFA (g/d), PUFA (g/d), vitamin D (μg/d) and calcium (mg/d) correlated significantly with either BUA or SoS, except protein (g/d). Protein intake correlated positively with SoS ($r^2 = 0.12$, $p = 0.03$). None of the food groups, including milk intake (mL/d), dairy products including milk (servings/d), fruits and vegetables (servings/d), TF (g/d), frequency of fish consumption (times/month) and frequency of marine mammals consumption (times/month) correlated significantly with BUA or SoS.

None of the biochemical variables, including 25(OH)D (nmol/L), PTH (pmol/L), calcium (mmol/L) or CRP (mg/L), correlated with SoS and BUA, except ALP (U/L). ALP correlated positively with SoS ($r^2 = 0.17$, $p = 0.02$). From the lifestyle variables, latitude inversely correlated with SoS ($r^2 = -0.11$, $p = 0.04$) while it did not correlate with BUA and the number of hours spent outside (h/d) did not correlate with either variables.

Children in the SoS and BUA quartiles were not different by age (years), sex (M/F), presence of an active hunter in the household (yes/no), carbohydrates intake, milk intake, fruits and vegetables intake, TF intake, fish and marine mammals, plasma CRP, calcium, PTH and latitude. However, children in the highest quartile of SoS or BUA had higher BMI and BAZ compared to those in quartiles 1 (Tables II and III). Also, children in SoS quartile 4 had higher ALP compared to those in quartile 3, while results were not different by BUA quartiles.

Using multivariate linear regression, higher BMI and MUFA intake plus older age were the only positive predictors of BUA in Inuit preschool children (Table IV), while age, sex, number of hours spent outside, vitamin D status ($0 < 75$ and $1 \geq 75$ nmol/L), energy, saturated fat, vitamin D, protein and calcium intake did not contribute to the model. On the other hand, only higher BMI was a significant predictor of SoS in Inuit preschoolers ($r^2 = 0.19$, $p < 0.01$) (Table V).

Discussion

This is the first study to report upon parameters of bone health in Inuit children. Children in the highest SoS and BUA quartile had higher adiposity, as measured using BMI than those in quartile 1. The strongest predictors of BUA were higher BMI, older age and higher MUFA while only higher BMI was a predictor of SoS.

Previous studies suggest Aboriginal children as a group are at a higher risk for low bone mass compared to other Canadian infants as a function of low vitamin D status (23). It appears from this sample that median vitamin D concentration of Inuit preschoolers was below the 50 nmol/L cut-off recommended by the Institute of Medicine (20). In contrast to the suggested hypothesis, vitamin D intake and plasma 25(OH)D concentration, regardless of the definition of optimal concentration (≥ 50 or 75 nmol/L) in the regression model, were not associated with SoS and BUA in our cross-sectional analysis. Very few children (18%) had 25(OH)D values in the range suggested to enhance bone health (≥ 75 nmol/L) based on adult studies (24). Our results were in concordance with other studies that used a quantitative

Citation: Int J Circumpolar Health 2012, **71**: 18999 - http://dx.doi.org/10.3402/ijch.v71i0.18999

Table II. Comparison of selected characteristics by broadband ultrasound attenuation of Inuit preschool children: Nunavut Inuit Child Health Survey, 2007–2008

