Journal of Hazardous Materials 344 (2018) 55–63

Contents lists available at ScienceDirect

Journal of Hazardous Materials

journal homepage: www.elsevier.com/locate/jhazmat

Risk assessment of dietary lead exposure among First Nations people living on-reserve in Ontario, Canada using a total diet study and a probabilistic approach

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HIGHLIGHTS

- Lead exposure was 1.7 times higher among First Nations.
- Lead intake increase the risk of high blood pressure.
- Traditional food accounted for 73% of dietary intake.
- Lead containing ammunition is a major source.

ARTICLE INFO

Article history:
Received 25 April 2017
Received in revised form
19 September 2017
Accepted 21 September 2017
Available online 5 October 2017

Keywords:
Lead
Total diet study
Dietary exposure
First Nations
Ontario
Probabilistic modelling

ABSTRACT

Indigenous peoples have elevated risk of lead (Pb) exposure as hunted traditional food can be contaminated with lead-containing ammunition. Recent scientific consensus states that there is no threshold level for Pb exposure. The objective of this study was to estimate dietary exposure to Pb among First Nations living on-reserve in the province of Ontario, Canada. A total diet study was constructed based on a 24-h recall and Pb concentrations for traditional foods from the First Nations Food, Nutrition, and Environment Study (FNFNES) and Pb concentrations in market foods from Health Canada. A probabilistic assessment of annual and seasonal traditional food consumption was conducted. Results indicate that traditional foods, particularly moose and deer meat. are the primary source of dietary Pb intake (73%), despite providing only 1.8% of the average caloric intake. The average dietary Pb exposure (0.21 μg/kg/d) in the First Nations population in Ontario was 1.7 times higher than the dietary Pb exposure in the general Canadian population. Pb intake was associated with an estimated average increase in systolic blood pressure of 1.2 mmHg. These results indicate that consumption of foods hunted with Pb containing ammunition and shot puts the population at elevated risk of Pb toxicity.

1. Introduction

Lead (Pb) is a well-studied environmental pollutant with both naturally occurring and anthropogenic sources [1]. Historically, Pb was widely used as an additive in common consumer items such as paints and gasoline as well as in plumbing infrastructure, which resulted in elevated exposures in populations globally [1]. In North America, the level of Pb in these items has been reduced or even eliminated since the 1970′s due to stringent regulations [2,3]. Despite this decline, management of Pb exposures remains a modern health priority in North America as well as other regions around the world [4].

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https://doi.org/10.1016/j.jhazmat.2017.09.035
0304-3894/

Epidemiological studies over the past 10 years have shown conclusive evidence of adverse human health effects despite declining magnitudes of chronic Pb exposures in populations; this is most prominently observed with neurodevelopmental impairment among children [5–9]. In adults, the strongest weight of evidence for adverse health effects is supported by the association between chronic low dose Pb exposures and coronary heart disease characterized by increases in systolic blood pressure, and chronic kidney disease characterized by a reduction in the glomerular filtration rate [10–13].

In North America, diet is the predominant route of Pb exposure, primarily in the inorganic form [1]. In the general Canadian adult population, the average Pb intake from dietary sources was 0.12 μg/kg/d, as reported by the 2007 Canadian Total Diet Study (TDS) [14]. One of the limitations of TDSs is that they are a general screening tool and are unable to reflect the dietary exposures in populations with unique dietary compositions, such as the Aboriginal people living on-reserve. In Canada, approximately 1.4 million people identify as Aboriginal (First Nations, Inuit, Metis), representing approximately 4.3% of the Canadian population based on results from the 2011 National Household Survey [15]. The largest group of Aboriginal peoples in Canada are the First Nations accounting for approximately 60% of the Aboriginal population. There are over 600 First Nation bands and communities representing over 50 Nations [16]. The largest population of First Nations, comprising more than 200,000 people and 21% of the total First Nations population, reside in the province of Ontario, Canada [16]. About 30% of First Nations peoples live on reserves [15]. The diet of First Nations peoples is composed of a mixture of store-bought (or market foods) and traditional foods. Traditional food systems, which vary between Indigenous communities and are uniquely adapted include foods obtained from "the local, natural environment that are culturally acceptable" [17], which makes it distinct from the diet of the general Canadian population. Pb is found in many foods indirectly because of its ubiquitous nature as a pollutant due to its many industrial uses and natural occurrence as a metal. In traditional foods, however, there is also the potential for direct contamination through hunting with lead-containing shots and ammunition [18,19]. The assessment of dietary exposures of Pb in First Nations peoples to date has focused on specific traditional food items in small population samples [20–22]. Although traditional foods are largely unregulated from a contaminant perspective compared to store-bought market foods, they are a key component of health and connectedness in Aboriginal populations on the levels of the individual, family, community, culture, and environment [23,24]. Therefore, the characterization of their contribution to dietary sources of Pb in First Nations populations is necessary for the development of strategic and culturally relevant risk communication. There are few comprehensive Pb exposure assessments among First Nations. Dietary Pb intake is particularly of interest as toxicological reference values for assessing dietary exposures have been revoked by regulatory agencies [25,26] given the consistent and evolving body of evidence supporting low dose effects. This has culminated in the general scientific consensus that no threshold for Pb toxicity exists [27].

