Whole blood mercury and the risk of cardiovascular disease among the Greenlandic population

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ABSTRACT

Background: Studies have found mercury to be associated with cardiovascular disease (CVD), however, primarily in populations with low exposure. The highest levels, and variations in the levels, of whole blood mercury (WBM) worldwide have been found in Greenland. We prospectively assessed the association between WBM and the risk of developing CVD in the Greenlandic population.
Methods: We assessed the effects of WBM levels on incident CVD among 3083 Greenlandic Inuit, participating in a population-based cohort study conducted from 2005 to 2010. WBM was measured at baseline. Participants were followed in the National Patient Registries for Denmark and Greenland and in the causes of death register for CVD events from inclusion in the study until CVD event, emigration, death or end of follow-up (30/9–2013). Using Cox regression analyses, we calculated the incidence rates and the hazard ratio of CVD events according to WBM levels. Potential interactions with sex were also investigated.
Results: The highest levels of WBM were found in men, who had a significantly higher median level (19 µg/L (IQR:1–44)), compared with women (15 µg/L (IQR: 1–32), (p < 0.001)). The crude hazard ratio (HR) for incident CVD was 1.00 (95% CI 1.00–1.00) for 5 µg/l increase in WBM. After adjusting for several potential confounders, there was still no association between WBM and incident CVD (HR 0.99; 95%CI:0.99–1.00). We found no interactions with sex.
Conclusions: In a population with high levels of WBM, we found no association between WBM and the risk of developing CVD in Greenland.

1. Introduction

Cardiovascular disease (CVD) remains a crucial challenge for public health. The World Health Organization (WHO), estimated that 17.5 million people died from CVD in 2012 alone, accounting for 30% of all deaths globally and serving as the number one cause of death (WHO, 2016a).

WHO has suggested that CVD mortality could be reduced by decreasing or removing exposure to environmental pollutants, such as heavy metals (WHO, 2016a, b). Mercury is a global pollutant and is considered among the most toxic heavy metals (ATSDR, 2017). It has been associated with several adverse health outcomes and studies have shown that mercury may contribute to the development of CVD directly or indirectly by acting as a risk factor for associated diseases (Genchi et al., 2017). Exposure to mercury has been suggested to induce mitochondrial dysfunction, lower ATP-synthesis and increase phospholipid, protein and DNA peroxidation, resulting in vascular oxidative stress and inflammation. Mercury has also been suggested to inactivate paraoxonase, which causes dysfunctional HDL to reduce reverse cholesterol transport to the liver (Genchi et al., 2017). In addition, exposure to inorganic mercury caused β-cell dysfunction and apoptosis in murine models (Chen et al., 2006), and methylmercury blood levels were associated with hepatic insulin resistance in a cross sectional study (Chang et al., 2011).

In 1991, the Arctic Monitoring and Assessment Programme (AMAP) was established to monitor identified pollutants and their effect on the Arctic ecosystem. AMAP has in Greenland and other Arctic countries, systematically monitored whole blood mercury (WBM) levels, and within the Arctic, the highest levels of WBM have been found in Greenland (AMAP, 2015). The main source of mercury exposure in the Arctic is from fish and marine mammal consumption. These food sources are also high in selenium, which has been suggested to

counterbalance some of the adverse effects of mercury (Genchi et al., 2017)

Health effects of mercury in Greenland nonetheless, have only been assessed in a few studies; in a cross-sectional study, there was no association between systolic blood pressure and WBM in Greenland, and the prevalence of hypertension actually decreased with increasing WBM concentrations for men (Nielsen et al., 2012). In another cross-sectional study, a weak association between WBM and glucose intolerance and type 2 diabetes among adult Inuit in Greenland was found (Jeppesen et al., 2015).

Some longitudinal studies of mercury exposure and CVD have been conducted. Thus, higher concentrations of mercury in the hair was associated with CVD among men in Finland, whereas higher levels of serum mercury were associated with a lower risk of acute myocardial infarction among Swedish women (Virtanen et al., 2005; Bergdahl et al., 2013). However, longitudinal studies of the association of mercury exposure and risk of CVD are generally lacking, particularly in highly exposed populations such as the Greenlandic population. We aimed to explore the association between WBM and the risk of developing CVD among the Greenlandic population.

2. Materials and methods

We conducted a prospective cohort study of 3350 Greenlandic Inuit enrolled in the population-based study '*Inuit Health in Transition Greenland survey 2005–2010*' (IHIT) in Greenland (Bjerregaard, 2011). We followed all participants in National registries from inclusion in the IHIT study until CVD event, emigration, death or end of follow-up (30/9–2013).

Adult Greenlandic residents (18 + years of age) were randomly selected from the civil registration system from 2005 to 2010, to be included in IHIT. Participants were sampled from 9 towns and 13 villages.