	Broadband ultrasound attenuation (Δdb/ΔMHz)			
Nutrients/foods/characteristics[a,b]	Quartile 1	Quartile 2	Quartile 3	Quartile 4
Median [IQR]	32.5 [30.2–34.5]	38.4 [37.2–39.5]	52.3 [49.6–59.9]	52.3 [49.6–59.9]
Anthropometry[c,d]	(n = 71)	(n = 71)	(n = 70)	(n = 71)
BMI (kg/m^2)	17.0 ± 1.1^{x}	18.2 ± 1.1^{x,y}	18.6 ± 1.1^{y,z}	19.5 ± 1.1^{z}
BMI for age *z*-score	1.2 ± 2.3^{x}	1.6 ± 2.0^{x,y}	1.7 ± 2.2^{x,y}	2.2 ± 1.7^{y}
Weight for age *z*-score	0.6 ± 3.3^{x}	0.8 ± 2.9^{x,z}	1.2 ± 2.2^{y,z}	1.3 ± 2.3^{y,z}
Height for age *z*-score	0.3 ± 4.0	0.4 ± 3.1	0.4 ± 3.0	0.4 ± 3.1
Dietary[c,e]	(n = 69)	(n = 69)	(n = 69)	(n = 69)
Vitamin D (μg/d)	5.7 ± 0.9	5.7 ± 1.2	4.4 ± 0.9	5.8 ± 1.1
Calcium (mg/d)	795 ± 71	829 ± 94	734 ± 74	918 ± 66
Energy (Kcal/d)	1764 ± 104	1789 ± 76	1689 ± 64	1892 ± 68
Total protein (g/d)	63.1 ± 1.9	64.6 ± 1.9	63.1 ± 1.7	63.1 ± 1.8
Total fat intake (g/d)	52.5 ± 2.0	53.0 ± 1.8	47.9 ± 1.7	57.5 ± 1.7
MUFA intake (g/d)	19.4 ± 2.9	21.4 ± 2.2	18.8 ± 1.5	22.6 ± 1.9
PUFA intake (g/d)	8.1 ± 1.4	8.5 ± 1.0	7.4 ± 0.8	9.5 ± 1.1
SFA intake (g/d)	18.2 ± 2.2	19.0 ± 1.6	18.1 ± 1.6	19.9 ± 1.6
Biochemistry[c]	(n = 55)	(n = 55)	(n = 55)	(n = 54)
25(OH)D (nmol/L)	41.7 ± 1.9	44.7 ± 1.7	47.9 ± 1.7	44.7 ± 1.7
Lifestyle[c]	(n = 67)	(n = 67)	(n = 67)	(n = 67)
The number of hours spent outside (h/d)	2.1 ± 2.5	2.2 ± 2.3	2.0 ± 2.4	2.2 ± 2.3

[a]Data were log-transformed or square rooted before statistical analysis.
[b]Values are median [IQR] and geometric means ± SD. For a variable, means for variables with superscripts without a common letter differ, $p \leq 0.05$.
[c]ANOVA, followed by Bonferroni.
[d]According to World Health Organization (20).
[e]Data derived from 24-h recall.

ultrasound measurement. Krieg et al. (25) and Zochling et al. (18) did not find an association between vitamin D status and QUS parameters among large samples of institutionalized elderly. On the other hand, using dual-energy X-ray absorptiometry, among a younger sample of 12–15 year-old adolescents recruited throughout the year, in the United Kingdom, vitamin D status (≥ 74.1 nmol/L) was not associated with heel BMD. However, within the same study, only girls with high vitamin D status (≥ 74.1 nmol/L) had significantly greater forearm BMD (26). It has been suggested that the effect of vitamin D on the skeleton may be site-specific (27).

Using multivariable linear regression, age was a significant predictor of BUA. The effect of age on BUA was in agreement with results derived from quantitative ultrasound (28,29). Further, BMI was a significant predictor of SoS and BUA in the multivariable linear regression model, as hypothesized. Similarly, anthropometry including weight and BMI were positively associated with BUA (29,30) and SoS (28,31) in children and adults, but not once obesity is reached. An increasing number of studies suggests that a BMI > 30 kg/m^2 might interfere with bone health (32). In the current study, at this age and in absence of obesity, the lifestyle variables support accommodation of bone to the larger weight bearing load.

We did not find differences between BUA and SoS parameters in either gender. Similar results were reported by others among school-aged children using quantitative ultrasound (29,33). However, Zhu et al. (28) found gender differences, measured by BUA, between the ages of 12 to 13 years old. It is likely that gender differences might not appear until adolescence.

Higher MUFA intake was also a predictor of BUA. Similarly, MUFA intake was positively associated with bone health, particularly BMD measured by single photon absorptiometry, in both men and women in an epidemiological study in Greece (34). It is important to point out the main source of MUFA, in the Greek diet, is olive oil, which also contains PUFA. Likewise, the Inuit diet is known to be high in MUFA and PUFA, particularly derived from marine mammals and their oils (35) which are also rich sources of vitamin D (36). In the present study, MUFA, SFA and PUFA contributed

Table III. Comparison of selected characteristics by speed of sound quartiles of Inuit preschool children: Nunavut Inuit Child Health Survey, 2007–2008.