The objectives of this study were to (i) characterize and quantify sources of Pb in the total diet of First Nations adults in Ontario; (ii) assess the health risk to the population using the new non-threshold approach; and (iii) identify the key contributing food items for Pb exposures. A total diet approach using results from 24-h recall data was used to describe the relative contribution of Pb exposure from market food, traditional food and drinking water consumption. A more detailed probabilistic approach using food frequency questionnaire data was used to assess the Pb exposure from the consumption of traditional food.

2. Method

2.1. Ethics

Ethics approvals were obtained from the Research Ethics Board of the University of Ottawa and Health Canada.

2.2. First Nations food, nutrition and environment study (FNFNES)

Dietary patterns and contaminant concentrations in traditional foods were obtained through the *First Nations Food, Nutrition, and Environment Study (FNFNES)* Ontario region results collected in 2011–2012 [28]. The FNFNES was designed to study the diet of First Nations adults across Canada living on-reserve south of the 60th parallel. A total of 18 First Nation communities from the province of Ontario were selected to participate based on a systematic random sampling method with probability proportional to the size of the community. Community selection was designed to be representative of the First Nations population in the region based on a combined ecozone/cultural area framework. In the province of Ontario, three ecozone exist: the Boreal Shield, the Hudson Plains, and the Mixedwood Plains; and two cultural areas: Northeast and Subarctic [29]. Using this framework, First Nations communities in Ontario were stratified into four strata: Boreal Shield/Subarctic (Ecozone 1), Boreal Shield/Northeast (Ecozone 2), Hudson Plains/Subarctic (Ecozone 3), and Mixedwood Plains/Northeast (Ecozone 4) (Fig. 1). A minimum of four communities per strata, with a maximum of six were allowed. Participating household in each community were randomly selected with a target response rate of 100 households per community in all but two communities which had larger populations and a target of 200 households. At each household, one adult who met the following inclusion criteria was invited to participate: 19 years of age or older; able to provide written informed consent; self-identified as being a First Nation person living on-reserve in Ontario; and whose birthday was next among the adult members of the selected household. Participation was voluntary and a total of 1429 individuals participated [28]. All dietary exposure estimates are derived from data obtained through random samples of communities, households and persons. All results are adjusted with weights provided by Statistics that are the product of a design weight (the inverse of the selection probability) and of one or many adjustment factors (non-response and other random occurrences that could induce biases in the estimates). These design weights and adjustment factors are specific to each stage of the sample design and to each stratum used by the design [28]. The weighted results therefore reflect the whole population of First Nations in Ontario and not merely the sampled individuals.

All participating individuals completed a household interview which included the following sections: a 24-h dietary recall; traditional food frequency questionnaire (FFQ); socio/health/lifestyle questionnaire; and food security questionnaire. Interviews took place between mid-September and mid-December (Fall season). Interviews were conducted in person by community research assistants. Visual aids were used to assist participants in identifying species on the FFQ and in the quantification of foods and beverages. For the 24 h recall, participants were asked to describe the types and amounts of beverages and food consumed in the previous 24 h. For the FFQ, participants were asked to retrospectively recall the number of days on which they consumed each food item for the past four seasons, which were defined as 90-day periods. The FFQ included 143 traditional food items categorized into eight categories based on consultation with literature and community representatives. Categories with number of food items were: fish (29), land mammals (22), wild birds (25), wild berries/nuts (27),