Prior to the IHIT study, a pilot study was conducted in 2003 in three villages, in Ilulissat on the West coast of Greenland. For the present study, all participants from the IHIT study and the pilot study with a valid measurement of WBM and with no history of CVD at enrolment, were included.

2.1. Data collection

All participants in the IHIT and the pilot study underwent a questionnaire interview and clinical examinations.

All blood samples were drawn by venepuncture at normal venous pressure. We measured selenium and total mercury in whole blood, stored at − 20 °C and analysed using inductively coupled mass spectrometry, in the laboratoire de Toxicologie/INSPQ, Sainte-Foy, Quebec, Canada. The detection limit for mercury was 0.10 µg/l and 7.9 µg/l for selenium (Bjerregaard, 2011). We termed total mercury measured in whole blood as whole blood mercury (WBM).

HbA_{1c} was assessed using high-pressure liquid chromatography (HPLC;Tosoh G7;Roche Diagnostics) (Bjerregaard, 2011), measured in % and converted to mmol/mol by calculation. Total cholesterol, HDL-cholesterol and triglycerides were assessed using enzymatic colorimetric tests using Hitachi 917 (Bjerregaard, 2011). Based on total cholesterol, HDL-cholesterol and triglycerides, LDL-cholesterol was estimated using Friedewald's formula. N-3 fatty acids were measured as the composition of phospholipids of erythrocyte membranes after total lipid extraction, using a DB-23 column in HP-Packard GC chromatograph. We then calculated the Omega-3 index from the fraction of eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) in the erythrocyte membrane. Height and weight was measured with the participant stripped to underwear and socks. Weight was measured on a standard electronic clinical scale and BMI was calculated. Waist circumference was measured midway between the rib cage and the iliac crest.

Blood pressure was measured on the right arm of the sitting participant after five minutes of initial rest, using an automatic measuring device (Kivex UA-779). Blood pressure was measured three times with at least one-minute interval. The two last measurements were averaged for the analysis (Bjerregaard, 2011).

Questionnaire derived measures included place of residence (village/town), education (student/completed primary school/completed high school/mid-level or long education (university)) and smoking (daily smoking/not daily smoking/non-smoker).

Physical activity was measured using a modified and validated version of the interviewer-administered seven-day International Physical Activity Questionnaire (IPAQ) (long version) (IPAQ-L) (Dahl-Petersen et al., 2013). Physical activity energy expenditure (PAEE) was calculated by multiplying time reported (minutes/week) by the net metabolic cost of each activity, which was expressed in metabolic equivalents. Age and sex was derived from the civil registration system.

2.2. CVD

All citizens of Greenland are assigned a unique personal identification number at birth (PIN), which enables tracing of all study participant persons through public registries.

We followed the participants in the Greenland National Patient Register, the Danish National Patient Register and in the Greenlandic Causes of Death Register, for incident CVD or death from CVD after enrolment in IHIT or the pilot study.

Based on ICD-10 coding, all deaths or hospital admissions from ischemic heart disease (I20–25), atrial fibrillation (I48), heart failure (I50), cerebrovascular diseases (I60–69) and vascular diseases (I70–79) were categorized as a CVD event. Both primary and secondary hospital diagnoses were included and the underlying and contributory causes of death. All CVD diagnoses identified in the registries among the study participants were validated by two medical doctors (Tvermosegaard et al., 2018).

2.3. Statistical analysis

We assessed the distribution of all baseline characteristics by sex due to sex-differences in both WBM levels and CVD incidence. We tested for differences using Chi2 for categorical and binary variables and using T-test for continuous variables.

Using Cox regression, we modelled the continuous effects of WBM levels on incident CVD in both univariate and multivariate analyses, using age as the underlying time scale. We calculated the overall incidence rates and the hazard ratio (HR) of a 5 µg/l increase in WBM on the risk of first CVD event after entry in the study. Participants entered the analyses at date of examination in IHIT or the pilot study and contributed risk time until date of CVD diagnosis/death, emigration, death from other causes or end of follow-up 30/09–2013.

We tested the exposure variable for deviations from linearity by including the quadratic, cubic and logarithmic transformation of the continuous variable in the analyses. No deviation from linearity was found for WBM and it was kept in its original form. Total cholesterol, HDL-cholesterol, LDL-cholesterol and triglycerides were inter-correlated. Total cholesterol was hence removed from the final analyses.

We selected potential confounders based on previous studies as well as availability in the dataset and adjusted for these in the regression analyses in various levels of adjustment; in model 1, we assessed the crude association between WBM and CVD. The second model was adjusted for age and sex. In addition to age and sex, the third model also comprised PAEE, selenium, place of residence and education. PAEE and selenium were included as continuous variables.

In addition to the third model, the fourth model additionally included smoking and HDL-cholesterol, calculated LDL-cholesterol, triglycerides, Omega-3 index, BMI, waist circumference, systolic/diastolic blood pressure and HbA_{1c} as continuous variables.