Nutrients/foods/ characteristics[a,b]	SoS quartiles (m/s)			
	Quartile 1	Quartile 2	Quartile 3	Quartile 4
Median [IQR]	1527.3 [1522.0–1530.1]	1537.7 [1535.1–1539.2]	1544.6 [1543.7–1546.6]	1557.9 [1553.4–1566.1]
Anthropometry[c,d]	(n = 71)	(n = 71)	(n = 70)	(n = 71)
BMI (kg/m^2)	18.0 ± 1.1^x	18.1 ± 1.1^{x,y}	18.2 ± 1.1^{x,y}	19.0 ± 1.1^y
BMI for age *z*-score	1.5 ± 2.5^x	1.6 ± 2.0^{x,y}	1.5 ± 2.1^{x,y}	2.1 ± 1.7^y
Weight for age *z*-score	0.9 ± 2.5^{x,y}	0.7 ± 3.7^x	0.9 ± 2.6^{x,y}	1.4 ± 2.0^y
Height for age *z*-score	0.3 ± 5.7	0.4 ± 2.7	0.4 ± 2.7	0.5 ± 2.7
Dietary[c,e]	(n = 69)	(n = 69)	(n = 69)	(n = 69)
Vitamin D (μg/d)	5.0 ± 1.0	4.7 ± 1.0	6.1 ± 1.2	5.6 ± 1.1
Calcium (mg/d)	727 ± 58	756 ± 85	954 ± 83	841 ± 75
Energy (Kcal/d)	1776 ± 107	1618 ± 72	1910 ± 81	1832 ± 49
Total protein (g/d)	60.2 ± 1.8	54.9 ± 2.0	72.4 ± 1.6	69.2 ± 1.6
Total fat intake (g/d)	56.2 ± 1.8	57.4 ± 1.6	47.3 ± 1.9	54.9 ± 1.7
MUFA intake (g/d)	19.2 ± 2.8	18.1 ± 1.9	23.7 ± 2.2	21.3 ± 1.5
PUFA intake (g/d)	7.5 ± 0.9	7.9 ± 1.1	9.6 ± 1.1	8.5 ± 0.8
SFA intake (g/d)	17.4 ± 2.2^x	16.0 ± 1.5^x	22.8 ± 1.5^y	19.3 ± 1.5^{x,y}
Biochemistry[c]	(n = 55)	(n = 55)	(n = 55)	(n = 54)
25(OH)D (nmol/L)	41.7 ± 1.8 (n = 42)	46.8 ± 1.8 (n = 42)	46.8 ± 1.7 (n = 42)	45.7 ± 1.7 (n = 43)
ALP (U/L)	173.8 ± 6.1^{x,y}	182.0 ± 1.2^{x,y}	173.8 ± 1.2^x	195.0 ± 1.3^y
Lifestyle[c]	(n = 67)	(n = 67)	(n = 67)	(n = 67)
The number of hours spent outside (h/d)	2.4 ± 2.4	2.0 ± 2.5	2.1 ± 2.4	2.0 ± 2.2

[a]Data were log-transformed or square rooted before statistical analysis.
[b]Values are median [IQR] and geometric means ± SD. For a variable, means for variables with superscripts without a common letter differ, $p \leq 0.05$.
[c]ANOVA, followed by Bonferroni.
[d]According to World Health Organization (21).
[e]Data derived from 24-h recall.

to 11.1, 10.0 and 4.6%, respectively, of total energy intake which is close to the intake of 1–8 year-old Canadian children in the Canadian Community Health Survey, Cycle 2.2 which reported an intake of MUFA, SFA and PUFA of 11, 12 and 4% of energy intake (37). Children in the 4th quartile of BUA or SoS tended to have higher intakes of PUFA and MUFA than children in the first quartile, but these trends did not reach statistical significance. It is likely that we did not observe any difference in PUFA intake, since we did not differentiate

Table IV. Predictors of broadband ultrasound attenuation of Inuit preschool children: Nunavut Inuit Child Health Survey, 2007–2008[a,b]

Variables	Coefficient [95% CI]	p
Age (years)	2.70 [0.71–4.70]	< 0.01
BMI percentile categories (1 < 85th, 2 = 85th–95th, 3 ≥ 95th)	10.33 [6.24–14.43]	< 0.01
MUFA intake (g/d)	0.29 [−0.08 to 0.49]	< 0.01
Latitude degree (°N)[c]	−0.08 [−0.56 to 0.40]	0.74
Constant	32.28 [−1.21 to 65.76]	0.06

[a]Multivariate linear regression.
[b]The number of hours spent outside, vitamin D status, sex, energy, protein, vitamin D and calcium intake did not contribute to the model.
[c]Latitude ranged from 56° 32′–72° 40′N.