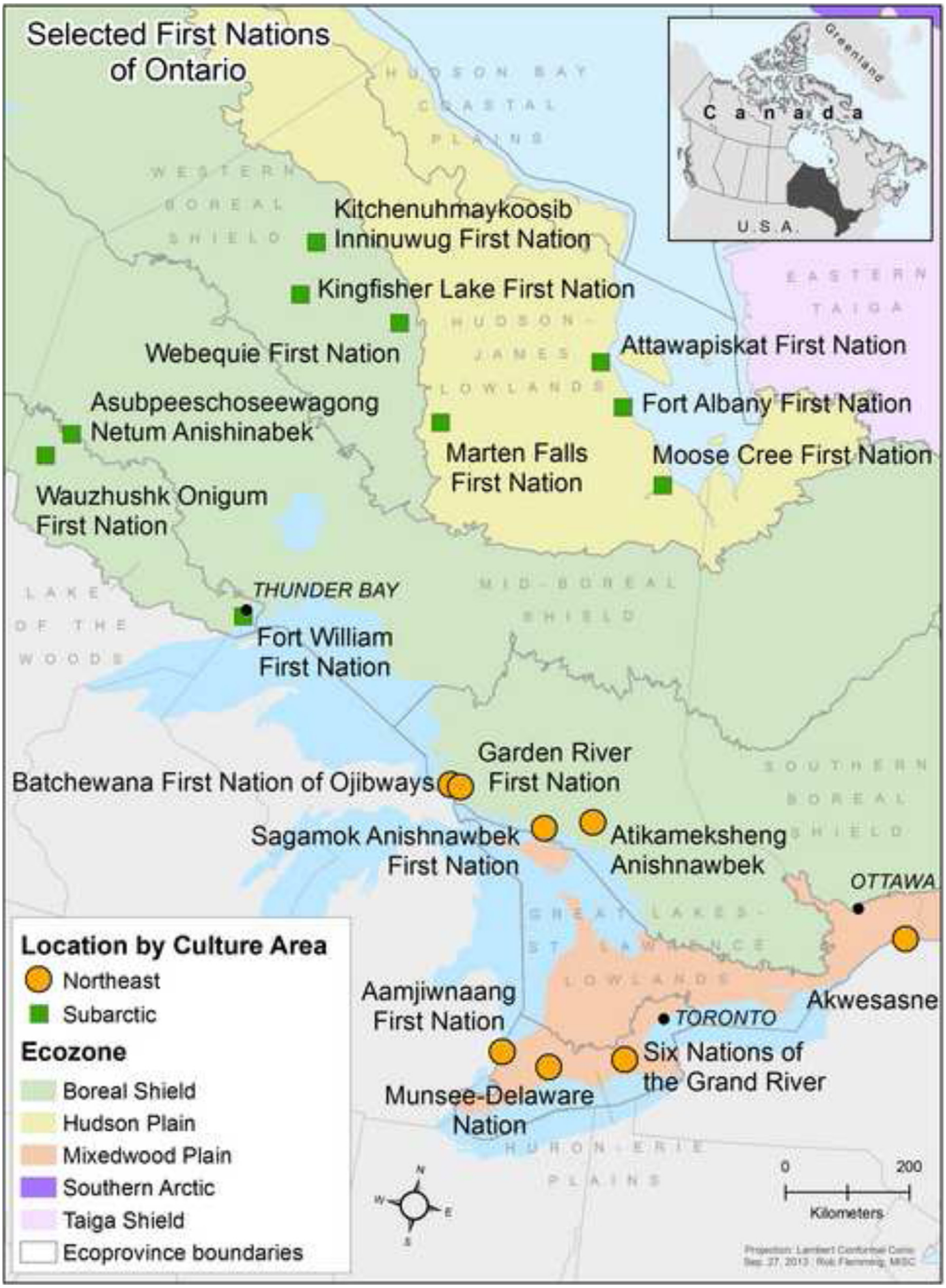

Fig. 1. Map of First Nations communities in Ontario participating in the FNFNES 2011–2012 [28].

wild plant roots, grains, shoots, and greens (29), tree foods (9), and mushrooms (2). When participants recalled consuming traditional food items not explicitly listed in the questionnaire, open-ended responses were recorded as "other" in the food category.

2.3. Tap water samples & analysis

In addition to the individual questionnaires, tap water samples were collected from a target of 20 participating households in each community through the FNFNES. Tap water samples were collected after water had been run for five minutes or until the water was cold in order to flush the stagnant water out of the pipe. Prior to analysis, water samples were filtered through a 0.45 μm pore filter and digested using nitric acid based on EPA method #200.2. Inductively Coupled Argon Plasma Mass Spectroscopy (ICP/MS) was used to analyse Pb concentration based on EPA method #200.8. For QA/QC, the percent recovery of certified reference material ranged from 87.7–106.7%, with the percent recovery of matrix spiked samples ranging similarly from 92.3–105%. The limit of detection (LOD) for Pb was 0.005 μg/L.

2.4. Traditional food samples & analysis

Traditional food samples were collected from participating FNFNES communities based on lists containing items that were (1) commonly consumed, (2) of importance for nutrition or environmental concerns, and (3) were known to accumulate higher concentrations of contaminants. Communities provided up to 30 composite food samples, with each composite comprising tissue from up to five replicates. A total of 419 composite food samples comprising a sum total of 1237 replicates and representing 141 different traditional food items were analyzed for Pb content. Pb content was analyzed from homogenized composite samples digested in an open vessel using a combination of nitric acid and hydrogen peroxide based on EPA 200.3/6020A. Inductively coupled plasma mass spectrometry (ICP/MS) was employed to quantify Pb concentrations with a limit of detection of 0.004 μg/g. Recovery of certified reference material ranged between 70 and 130%. Pb concentrations in traditional food was compared to the CODEX alimentarius standard established by the FAO and WHO [30].

2.5. Total diet study

Total diet studies are conducted to assess intakes of key nutrients and contaminants in a population [31]. Using data from the 24-h recall survey of FNFNES, the individual intakes of recalled food (expressed as grams per day) was multiplied by the average concentration of Pb in that food item in the database of contaminant concentration to determine the dietary Pb exposures. Pb concentrations of market food were collected in 2011 and 2012 through the Canadian Total Diet Study by Health Canada [32]. Pb concentrations in traditional food items were measured as described above. Water consumption was included in the calculation of Pb intakes as per harmonized total diet study guidance [31], with concentrations in tap water represented by community-specific values measured by FNFNES. Contaminant concentrations values below the limit of detection were represented by an upper-bound approach to provide conservative estimate as the limit of detection varied between different data sources. Dietary Pb intakes have been reported as weighted values to account for factors such as design weight (the inverse of the selection probability) and adjustment factors (non-response rate).

2.6. Risk assessment of dietary intake

In adult populations, the adverse outcome of chronic Pb exposure with the strongest weight of evidence as assessed by the Joint FAO/WHO Expert Committee on Food Additives (JECFA) was an increase in systolic blood pressure [33]. Based on epidemiological evidence, JECFA concluded that a dietary exposure of 1.3 μg/kg/d was associated with a 1 mmHg increase in systolic blood pressure [33]. Dietary exposures in this study were compared to this JECFA established level using a margin of exposure (MOE) approach ESFA, 2010. The MOE is the ratio between the observable effect level and the exposure level of the studied population and MOEs less than 1 means that the studied population has in increased health risk from the exposure.

