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**Title: Early exposure to mercury and cardiovascular function of seven-year old children in Guadeloupe (French West Indies)**

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**Running head:** Mercury exposure and cardiac function in 7-year-olds

## 1 Abstract

## 2 Background

3 The cardiotoxicity of prenatal exposure to mercury has been suggested in populations having  
4 regular contaminated seafood intake, though replications in the literature are inconsistent.

## 5 Methods

6 The Timoun Mother-Child Cohort Study was set up in Guadeloupe, an island in the Caribbean  
7 Sea where seafood consumption is regular. At seven years of age, 592 children underwent a  
8 medical examination, including cardiac function assessment. Blood pressure (BP) was taken  
9 using an automated blood pressure monitor, heart rate variability (HRV, 9 parameters) and  
10 electrocardiogram (ECG) characteristics (QT, T-wave parameters) were measured using Holter  
11 cardiac monitoring during the examination. Total mercury concentrations were measured in  
12 cord blood at birth (median = 6.6  $\mu\text{g/L}$ , N=399) and in the children's blood at age 7 (median =  
13 1.7  $\mu\text{g/L}$ , N=310). Adjusted linear and non-linear modelling was used to study the association  
14 of each cardiac parameter with prenatal and childhood exposures. Sensitivity analyses included  
15 co-exposures to lead and cadmium, adjustment for maternal seafood consumption, selenium  
16 and polyunsaturated fatty acids (n3-PUFAs), and for sporting activity.

## 17 Results

18 Higher prenatal mercury was associated with higher systolic BP at 7 years of age ( $\beta_{\log 2} = 1.02$ ;  
19 95% Confidence Interval (CI) = 0.10, 1.19)). In boys, intermediate prenatal exposure was  
20 associated with reduced overall HRV and parasympathetic activity, and longer QT was  
21 observed with increasing prenatal mercury ( $\beta_{\log 2} = 4.02$ ; CI= 0.48, 7.56). In girls, HRV tended  
22 to increase linearly with prenatal exposure, and no association was observed with QT-wave  
23 related parameters. Mercury exposure at 7 years was associated with decreased BP in girls ( $\beta_{\log 2}$   
24 = -1.13, CI= -2.22, -0.004) for diastolic BP). In boys, the low/high-frequency (LF/HF) ratio  
25 increased for intermediate levels of exposure.

1 **Conclusion**

2 Our study suggests sex-specific and non-monotonic modifications in some cardiac health  
3 parameters following prenatal exposure to mercury, in pre-pubertal children from an insular  
4 fish-consuming population.

5

6 **Keywords:** Mercury, blood pressure, heart rate variability, children, prenatal exposure,  
7 cardiotoxicity, electrocardiogram, QT interval, QT-peak, T-wave, T-peak-to-end

8

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## DECLARATIONS

**Ethics approval and consent to participate:** The study was approved by the relevant ethical committee for studies involving human subjects (Comité de Protection des Personnes Sud-Ouest et Outremer III; n° 2011-AOOSI--40). Each participant provided written informed consent.

**Availability of data and materials:** Due to ethical concerns, supporting data cannot be made openly available. The Timoun team can provide the data on request, subject to appropriate approvals. Please contact the corresponding author to application.

**Competing interests:** The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this article.

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### Individual contributions:

**Nathalie Costet:** Methodology; Software; Formal analysis; Visualization; Writing – original draft. **Matthieu Doyen:** Data curation; Formal analysis; Software; Writing – original draft.

**Florence Rouget:** Investigation; Validation; Writing – review & editing. **Léah Michineau:** Investigation; Data curation; Writing – review & editing. **Christine Monfort:** Investigation; Data curation; Writing – review & editing. **Ciprian-Mihai Cirtiu:** Methodology; Validation; Writing – review & editing. **Philippe Kadhel:** Conceptualization, Funding acquisition; Project administration; Writing – review & editing. **Luc Multigner:** Conceptualization; Funding acquisition; Project administration; Writing – review & editing. **Patrick Pladys:** Conceptualization; Validation; Writing – review & editing. **Sylvaine Cordier:** Conceptualization; Funding acquisition; Supervision; Writing – original draft.

