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## Consumption of tomato products is associated with lower blood mercury levels in Inuit preschool children

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### ABSTRACT

Some evidence suggests that various diet components and nutrients, including vegetables, fruit and foodderived antioxidants, could mitigate contaminant exposure and/or adverse health effects of contaminants. To examine the effect of the consumption of tomato products on blood mercury levels in Inuit preschool children, 155 Inuit children ( $25.0 \pm 9.1$  months) were recruited from 2006–2008 in Nunavik childcare centers (northern Québec, Canada). Food frequency questionnaires were completed at home and at the childcare center, and total blood mercury concentration was measured by inductively coupled plasma-mass spectrometry. Multivariate regression analysis was performed after multiple imputation. The median blood concentration of mercury was 9.5 nmol/L. Age, duration of breastfeeding, annual consumption frequency of seal meat, and monthly consumption frequency of tomato products were significant predictors of blood mercury levels, whereas annual consumption frequencies of beluga muktuk, walrus, Arctic char, and caribou meat were not. Each time a participant consumed tomato products during the month before the interview was associated with a 4.6% lower blood mercury level (p = 0.0005). All other significant predictors in the model were positively associated with blood mercury levels. Further studies should explore interactions between consumption of healthy store-bought foods available in Arctic regions and contaminant exposure.

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## 1. Introduction

The Canadian Arctic Indigenous Peoples are intimately connected to their environment. Foods they harvest through fishing, hunting, and gathering are an essential part of their identity and provide them cultural, socioeconomic, and nutritional benefits. These foods are an important contributor of key nutrients to their diet, increasing the levels of protein and many vitamins and minerals (Gagné et al., 2012; Johnson-Down and Egeland, 2010; Kuhnlein and Receveur, 2007). However, the multiple benefits of traditional foods must be weighed against the detrimental aspects of contaminants found in the Arctic food chain (Dewailly, 2006). Owing to their traditional lifestyle and dependence on traditional foods, Inuit are particularly vulnerable to mercury present in the Arctic environment (AMAP, 2011). Mercury is carried by air and water currents from southern to northern latitudes and deposited in Arctic ecosystems. In the environment, mercury is methylated into methylmercury by microbial action, notably in aquatic ecosystems, and biomagnified in the food webs (AMAP, 2011; Clarkson and Magos, 2006). The consumption of fish and marine mammals is the most significant source of methylmercury exposure in Inuit (Dewailly et al., 2001; Hansen and Gilman, 2005; Muckle et al., 2001).

Methylmercury exposure is known to interfere with neurodevelopment in children, especially in the domains of verbal function, visuo-motor integration, and memory (AMAP, 2009; Karagas et al., 2012). However, despite an abundant literature on methylmercury, the threshold dose for neurotoxic effects is still unclear, in particular when it comes to subtle effects on neurobehaviors (Castoldi et al., 2008). It has been suggested that several factors including genetics, age, sex, health status, and nutritional factors may influence a population's vulnerability to the effects of methylmercury (US National Research Council, 2000). Two cohort studies investigating the effects of prenatal exposure to methylmercury revealed that although some children have been exposed to similar doses of methylmercury, children from the Faroe Islands have experienced subsequent neurotoxic effects (Debes et al., 2006; Grandjean et al.,



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1999), whereas children from the Seychelles have not (Davidson et al., 2011; van Wijngaarden et al., 2009). It has been hypothesized that methylmercury-induced toxicity on the developing brain could be greater following episodic high-level (peak) exposures (e.g. pilot whale meat in the Faroe Islands) than that resulting from average (continuous) exposures (e.g. ocean fish in the Seychelles) over the course of pregnancy, even though the average body burdens may be similar (Clarkson and Magos, 2006).

The possible effects of nutritional factors in modulating methylmercury metabolism and/or toxicity have been recognized by different authors (Castoldi et al., 2008; Chapman and Chan, 2000; Mahaffey, 1990; Peraza et al., 1998; Stokes-Riner et al., 2011). In animal studies, several food-derived antioxidants such as lycopene, proanthocyanidins and tea polyphenols have been identified as potential protective factors against toxicity induced by inorganic mercury (Augusti et al., 2007; Cavusoglu et al., 2009; Deng et al., 2012) and/or methylmercury (Liu et al., 2012; Yang et al., 2012). In humans, two studies carried out in the Brazilian Amazon revealed that fruit consumption provides a protective effect against methylmercury exposure (Passos et al., 2003, 2007). According to these authors, the soluble dietary fiber content as well as other prebiotic nutrients found in fruits could be interfering with methylmercury absorption in the gastrointestinal tract (Passos et al., 2007).

Exploratory analysis of our data collected among Inuit children attending childcare centers in Nunavik (northern Quebec, Canada) revealed negative correlations between blood mercury levels and consumption frequencies of several vegetables and fruits during the month prior to the interview (Turgeon O'Brien et al., 2010). Among these foods, tomato/vegetable juice and tomatoes were of interest because tomato products are the best dietary sources of lycopene (Murphy et al., 2012), a potent carotenoid antioxidant that is stable over long periods of storage (Agarwal et al., 2001). Additionally, tomato products are healthy, convenient, and relatively affordable in Nunavik.

The objective of the present study was to examine the effects of the consumption of tomato products on blood mercury levels in preschool-aged Inuit children. This study is part of the *Nutrition Program in Nunavik Childcare Centers* which includes both a nutrition intervention and a broad research project.

#### 2. Material and methods

#### 2.1. Study population and data collection

A study was conducted between 2006 and 2010 to document contaminantnutrient interactions and the effects of a nutrition intervention on the health of Inuit children attending childcare centers in Nunavik. The nutrition intervention of the *Nutrition Program* includes (i) a four-week cycle menu (breakfast, lunch, and mid-afternoon snack) that contains traditional foods upon availability and healthy store-bought foods, (ii) a nutrition policy, (iii) training sessions for cooks, educators, and directors, and (iv) educational activities for children. This paper covers crosssectional data collected among preschool Inuit children recruited in 2006–2008.