2.7. Traditional food probabilistic exposure assessment

A probabilistic approach was used to estimate Pb exposure from annual traditional food consumption. This provided a detailed assessment that reflected the variability in the types and amounts of traditional foods consumed throughout the year and the ability to identify and prioritize patterns of exposures in this dietary component. Monte Carlo simulations were constructed in Excel 2010 add-in Crystal Ball (Oracle; version 11.1.2.3). Pb intake for each iteration $_j$ ($\mu g \cdot kg^{-1} \cdot d^{-1}$) was modelled based on the sum of the product of the consumption of each food $_i$ ($g \cdot d^{-1}$) by Pb concentration $_i$ ($\mu g \cdot g^{-1}$), divided by body weight $_j$ (Eq. (1)).

$$\text{Pb Intake}_j(\mu \text{ g/kg/d}) = \sum_{i=1}^{69} \frac{\left[\text{food}_i(\text{g/d})\right] \times [\text{Pb}]_i(\mu \text{ g/g})}{\text{Body Weight}_j(\text{kg})} \quad (1)$$

Consumption frequencies from FFQ were converted into grams by applying age and sex specific serving size data for food groups reported through 24-h recall responses. Daily consumption values in grams per day were computed by averaging intakes over a one-year period. Traditional food items were included in the simulation if consumption was reported in more than 5% of the population to limit input parameters with negligible bearing on simulation outputs. The total numbers of traditional food items included were 69 at the regional level, 47 in Ecozone 1, 63 in Ecozone 2, 41 in Ecozone 3, and 55 in Ecozone 4. Consumption data were parameterized using the custom distribution function in Crystal Ball as the sample data were representative of the provincial and ecozone populations of First Nations in Ontario. Input distributions of Pb concentrations in each traditional food item was represented through FNFNES traditional food composite analysis fitted to lognormal distributions described by the average, the standard deviation derived as an assumed coefficient of variation of 100%, and bounded by LOD/2 and three standard deviations. For traditional food items with no direct Pb concentrations from the FNFNES samples, surrogate food items were selected from the data set based on trophic level and species similarities. Body weight data were obtained through the FNFNES and included as an input through a custom distribution function. Simulations were constructed for the total provincial population, ecozone populations, women of child-bearing age subpopulation ($n = 562$), and seasonal simulations for the total provincial population. Simulations were run for 10 000 iterations with a Monte Carlo sampling method.

2.8. Statistical analysis

JMP statistical software (version 12.1.0) was used to obtain summary statistics. Output distributions did not conform to assumptions of normality prior and after a logarithmic transformation. Non-parametric statistical tests were used to compare outputs. Tests of statistical differences utilized Kruskal-Wallis test, with significance considered as $p < 0.05$.

3. Results

3.1. Estimate of dietary Pb exposure

Results from the total dietary study show that Ontario First Nation had an average Pb intake of 0.21 μg/kg/day which is higher than the average of the general Canadian population (0.12 μg/kg/d) (Table 1). Traditional food sources represent 72.7% of the average Pb intake for the population, while representing only 1.8%

Table 1
Summary of Pb intake (μg/kg/d) for results of the Total Diet Study in Ontario First Nations adults living on-reserve, compared to results of the 2007 Canadian Total Diet Study.

	n	Mean Pb Dose (μg/kg/d)	Standard Error	50th	90th	95th	97.5th	99th	
Canada Total Diet Study 2007 [a]			0.12						
Total Population	Ontario	1429	0.21	0.024	0.056	0.15	1.6	2.8	5.0
	Ecozone 1	359	0.24	0.046	0.055	0.58	2.4	4.3	5.5
	Ecozone 2	344	0.14	0.025	0.057	0.11	0.18	1.1	2.9
	Ecozone 3	266	0.61	0.094	0.06	2.0	3.4	4.8	6.3
	Ecozone 4	460	0.16	0.047	0.053	0.11	0.14	0.2	1.8
Traditional Food Consumers	Ontario	190	1.5	0.18	0.82	4.5	5.5	8.0	13
	Ecozone 1	70	1.4	0.25	0.59	5.0	5.5	12.2	24
	Ecozone 2	26	1.2	0.28	0.47	3.1	4.0	4.4	4.4
	Ecozone 3	62	2.6	0.30	1.7	4.8	5.8	7.4	7.8
	Ecozone 4	32	1.4	0.60	0.076	2.2	12	12	12
non-Traditional Food Consumers	Ontario	1239	0.059	0.0011	0.052	0.097	0.12	0.15	0.18
	Ecozone 1	289	0.060	0.0025	0.048	0.099	0.12	0.16	0.19
	Ecozone 2	318	0.061	0.0019	0.055	0.097	0.12	0.14	0.18
	Ecozone 3	204	0.050	0.0020	0.049	0.087	0.10	0.12	0.15
	Ecozone 4	428	0.057	0.0016	0.052	0.10	0.12	0.16	0.20

[a] Health Canada [32].