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**LIST OF ACCRONYMS**

- 1 AIC: Akaike Information Criterion (estimator of prediction error)
- 2 BMI: Body Mass Index
- 3 CDC: Center for Disease Control and Prevention (USA)
- 4 GAM: Generalized Additive Model
- 5 HTA: Hypertension (High blood pressure)
- 6 LOD: limit of detection
- 7 MeHg: methyl-mercury
- 8 PCA: Principal Component Analysis
- 9 n3-PUFAs: Poly Unsaturated Fatty Acids (omega-3)
- 10 SGA: Small for Gestational Age (10<sup>th</sup> percentile)
- 11
- 12 **Heart function indicators**
- 13 (see S-Table 1 and S-Figure 1 for definitions)
- 14 AETP: Approximate Entropy (heart rate variability)
- 15 ANS: Autonomic Neural System
- 16 BP: Blood Pressure
- 17 bpm: Beats per minute
- 18 DBP: Diastolic Blood Pressure
- 19 DFA1: Detrended short term Fluctuations
- 20 DFA2: Detrended long term Fluctuations
- 21 ECG: ElectroCardiogram
- 22 HF: HRV frequency measurement (High Frequencies: 0.15–0.4 Hz)
- 23 HR: Heart Rate
- 24 HRV: Heart Rate Variability
- 25 LF: HRV frequency measurement (Low Frequencies: 0.04–0.15 Hz)
- 26 QRS: Duration between Q and S waves (ECG)
- 27 QT: Duration between Q and T waves (ECG)
- 28 QTc: QT corrected (Bazett) for heart rate
- 29 QTp: Duration between Q wave and peak of the T-wave
- 30 QTpc: QTp corrected (Bazett) for heart rate
- 31 RMSSD: Root Mean Square of the Successive Differences of RR intervals
- 32 RR: Duration between 2 successive heartbeats (ECG)

- 1 SBP: Systolic Blood Pressure
- 2 SDNN: Standard Deviation of the normal R-R intervals
- 3 SETP: Sample Entropy (heart rate variability)
- 4 Tamp: Amplitude of the T wave (ECG)  
TpTe: Duration between Peak and end of the T wave (ECG)
- 5

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## 1 1. INTRODUCTION

2 Mercury (Hg) neurotoxicity has been demonstrated following massive poisoning outbreaks in  
3 the populations of Minamata Bay and Iraq, as well as in populations with a lower chronic  
4 environmental exposure to mercury as a result of regular and high consumption of contaminated  
5 seafood [1,2]. In-utero mercury exposure has been associated in particular with developmental  
6 neurotoxicity [3]. Mercury (inorganic, or organic as methylmercury (MeHg)) crosses the  
7 placenta easily [4], and because fetal blood has larger hematocrit and higher concentrations of  
8 hemoglobin and plasma albumin, it accumulates 30–70% more MeHg than maternal blood does  
9 [5–8], so that the developing fetal brain is more exposed. The Autonomic Nervous System  
10 (ANS), which regulates cardiovascular, respiratory and gastrointestinal functions in response  
11 to environment, undergoes a critical period of maturation during the second half of gestation  
12 and the early neonatal period [9]. Adverse prenatal and neonatal conditions and exposures  
13 during these critical periods can impair or alter its development [10].

14 The cardiotoxic effect of prenatal exposure to MeHg in relation to child blood pressure and  
15 cardiac autonomic function has been investigated in a number of prospective studies across the  
16 world, with mixed results [11–13]. One reason for these discrepancies may be the different  
17 exposure levels in various studies, in relation to the putative shape of the dose-response curve.  
18 One early study suggested that the risk of impairments to the cardiac function of children (such  
19 as increased blood pressure (BP) or decreased heart rate variability (HRV)) increased linearly  
20 with mercury exposure for levels of exposure below 10 $\mu$ g/L in cord blood, but not thereafter  
21 [14]. However, in subsequent epidemiological studies, the shape of the dose-response  
22 relationship has rarely been explored.

23 A survey of MeHg exposure from fish or seafood consumption in populations throughout the  
24 world [15] ranked populations as follows: the highest exposure was observed in communities  
25 living next to gold mining sites, followed by the arctic regions (i.e. Faroe Islands, Canada), then

1 in populations relying on self-caught local waterways, those living in coastal areas/island-  
2 dwelling (i.e. Seychelles islands) and populations living inland (Europe, USA). A survey of  
3 pregnant women from 10 Caribbean countries reported mercury blood concentrations two times  
4 higher than in U.S and Canada [16]. These concentrations were related to consumption of large  
5 predatory fishes such as swordfish, marlin, and shark known as containing high levels of MeHg.  
6 In Guadeloupe, one island in the Caribbean Sea, inhabitants regularly eat large quantities of  
7 seafood (estimated at 35-37 kg/habitant/year), mostly smaller predatory fishes such as snapper,  
8 dolphinfish, catfish, grouper or tuna, known as moderately contaminated by methylmercury  
9 [17–19].