Citation: Int J Circumpolar Health 2012, **71**: 18999 - http://dx.doi.org/10.3402/ijch.v71i0.18999

Table V. Predictors of speed of sound of Inuit preschool children: Nunavut Inuit Child Health Survey, 2007–2008[a,b]

Variables	Coefficient [95% CI]	p
Age (years)	1.26 [−1.25 to 3.78]	0.32
BMI percentile categories (1 < 85th, 2 = 85th–95th, 3 ≥ 95th)	8.34 [3.16–13.52]	< 0.01
MUFA intake (g/d)	0.23 [−0.33 to 0.49]	0.09
Latitude degree (°N)[c]	−0.59 [−1.2 to 0.20]	0.06
Constant	1570.28 [1527.89–1612.67]	< 0.01

[a]Multivariate linear regression.
[b]The number of hours spent outside, vitamin D status, latitude, sex, age, energy, protein, vitamin D and calcium intake did not contribute to the model.
[c]Latitude ranged from 56° 32′–72° 40′N.

between omega 3 and omega 6 and the ratio of omega 6: omega 3 due to the lack of the data in TF in the Canadian Nutrient File. Evidence suggests that BMD is positively correlated with omega 3 intake and is negatively associated with omega 6: omega 3 ratio in both men and women (38). Further, children in the higher 2 BUA or SoS quartiles did not have higher milk intake, dairy intake, TF intake, and monthly consumption of fish and marine mammals. All of these dietary variables were consumed scarcely, for instance mean daily milk intake of Inuit preschoolers was below 1 serving (data derived from the 24 h recall) and almost one-third of children did not consume fish and marine mammals in the previous month (data derived from the FFQ). Accordingly, it might be difficult to detect statistical differences in SoS or BUA. Several investigations have evaluated the role of calcium intake in bone health. Strong evidence suggests that calcium intake and absorption from childhood through early adulthood, is a crucial determinant of bone health throughout life (39), however, in the current study, this association was not detected, in contrast to the hypothesis. This observation is similar to another study among a large sample (n = 1,016) of children (6–13 years old) in Taiwan (29). It has been suggested that cross-sectional studies are less sensitive in finding the influence of calcium intake on bone health, in comparison to prospective studies involving calcium interventions (40). Recently, we published nutrient intakes of Inuit preschoolers using all available 24-h recall data (n = 374) (12). Mean intake of most nutrients, including protein, vitamin D and calcium was above the recommendations set by the Institute of Medicine (19). Only 8.8 and 24.0% met the recommended daily servings of fruits and vegetables and dairy products, respectively, of Canada's Food Guide for First Nations, Inuit and Métis.

The strength of this study is that, to our knowledge, it is the first to assess bone parameters in Inuit children, in combination with vitamin D intake, age, gender, latitude, BMI and other known predictors of bone health. However, the study findings cannot be generalizable beyond Inuit preschoolers and because the study was cross-sectional in nature, causality cannot be inferred. Further, for PTH measurements, intact PTH assays were used, instead of bioactive 1–84 PTH assays, that might have cross-reacted with carboxyl-terminal PTH fragments and lead to an overestimation of biologically active PTH (41). Physical activity was not assessed among Inuit preschoolers, and the literature has shown positive association between physical activity and bone health (39), future studies should take this factor into consideration. It is very challenging to measure physical activity in such a young population. Measuring physical activity through a pedometer or accelerometer would be an accurate measure; however compliance issues are problematic among preschoolers. Even though, peripheral quantitative ultrasound is the most suitable method to assess BMD in large populations in the field for its several advantages, being easy, portable, safe, cost-effective, rapid, and radiation-free, this technique is limited by the difficulty to compare its results with those of the dual-energy X-ray absorptiometry (16).

In summary, these data suggest that factors in support of bone health in young Inuit children are higher MUFA intake, older age and BMI. Since the dietary data was mainly derived from the 24 h recall data, further investigation assessing usual intakes of TF and nutrients affecting bone health using multiple 24 h recalls and FFQ along with serial assessment of vitamin D status of Inuit children across different seasons is required to confirm this observation. Further, future studies should assess physical activity, including weight bearing activities, and body composition, particularly body fat, as important predictors of bone health.

Acknowledgements

We thank Mrs Nelofar Athar Sheikh for her contribution in the data management and statistical advice, Mrs Louise Johnson-Down for her assistance in the analysis of the dietary data, Mrs Sherry Agellon for her help with the measurements of 25(OH)D and PTH concentrations, Mrs Angela Pacey for the data entry of BMD measures.

Authors' contributions

The author's responsibilities were as follows: GE and HW designed research; JH, GE and HW conducted research; GE and HW provided essential materials and reagents; JH analyzed data; JH wrote the manuscript and GE and HW critically reviewed the manuscript; JH had primary responsibility for final content. All authors read and approved the final manuscript.

Conflict of interest and funding

This study was supported by the Canadian Federal Program for International Polar Year, Canadian Institutes for Health Research, ArcticNet, Government of Nunavut, Indian and Northern Affairs Canada, and Health Canada.

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Citation: Int J Circumpolar Health 2012, **71**: 18999 - http://dx.doi.org/10.3402/ijch.v71i0.18999

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