Table 2
Summary of Pb intakes (μg/kg/d) from Market Food sources for non-traditional food consumers (n = 1239) based on 24-h recall consumption data.

	n	Mean Pb intake (μg/kg/d)	Standard Error	50th	90th	95th	97.5th	99th
Ontario	1239	0.056	0.0010	0.049	0.095	0.11	0.14	0.18
Ecozone 1	289	0.056	0.0025	0.044	0.097	0.12	0.16	0.19
Ecozone 2	318	0.059	0.0018	0.053	0.095	0.12	0.14	0.18
Ecozone 3	204	0.050	0.0020	0.048	0.086	0.10	0.12	0.15
Ecozone 4	428	0.054	0.0016	0.050	0.097	0.12	0.16	0.20

of the average caloric intake (Supplementary Table A2). Based on responses to the 24-h recall, 13% of the population reported consuming traditional foods. Therefore, First Nations adults in Ontario who consume traditional foods had higher Pb intakes the those who did not consume traditional foods on the 24-h recall (Table 1). Moreover, the northern-most ecozones, ecozone 1 and ecozone 3, had greater percentages of their populations (19.5% and 23.3% respectively) consuming traditional food compared to the provincial First Nations adult population living on-reserve (Table 1).

For the non-traditional food consumers, market foods represented an average 96% of the Pb intake or 0.056 μg/kg/d (Table 2) with the remaining 4% of the Pb intake arising from tap water consumption. In comparison, among adults reporting traditional food consumption on the 24-h recall, market foods accounted for an average of only 3% whereas traditional foods accounted for 97% of the total Pb intake. The contribution of traditional foods to the Pb intake at the ecozone level ranged from 93% in ecozone 2–97% in ecozones 1 and 3 (northern-most ecozones). The dietary Pb intake from traditional food sources is presented in Table 3a. Ecozone 3 had the highest average Pb intake from traditional food consumption; however, ecozone 1 had the widest range of Pb intakes. The summary of estimated Pb intakes from annual traditional food consumption, as calculated through a Monte Carlo simulation using consumption data from the FFQ is presented in Table 3b. Ecozone 3 has the highest mean Pb intakes, which is consistent with findings from Pb intakes computed through a total diet study approach (Table 3a). Dietary Pb intakes expressed as a daily average from annual traditional food consumption (Table 3b) are lower than intakes estimated through the total diet study (Table 3a) as the patterns of traditional food consumption in the latter include low traditional food consumers, as well as fluctuations in seasonally available food items.

The top five market foods contributing to the total dietary Pb intakes accounted for 9.8% of the average total Pb intake (Table 4). In southern ecozones (ecozones 2 and 4), market foods were a greater contributor to Pb intakes (16.9% and 13.3%, respectively). The top five traditional foods contributing to the total dietary Pb intakes are presented in Table 5a. Moose and deer meat were the main sources of Pb intakes. Table 5b presents the top five traditional foods based on annual traditional food consumption, and therefore reflects seasonal availability, unlike Table 5a which was based on a 24-h recall conducted in the fall season. Despite the temporal differences, moose and deer meat remain the leading traditional foods contributing to the Pb dose which suggests both methods capture similar leading sources of dietary Pb.

Pb concentrations in five of the commonly consumed traditional food items were compared to Codex alimentarius guidance on the maximum level of Pb in the meat of cattle, pig, sheep and poultry (without bones) (Table 6). The Codex threshold (0.1 ug/g) for Pb was exceeded the in 24% to 63% of composite samples of these food items (moose and mallard, respectively).

3.2. Risk assessment of total dietary Pb exposure (Total diet study)

The calculated mean (0.21 μg/kg/d) and 95th percentile intakes (1.6 μg/kg/d) were below the previous PTWI from Health Canada (25 μg/kg/wk) for total dietary Pb exposure at the province, as well as for the ecozone level. In light of this reference dose being reviewed, we used an alternative margin of exposure (MOE) approach which is proposed by the European Food Safety Authority (EFSA) CONTAM panel for risk characterization of dietary Pb exposures [25]. Pb exposures from the total diet study were compared to the exposure level (1.3 μg/kg/d) associated with an increase of 1 mmHg in the systolic blood pressure in adults as defined by JECFA [33]. Based on the total diet, the 95th percentile of the provincial population as well as in ecozones 1 and 3 showed a MOE of less than 1 (Table 7). However, the population subgroups of individuals who reported no consumption of traditional food in the past 24 h showed a 20 times reduction in risk (with MOE higher than 1) compared to the population who reported traditional food consumption. This trend remains at the upper percentiles of the Pb intake, as the margin of exposure was greater than 10 for the

Table 3a
Summary of Pb intakes (μg/kg/d) from traditional food sources in traditional food consumers using data from the 24-h recall (n = 190).

	n	Mean Pb Intake (μg/kg/d)	Standard Error	50th	90th	95th	97.5th	99th
Ontario	190	1.5	0.18	0.78	4.4	5.4	8.0	13
Ecozone 1	70	1.4	0.25	0.56	5.0	5.5	12	24
Ecozone 2	26	1.2	0.28	0.39	3.0	3.9	4.3	4.3
Ecozone 3	62	2.5	0.30	1.7	4.8	5.8	7.4	7.8
Ecozone 4	32	1.4	0.60	0.014	2.2	11	12	12

Table 3b
Summary of Pb intakes (μg/kg/d) from traditional food consumption using Food Frequency Questionnaire and simulated through a Monte Carlo Simulation. (n = 10,000 iterations).

	Mean Pb Intake (μg/kg/d)	Standard Error	50th	90th	95th	97.5th	99th
Ontario	0.18	0.0047	0.051	0.44	0.73	1.2	2.1
Ecozone 1	0.21	0.0033	0.10	0.50	0.77	1.1	1.5
Ecozone 2	0.20	0.0054	0.035	0.48	0.93	1.5	2.5
Ecozone 3	0.24	0.0052	0.11	0.49	0.79	1.2	2.1
Ecozone 4	0.12	0.0054	0.0082	0.24	0.49	0.99	1.8

Table 4
Top 5 market foods contributing to the total Pb intake with mean intake (ug/kg/d), standard error (SE), and the percent contribution to the total Pb intake.