10 In 2004, a mother-child cohort (Timoun) was established there to investigate the consequences  
11 of prenatal and childhood exposure to pesticides and other environmental pollutants (including  
12 heavy metals) for pregnancy and child development [20]. The aim of the present study was to  
13 evaluate the role of prenatal and childhood exposure to mercury on child cardiovascular health,  
14 including blood pressure, heart rate variability (HRV) and QT-wave related parameters at seven  
15 years, with a particular interest in the shape of the dose-response relationship and sex-specific  
16 associations.

## 17 **2. METHODS**

### 18 **2.1. Study participants**

19 This study was conducted within the Timoun Mother Child Cohort, which enrolled 1,068  
20 pregnant women from the general population in Guadeloupe (French West Indies, a Caribbean  
21 archipelago) between November 2004 and December 2007. Women were recruited in the  
22 course of third-trimester appointments at public and private health centers (University Hospital  
23 of Guadeloupe, General Hospital of Basse-Terre, Polyclinic of Guadeloupe, and antenatal care  
24 dispensaries). [20] At inclusion, women were interviewed by trained midwives to assess their  
25 medical history, socioeconomic conditions, and dietary habits. At delivery, medical data on the

1 pregnancy and newborn health status data were collected by the medical staff, and maternal and  
2 cord blood samples were obtained. In a selected sub-cohort, follow-up appointments were made  
3 for the children at three months, seven months, and eighteen months of age. [21–23] At seven  
4 years of age (May 2011-October 2015), all mothers initially included in the cohort were invited  
5 to a follow-up interview and a medical examination of their child at the University Hospital of  
6 Guadeloupe. 592 (55.4%) of the initial cohort brought their children to be examined. The  
7 medical examination included anthropometric measurements, Tanner stage assessment, blood  
8 pressure measurements, a Holter cardiac monitoring, hearing and functional respiratory tests,  
9 neuro-developmental tests and the collection of blood and urine samples. During the  
10 examination, face-to-face interviews were also conducted with the mothers, to collect  
11 information about the socio-economic context in which the child was being raised  
12 (environment, lifestyle, dietary habits, etc.) and the health of the child and his/her parents. For  
13 the present study, we excluded five children because they had medical conditions that could  
14 impact cardiac parameters (two with Down syndrome, one with leukemia, and two with a severe  
15 cardiac pathology) and 186 children for whom no cord blood was available, so that 401 children  
16 were eligible for our study.

17

## 18 **2.2. Cardiac function procedures and measurements**

19 The definition of cardiac function parameters used in this study are summarized in S-Table 1  
20 and S-Figure 1.

### 21 **2.2.1. Blood pressure**

22 Systolic (SBP), diastolic (DBP) and mean blood pressures were measured twice (at the  
23 beginning and end of the examination) with the child seated, using an automated blood pressure  
24 monitor (Dinamap CARESCAPE V 100, GE Medical Systems Information Technology, Inc.

1 Milwaukee, WI, USA) with an appropriate pediatric cuff. The first measurement was taken  
2 just after a period of standing followed by 3 minutes sitting (corresponding to the Holter  
3 monitoring set up), and the second was taken in the sitting position, following a long (around  
4 20 minutes) period of calm in the sitting position, while the questionnaire was administered to  
5 the mother. The minimum of both measurements was retained, to estimate the child's SBP and  
6 DBP at 7 years of age. The measurements for one child were missing, and those of another were  
7 inconsistent. In the end, 399 children with reliable BP measurements were included in the BP  
8 analyses.