A convenience sample of villages was used with the priority given to the ones where the *Nutrition Program* was implemented in the childcare center for at least a few months. Three villages were included in the research project in 2006 (Inukjuak, Salluit, Kuujjuaq), then the project was expanded with the addition of more villages in 2007 (Kangiqsualujjuaq) and 2008 (Quaqtaq, Kangiqsujuaq, Ivujivik, Akulivik). Recruitment and data collection took place during the fall. Parents of Inuit children aged 1–4 years were contacted and invited to participate by an Inuk, usually from their own village. Those interested in the study were met by a research team member. Information on the study was given individually, orally or through a DVD available in three languages (Inuktitut, English, and French). Parents who agreed for their child to participate in the study gave their written consent. This study was reviewed and approved, prior to its conduct, by the Research Ethics Board of the Centre hospitalier universitaire de Québec, Québec (Québec), Canada.

#### 2.2. Measurement of total mercury in blood

For each participating child, a blood sample (6 ml) was collected by venipuncture in a vial containing ethylenediaminetetraacetic acid (EDTA) as the anticoagulant (BD 367863, Becton, Dickinson and Co., USA). Blood samples were either kept at room temperature for a maximum of 20 min, kept in an insulated container with ice packs, or refrigerated at 4 °C prior to processing. A sample of whole blood was aliquoted for mercury analysis and aliquots were frozen and stored at -18 °C/ -20 °C within 3 h of collection. Frozen aliquots were kept in insulated containers with ice packs during transportation to the Laboratoire de toxicologie of the Institut national de santé publique du Québec (INSPQ) where mercury analyses were performed.

Total mercury was determined in whole blood samples by inductively coupled plasma-mass spectrometry (ICP-MS; INSPQ method M-572). Blood samples were diluted in ammonium hydroxide and mercury was brought to its elementary state by passing through argon plasma before being selectively identified by mass spectrometry. All samples were analyzed on Perkin Elmer Sciex ICP-MS instruments and mercury was quantified on the DRC II. Limit of detection was 0.5 nmol/L (1 nmol = 0.20  $\mu$ g). Accuracy and precision were measured using reference materials from the QMEQAS scheme (Quebec multielement external quality assessment scheme). The coefficient of variation was 2.8% and the relative bias was nil for the mercury reference materials analyzed on 17 different days (consensus median value from participating laboratories = 30 nmol/L).

#### 2.3. Dietary assessment

Data on feeding practices during infancy and early childhood, including breastfeeding and the use of infant formula, were obtained by means of a questionnaire completed by a registered dietitian or a nurse with the parent or primary caregiver. Consumption frequencies (i.e. the number of eating occasions) of selected traditional foods (27 items) and store-bought foods (77 items) were collected by a registered dietitian using an adapted version of the food frequency questionnaire used in the 2004 Nunavik Inuit Health Survey (Blanchet and Rochette, 2008). It is well known that the consumption frequency of traditional foods generally varies by season (Blanchet and Rochette, 2008). Thus, to obtain children's usual traditional food intake, consumption was assessed by season for the year before the interview. For store-bought foods, a one-month period was used as this was assumed to be long enough to reflect the usual intake of participating children, particularly in light of the fact that Nunavik childcare centers offer a four-week cycle menu. Two food frequency questionnaires were completed for each participant, one with the parent or primary caregiver for the foods consumed at home, and one with childcare centers' cook and educators for the foods eaten at the childcare center.

Food consumption frequencies at the childcare center were adjusted for the child's length of attendance. Afterwards, total consumption frequency of each food was computed as follows: (i) for traditional foods, seasonal consumption frequencies were totaled first, then annual consumption frequencies at home and at the childcare center were summed to obtain the total consumption frequency, and (ii) for store-bought foods, we totaled the consumption frequencies at home and at the childcare center during the month before the interview. When a frequency was missing for one food (at home or at the childcare center, or for traditional foods, during any season), the total consumption frequency for this specific food was considered as a missing value. To assess the consumption frequency of tomato products, we summed up the consumption frequencies was missing, the consumption frequency of tomato products was considered as a missing value.

We also made the following adjustments for the participants from one village where we have been unable to meet the former cook and several educators in 2006. For all of the 26 participants, we filled the missing data for each traditional food item with the average annual consumption frequency in this childcare center between 2007 and 2009, adjusted for the child's length of attendance. For 23 of the 26 participants for whom we knew how often store-bought foods were served, but were unable to interview their educator to confirm their consumption, we assumed that children ate all the foods that were served by the cook, and we equalled children's consumption frequencies to the serving frequencies as reported by the cook. The decision concerning traditional foods was based on a cluster analysis examining the pattern of consumption between childcare centers, while the decision made on store-bought foods was based on our observation that educators often reported the same information for all children in their group. One reason for this could be that only one daily menu was available to children. The fact that data for these 26 participants were not missing at random prevented us from using the multiple imputation procedure presented below. For these participants, total consumption frequency of each food was computed as described above.

#### 2.4. Statistical analyses

Double data entry was performed and then statistical analyses were carried out using SAS 9.2 (SAS Institute, Cary, NC, USA). We included only participants having data from the food frequency questionnaire for both home and the childcare center. Among the 155 children recruited between 2006 and 2008, 31 were then excluded from analyses because food consumption frequencies were missing either at home (n = 19), at the childcare center (n = 3), or both (n = 9). Therefore, analyses were performed on 124 children, with 79 of them having a complete data set for the variables of interest.

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