Food	Mean Pb Intake (μg/kg/d)	SE	% of Total Intake
Ontario			
Coffee	0.0076	0.00024	3.6%
Soft drinks, canned	0.0058	0.00029	2.8%
Luncheon meat, cold cuts	0.0029	0.00023	1.4%
Bread, white	0.0026	0.000099	1.2%
Cereal, oatmeal	0.0018	0.00014	0.9%
Total	0.21	0.024	9.8%
Ecozone 1			
Soft drinks, canned	0.0062	0.00067	2.6%
Coffee	0.0061	0.00039	2.5%
Luncheon meat, cold cuts	0.0030	0.00050	1.3%
Cereal, oatmeal	0.0027	0.00035	1.1%
Bread, white	0.0023	0.00018	1.0%
Total	0.24	0.046	8.5%
Ecozone 2			
Coffee	0.0098	0.00053	7.0%
Soft drinks	0.0055	0.00046	3.9%
Luncheon meat, cold cuts	0.0033	0.00047	2.4%
Bread, white	0.0031	0.00020	2.2%
Cereal, wheat, rice, bran	0.0020	0.00031	1.4%
Total	0.14	0.025	16.9%
Ecozone 3			
Soft drinks, canned	0.0056	0.00052	0.9%
Coffee	0.0054	0.00047	0.9%
Bread, white	0.0030	0.00023	0.5%
Pasta	0.0021	0.00044	0.3%
Luncheon meat, cold cuts	0.0019	0.00031	0.3%
Total	0.61	0.094	3.0%
Ecozone 4			
Coffee	0.0082	0.00049	5.1%
Soft drinks	0.0057	0.00048	3.6%
Luncheon meat, cold cuts	0.0028	0.00039	1.8%
Bread, white	0.0026	0.00019	1.6%
Cereal, wheat, rice, bran	0.0019	0.00035	1.2%
Total	0.16	0.047	13.3%

Table 5a
Top 5 traditional foods with mean Pb intake (μg/kg/d), standard error (SE), and the percent contribution of the total dietary Pb intake based on the Total Diet Study.

Food	Mean Pb Intake (μg/kg/d)	SE	% of Total Intake
Ontario			
Moose Meat	0.12	0.017	57.1%
Deer Meat	0.043	0.018	20.5%
Canada goose	0.002	0.0014	1.0%
Trout	0.00031	0.00011	0.1%
Pickerel	0.00023	0.000055	0.1%
Total	0.21	0.024	78.8%
Ecozone 1			
Moose Meat	0.17	0.040	70.8%
Deer Meat	0.0095	0.024	4.0%
Trout	0.00071	0.00034	0.3%
Pickerel	0.00047	0.00014	0.2%
Whitefish	0.00038	0.00015	0.2%
Total	0.24	0.046	75.4%
Ecozone 2			
Moose Meat	0.042	0.015	30.0%
Deer Meat	0.032	0.020	22.9%
Rabbit meat	0.00083	0.00064	0.6%
Moose Tongue	0.00040	0.00070	0.3%
Moose Liver	0.00024	0.00024	0.2%
Total	0.14	0.025	53.9%
Ecozone 3			
Moose Meat	0.52	0.093	85.2%
Canada goose	0.035	0.014	5.7%
Caribou meat	0.0033	0.0012	0.5%
Turkey meat	0.00055	0.00081	0.1%
Whitefish	0.00019	0.00021	0.0%
Total	0.61	0.094	91.6%
Ecozone 4			
Deer Meat	0.097	0.046	60.6%
Moose Meat	0.0076	0.0050	4.8%
Maple Syrup	0.00037	0.00017	0.2%
Perch	0.00018	0.000098	0.1%
Winter squash	0.000091	0.000047	0.1%
Total	0.16	0.047	65.8%

95th percentile, with exposures at less than 10% of the exposure guidance value. In the subpopulation reporting traditional food consumption in the past 24 h, all regions except ecozone 2 had a margin of exposure less than 1 between the mean total dietary Pb dose and the exposure value, suggesting an elevated risk. For the province as a whole, the Pb intake was associated with an estimated average increase in systolic blood pressure of 1.2 mmHg, and up to 2 mmHg for ecozone 3. At the 95th percentile of the subpopulation reporting traditional food consumption in the past 24 h, the total dietary Pb intake corresponded to an estimated 4.22 mmHg increase in systolic blood pressure for the population, with a regional range up to an increase of 8.9 mmHg in ecozone 4.

Table 5b
Top five traditional foods with mean Pb intake (μg/kg/d), standard error (SE), and mean consumption (g/d) based on annual traditional food consumption as reported in the FFQ and modelled with Monte Carlo Simulation.