### 9 **2.2.2. Heart Rate Variability**

10 A SpiderView 12-lead Digital Holter Monitor (SORIN, Milan, Italy) with high temporal  
11 resolution (1000Hz) was used to record the electrical activity of the heart (referred to as an  
12 electrocardiogram (ECG)) throughout the examination for a median length of 51 minutes  
13 (Q1=44 min, Q3=59 min). During the recording, children were subjected to the same  
14 examinations and tasks as described in Grandemange et al.(Fig. S1). [24]

15 The time intervals (RR intervals, in ms) between consecutive heart beats (QRS complex) were  
16 derived from the ECG records using a Multi-Feature Probabilistic QRS Detector [25] during a  
17 calm 20-min period in which the child was at rest, quiet, and seated during the mother's  
18 interview. QRS detections were first verified and manually corrected by expert clinicians, then  
19 the most stationary 5-min RR sequence without ectopic beats was selected to derive mean heart  
20 rate (HR) in beats per minute (bpm) and Heart Rate Variability (HRV) indices. In line with  
21 current guidelines [26], three types of HRV measurements were computed: time-domain,  
22 frequency-domain and non-linear measurements. The time domain indicators included:  
23 standard deviation of the cardiac cycle length (SDNN, representing global HRV, in ms) and  
24 root mean square of the successive difference of RR intervals (RMSSD, representing vagally  
25 mediated and respiratory related HRV, in ms). The frequency-domain analysis was performed

1 on RR series resampled at 4 Hz and power spectrum measured using an autoregressive Burg's  
2 lattice-based method with an order of  $p=12$ . The frequency variables studied were obtained  
3 using two frequency bands: (i) between 0.04–0.15 Hz for the Low Frequency (LF), which is  
4 known to be mainly determined by blood pressure ripples and to mainly reflect baroreceptor  
5 activity during resting conditions [27]; (ii) 0.15–0.4 Hz for the High Frequency (HF), which is  
6 known as the respiratory band because it reflects parasympathetic activity related to the  
7 respiratory cycle [27]; (iv) LF/HF ratio was used as an indicator of sympatho-vagal balance. A  
8 lower LF/HF ratio reflects a parasympathetic dominance, while a higher LF/HF ratio suggests  
9 a sympathetic dominance. There is however some debate in the literature about the  
10 interpretation of this ratio [28] that may also be dependent on measurement conditions [27].  
11 HRV regulation results from the dynamic and complex interactions between multiple  
12 physiological mechanisms involving non-linear contributions [29]. Thus, the use of non-linear  
13 indices can lead to a better understanding of HRV dynamics, reflecting the unpredictability of  
14 variations in the heart beat cycle, due to the complexity of the HRV mechanisms [27]. Various  
15 studies have reported their better discriminant power than linear methods [30–33]. Four  
16 commonly-used non-linear analyses were performed. To measure the complexity and regularity  
17 of the signal [34], we computed the Approximate and Sample Entropies (AETP and SETP): a  
18 low value indicates high fluctuation predictability. Detrended fluctuation parameters, which  
19 measure correlations between successive RR intervals of different lengths, were also calculated:  
20 DFA1 represents short term fluctuations (within 4-16 beats) and DFA2 long-term fluctuations  
21 (within 16-64 beats) [35].

22 HRV parameters were derived for 426 (72%) of the 592 children participating in the medical  
23 examination. Owing to technical problems in transmission, compression of ECG records,  
24 misidentification of the child or impossible HRV parameter computation as a result of

1 significant noise perturbation in signals (or abnormal duration), we were left with reliable HRV  
2 parameters and mercury measurement in cord blood available for 300 children.

### 3 **2.2.3. QT-wave related parameters**

4 The QT interval (S-Figure 1) measures the duration of the ventricular depolarization (Q-wave  
5 onset) -repolarization (end of the T-wave) cycle. An abnormal value (prolonged QT interval) is  
6 often associated with an electrophysiological environment that is conducive to the development  
7 of cardiac arrhythmias [36]. Q-wave and T-wave delineations were performed on the ECG at  
8 rest, using a fully automatic segmentation method based on wavelet transform decomposition  
9 in combination with an evolutionary algorithm designed to optimize both the parameters and  
10 the threshold of the decision process [37]. The mean QT interval was then calculated in the  
11 most stationary 5-min RR sequence, where Q and T-wave detections were considered reliable  
12 by an expert clinician. Because the QT interval shortens with physiologic decreases in the RR  
13 interval, a heart rate correction was made to the QT interval, using the classical Bazett  
14 correction formula ( $QTc = \frac{QT}{\sqrt{RR}}$ ) [38]. The mean value of the duration between Q onset and  
15 T peak was also computed (QTp), as was the QTpc (with Bazett correction). In addition, we  
16 computed the indicators of the T-wave (repolarization phase) morphology: its amplitude ( $T_{amp}$ ),  
17 and interval from T-wave peak to end (TpTe), which represents the repolarization dispersion  
18 [39]. These additional features are useful in characterizing repolarization abnormalities.  
19 Ultimately, 266 children having both reliable QT characteristics and available cord blood  
20 mercury measurement were included in the analyses.