Food	Mean Pb Intake (μg/kg/d)	SE	Mean Intake (g/d)
Ontario			
Deer Meat	0.078	0.0040	1.6
Moose Meat	0.065	0.0020	6.7
Beaver	0.018	0.0020	0.37
Goose	0.0078	0.00037	3.6
Mallard	0.0073	0.0010	0.44
Total	0.18	0.0047	38
Ecozone 1			
Moose Meat	0.099	0.0021	10
Beaver	0.043	0.0020	0.77
Deer Meat	0.034	0.0014	0.70
Goose	0.013	0.00040	5.2
Mallard	0.0087	0.00064	0.50
Total	0.216	0.0033	61
Ecozone 2			
Deer Meat	0.13	0.0049	2.6
Moose Meat	0.060	0.0022	6.3
Deer Liver	0.0033	0.00048	0.22
Beaver	0.0015	0.00019	0.030
Mallard	0.00062	0.000099	0.030
Total	0.20	0.0054	32
Ecozone 3			
Moose Meat	0.11	0.0027	12
Beaver	0.045	0.0041	0.87
Goose	0.028	0.00074	12
Mallard	0.026	0.00090	1.6
Teal	0.0093	0.00057	0.55
Total	0.24	0.0052	53
Ecozone 4			
Deer Meat	0.11	0.0054	2.3
Moose Meat	0.0070	0.00046	0.75
Black Bear	0.00065	0.000083	0.010
Beaver	0.00050	0.000059	0.010
Mallard	0.00045	0.000043	0.030
Total	0.12	0.0054	14

3.3. Monte Carlo simulation

To assess trends in Pb intake as a function of seasonal traditional food consumption, inputs for the consumption distributions in the simulation were represented by seasonal average (fall, winter, spring, summer). Since seasonality influences the species availability, a moderate trend coinciding with peak hunting seasons was expected. Elevated Pb intakes were observed in the fall and winter seasons for the population, and at all ecozones except ecozone 3 which had Pb intake peaks during the fall and spring/summer which is reflective of a fall game hunt and a spring bird hunt. Seasonal differences were statistically significant for each ecozone (Supplementary Table A3). Simulations for women of childbearing age resulted in slight, but significantly ($p < 0.0001$) lower mean Pb intake at the provincial level, as well as in ecozones 1 and 3 with differences of 0.027, 0.063, 0.102 μg/kg/d, respectively.

4. Discussion

Previous studies have observed terrestrial game, waterfowl, and birds hunted with lead-containing ammunition to have elevated Pb concentrations in homogenized tissue samples due to fragmentation of bullets and shots [35,36,19,37]. Concentration of Pb in moose, deer, and goose hunted with non-lead shot and ammunition have been previously documented to have background Pb concentrations below the Codex maximum contaminant level [38–40]. In water fowl, the background concentration of Pb is difficult to discern due to the potential ingestion of legacy and stray lead-containing shots that may become a source of internal exposure to elevated tissue Pb concentrations. The elevated Pb concentrations observed in mallard samples in this study may be the result of internal distribution from such sources.

Results from the probabilistic assessments are in agreement with the conclusion of the total diet assessment in that the upper percentiles of the population distribution are at risk of adverse effect when traditional foods which may be harvested by a mix of hunting method (i.e. different types of ammunition, traps etc.) are consumed. Previous studies in Indigenous populations corroborate the conclusion that foods hunted with lead-containing ammunition and shot elevates blood Pb values, especially consumption of birds and waterfowl [18,41,42]. The consumption of game meats, primarily deer and moose, hunted with lead-containing shots have also been associated with increased blood Pb concentrations in general hunting populations [43,44]. In comparison to blood Pb levels in southern urban population, previous studies have indicated the sources of Pb in the Inuit and First Nations populations to be predominantly from lead-containing hunting shots and ammunition based on the analysis of isotopic ratios [45,20,46,22].

The risks of Pb exposure from dietary items may be underestimated; since lead-containing shots and ammunition can be unintentionally ingested as whole pellets, and they may become an internal source of Pb exposure if lodged in the gastrointestinal tract or appendix [47–50]. A study of Northern Ontario First Nations suggested approximately 15% of individuals have ingested lead shots lodged in abdominal regions based on a review of abdominal x-rays [50]. Despite these considerations, national biomonitoring results from the 2011 First Nations Biomonitoring Initiative (FNBI) indicate blood Pb values in First Nations to be significantly lower than the general Canadian population assessed in Cycle 1 (2007–2009) of the CHMS [51,52]. However, in comparison to CHMS Cycle 2 (2009–2011) results, FNBI blood Pb levels appear to be in closer agreement with the Canadian population average although the significance of the trend is unknown and warrants further investigation [53]. Limitations in the FNBI include limited assessments of lifestyle or dietary parameters relevant to Pb exposures and therefore limited profiling and insights on the traditional food consumption of participants can be concluded [54]. Future assessments should be conducted on the living, recreational and working environments to provide detailed context on Pb exposures in this population.

Based on the total diet study results, traditional foods are the primary dietary contributor to Pb intakes in the First Nations population in Ontario. Compared to dietary Pb exposure of the general Canadian population [34], the provincial First Nations population has an average dietary Pb exposure 1.7 times greater. However, in First Nations individuals with no reported traditional food consumption in the past 24-h, the total dietary Pb intake is approximately half the Canadian average, whereas for traditional food consumers, the total dietary Pb intake is more than 7 times greater than the Canadian average. Similar patterns were observed with mean Pb intakes generated by the probabilistic assessment where levels were similar to the Canadian average in ecozone 4, but elevated between 1.5–2 times in the total province and ecozone 3, respectively (Table 3b). This is reflective of higher traditional food consumption in northern ecozones, whereas southern ecozones consume less traditional foods. The consumption of different types of traditional food items (i.e. game vs. bird vs. fish) has been hypothesized to be closely correlated complicating the identification of key contributing sources to the blood Pb burden in subsistence populations. Across the province of Ontario, this study found a strong correlation between the consumptions of moose and Canadian goose ($\rho = 0.6421$, $p < 0.0001$). In regional ecozones, this correlation was strongly observed only in ecozone 1, ($\rho = 0.6485$,

Table 6
Concentrations of Pb with ranges in five common food items consumed across the province.

	n (replicates)	n (composites)	Mean (SD)[a] ug/g	GM	Range ug/g	<Codex Threshold [b]		>Codex Threshold [c]	
						Mean (SD) [a] ug/g	% of n	Mean (SD) [a] ug/g	% of n
Beaver	30	11	9.4 (28)	0.044	< LOD – 99	0.017 (0.024)	73%	34 (56)	27%
Deer	42	10	4.5 (13)	0.032	< LOD – 42	0.035 (0.024)	70%	22 (24)	30%
Moose	83	17	0.87 (3.0)	0.022	< LOD – 13	0.013 (0.022)	76%	3.7 (6.2)	24%
Canada Goose	28	8	0.39 (0.48)	0.039	< LOD – 1.2	0.0027 (0.0014)	50%	0.78 (0.46)	50%
Mallard	26	8	1.6 (2.7)	0.25	< LOD – 8.5	0.032 (0.031)	38%	2.5 (3.4)	63%

< LOD included as LOD/2; LOD = 0.004 ug/g.
b - Pb [] < 0.1 ug/g.
c - Pb [] > 0.1 ug/g.

Table 7
Summary of Margin of Exposures (MOE) between total Pb dietary intakes (from 24-h recall) and the exposure level of 1.3 μg/kg/d associated with an increase of 1 mmHg in the systolic blood pressure in adults as defined by JECFA. MOEs less than 1 indicate elevated exposure risk, and have been bolded.

		n	Mean	50th	90th	95th	97.5th	99th
Total Population	Ontario	1429	6.2	23	8.7	**0.81**	**0.46**	**0.26**
	Ecozone 1	359	5.4	24	2.2	**0.54**	**0.30**	**0.24**
	Ecozone 2	344	9.3	23	12	7.2	1.2	**0.45**
	Ecozone 3	266	2.1	22	**0.65**	**0.38**	**0.27**	**0.21**
	Ecozone 4	460	8.1	25	12	9.3	6.5	**0.72**
Traditional Food Consumers	Ontario	190	**0.87**	1.6	**0.29**	**0.24**	**0.16**	**0.10**
	Ecozone 1	70	**0.93**	2.2	**0.26**	**0.24**	**0.11**	**0.05**
	Ecozone 2	26	1.1	2.8	**0.42**	**0.33**	**0.30**	**0.30**
	Ecozone 3	62	**0.5**	**0.76**	**0.27**	**0.22**	**0.18**	**0.17**
	Ecozone 4	32	**0.93**	17	**0.59**	**0.11**	**0.11**	**0.11**
non-Traditional Food Consumers	Ontario	1239	22	25	13	11	8.7	7.2
	Ecozone 1	289	22	27	13	11	8.1	6.8
	Ecozone 2	318	21	24	13	11	9.3	7.2
	Ecozone 3	204	26	27	15	13	11	8.7
	Ecozone 4	428	23	25	13	11	8.1	6.5

$p < 0.0001$). However, the inclusion of correlation coefficients for strongly correlated variables in the Monte Carlo simulation did not significantly change the output distribution of total Pb intake, and therefore were not included.

Despite contributing to the Pb intake, traditional foods are an important determinant of health among First Nations peoples. The FNFNES regional analysis indicated the quality of the total diet improves in terms of essential nutrient intake and a reduction in saturated fats, sugars, and sodium on days when traditional foods are consumed [28], consistent with findings from previous studies in Canadian Indigenous populations [55]. Moreover, despite the known relationship between blood lead concentrations and health outcomes, population studies on dietary lead exposure and health outcomes are limited. Risk managers need to conduct detailed risk-benefit assessment of associated with individual traditional food and estimate the net effect on the health of the populations before issuing any dietary advice.

5. Conclusion

This study comprehensively quantifies the dietary Pb exposures in First Nations adults living on-reserve in Ontario for the first time in a total diet context as well as more detailed characterization of annual traditional food consumption patterns. Results indicate the variability in Pb concentration in traditional food items to be the most sensitive input and predictor of risk. Given the range of Pb concentrations in traditional foods beyond previously characterized background levels, it is presumed that a diet which occasionally includes foods hunted with lead-containing ammunition and shot puts the population at elevated risk of Pb toxicity. Future studies on dietary Pb exposures in this population should include an assessment of the hunting method as well as blood Pb concentrations, and economic and social implications of a lead shot policy.

Acknowledgements

We would like to express our gratitude to all participants for their cooperation and participation in the First Nations Food, Nutrition and Environment Study (FNFNES). We thank all First Nations community members who collected food and water samples, assisted in data collection, coordinated research activities, and arranged meetings and public gathering to share information. FNFNES is funded by Health Canada and the communication of the results to the communities was also funded by the Canadian Institute of Health Research. A.K. Juric was supported by a NSERC-CREATE Research in Environmental Analytical Chemistry and Toxicology scholarship. H.M. Chan is supported by the Canada Research Chair Program. The authors have no potential conflicts of interest.

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at https://doi.org/10.1016/j.jhazmat.2017.09.035.

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