21 The flowchart presented in S-Figure 2 summarizes final study sample selection.

22

### 23 **2.3. Exposure to mercury, and co-exposures**

1 Samples of cord blood and child blood taken at the medical examination at 7 years of age were  
2 collected in EDTA tubes to document prenatal and childhood exposure to mercury, and co-  
3 exposure to some other environmental contaminants (lead and cadmium) as well as to nutrients  
4 such as selenium and omega3 polyunsaturated fatty acids (n-3 PUFAs). Blood samples were  
5 stored at  $-30^{\circ}\text{C}$  before their transfer on dry ice by airmail to the Centre de Toxicologie du  
6 Québec laboratory (Institut National de Santé Publique du Québec, Québec, Canada). Total  
7 mercury (Hg), lead (Pb), cadmium (Cd), and selenium (Se) concentrations were quantified in  
8 whole blood cord and child blood, using inductively-coupled plasma mass spectrometry (ICP-  
9 MS). To determine Hg, blood samples were diluted 20-fold in a solution containing ammonium  
10 hydroxide prior to analysis. The cord blood LOD values were  $0.1\ \mu\text{g/L}$  for Hg,  $0.21\ \mu\text{g/L}$  for  
11 Pb,  $0.04\ \mu\text{g/L}$  for Cd, and  $7.87\ \mu\text{g/L}$  for Se. The child blood LOD values for Hg, Pb, Cd, and  
12 Se were  $0.4\ \mu\text{g/L}$ ,  $2\ \mu\text{g/L}$ ,  $0.1\ \mu\text{g/L}$ , and  $20\ \mu\text{g/L}$ , respectively. Concentrations below LOD were  
13 present for Hg in child blood (5%) and Cd in cord blood (47%). For Hg, we imputed them by  
14 values under LOD randomly selected from a left-censored log-normal distribution estimated  
15 using likelihood maximum approach [40], and we categorized cord blood Cd concentrations  
16 into three levels (<median, third quartile, fourth quartile). The serum phospholipid fatty acid  
17 composition was assessed using gas chromatography, according to the method described in  
18 Chajès et al. (2008) [41]. The n-3 PUFAs levels were expressed as percentage of total fatty  
19 acids.

#### 20 21 **2.4. Statistical analysis**

22 Concentrations of cord and child blood mercury, lead and selenium were considered as  
23 continuous and  $\log_2$ -transformed for the analyses because of skewed distributions. Owing to an  
24 insufficient detection rate (53%), Cd concentrations were categorized into three levels (below  
25 LOD, plus two upper categories, split at the median of the detected values).

1 Cardiac function indicator distributions were also visually checked for normality, and  
2 logarithmic transformation was applied when skewness was significant (SDNN, RMSSD, LF,  
3 HF, LF/HF, and TpTe). Associations between cardiac health indicators were examined using  
4 their Spearman correlation matrix and a Principal Components Analysis (PCA). Their  
5 bivariate associations with maternal and child characteristics were tested by linear regression  
6 modeling (anova and t-tests). We used sex-, age-, and height-specific z-scores for both  
7 diastolic and systolic blood pressure to identify children with (pre-) hypertension (i.e. those  
8 having a DBP or/and SBP above the 90th or the 95th percentiles of reference distribution,  
9 respectively) using the *childsds* R package with CDC-references for height z-scores [42].

10 We used multivariate linear regression modeling to assess the association between prenatal and  
11 current exposure to Hg (log<sub>2</sub> scale) and each child cardiac outcome separately. Non-linear  
12 associations were modeled with Generalized Additive Models (GAM), including restricted  
13 cubic splines with automatic smoothness estimation by cross-validation (*mgcv* R package) [43]  
14 and represented in graphs with 3 nodes pre-positioned at the 10<sup>th</sup>, 50<sup>th</sup> and 90<sup>th</sup> percentiles of  
15 the exposure distribution [44]. Non-linear associations were checked using several criteria: the  
16 estimated degree of freedom was > 1.5, the smooth term for exposure was significant (p<=0.10),  
17 the Likelihood Ratio test comparing the fit statistics (AIC) of linear models *versus* GAM was  
18 significant (Khi<sup>2</sup> test, p≤ 0.10), and graphical examination of the GAM plots. Because of the  
19 log<sub>2</sub> transformation of the exposure variable, variations in cardiac parameters were estimated  
20 for each doubling of the exposure level. If the cardiac parameter was log-transformed, effect-  
21 sizes were given as 10<sup>β</sup> and interpreted as multiplicative effects. For non-transformed cardiac  
22 parameters, they were given as β and interpreted as additive effects. Models were run for  
23 prenatal exposure and at 7 years of age separately. These were adjusted a priori for covariates  
24 known to be associated with childhood blood pressure, HRV or QT indicators and with mercury  
25 exposure (confounders) or associated with cardiometabolic health (precision variables). They



1 included child sex, exact age at measurement (in months), height, BMI, birth weight, maternal  
2 hypertension and smoking during pregnancy, maternal educational level and geographical  
3 origin of the child's maternal grandparents (as a proxy for ethnic origin).

4 For HRV parameters, we also adjusted the models for HR, to take into account its strong  
5 influence on HRV parameters and thereby allow for assessment of the specific effect of  
6 exposure on cardiac autonomic regulation. [45,46]

7 A sex stratification was a priori applied because of known differences between boys and girls  
8 in certain cardiac function parameters [47] and differential susceptibility to exposure to heavy  
9 metals reported in epidemiological and experimental studies [4]. We also formally tested a sex-  
10 exposure interaction term in the multivariate linear models with a significance threshold of 0.10.

11 We then conducted the following sensitivity analyses: First, we ran models mutually adjusted  
12 for prenatal and current exposures, to explore their relative contribution in the association with  
13 cardiac health. Further analyses were also adjusted for prenatal co-exposure to lead, cadmium,  
14 and nutrients (Se and n-3 PUFAs), and for maternal seafood intake to take into account common  
15 sources of exposures and similar or reverse expected effects compared to mercury [48,49]. We  
16 also tested additional adjustment for physical activity (3 categories: no sport, <2hrs/week, >=  
17 2hrs/week), as this is expected to impact heart function [47,50]. Finally, we ran analyses without  
18 adjusting for birth weight, because birth weight may be in the causal pathway between prenatal  
19 exposure to mercury and cardiac function. [10,51]

20 The analyses were computed using SAS® 9.4 and R (package mgcv for the GAM analyses).

21

## 1 **3. RESULTS**

### 2 **3.1. Description of the population under study**

3 Table 1 presents the characteristics of the mothers and children included in the various analyses.  
4 Children from the Timoun cohort participating in the follow-up at 7 years of age were mostly  
5 (87%) born to mothers of Caribbean origin (French West Indies or other Caribbean islands)  
6 (Table 1). They were examined at an average age of 7 years and 8 months (minimum = 7 years,  
7 maximum = 8 years). Their mean BMI z-score was 0.3 (SD=1.6), and around 17% of them were  
8 overweight and 10% obese. All attended school, and 45% declared practicing a sport outside of  
9 school (weekly median practice time = 2 hours). 13% of the children had been born preterm.  
10 Approximately 40% of their mothers were overweight or obese pre-pregnancy, and 12% had  
11 hypertension (pre-existing or gestational). Smoking tobacco during pregnancy was reported in  
12 up to 5% of the mothers, and alcohol consumption by less than 3%. Exposure to passive  
13 smoking during childhood (by the mother or other persons living with the child) was reported  
14 in 20-23% of children. The three sub-samples used for the BP, HRV and QT-wave related  
15 parameters, respectively, did not differ substantially in terms of covariates distributions.  
16 However, the QT sub-sample included less obese mothers but more overweight mothers than  
17 the other sub-samples. The BP sub-sample included less highly educated mothers, less smokers  
18 during pregnancy, less children obese and exposed to passive smoking at 7 years.  
19 Compared to the initial Timoun cohort, children participating in the 7 year-follow with cardiac  
20 function and Hg cord blood measurements available were less often born preterm or SGA, and  
21 their mothers were more educated and fewer to declare themselves as smokers during  
22 pregnancy. No differences were observed regarding maternal BMI and hypertension or other  
23 characteristics (S-Table 1).

24

### 25 **3.2. Cardiac function at 7 years of age**

1 Distribution statistics for BP, HRV and QT parameters are presented in Table 2, and their  
2 correlations are shown in S-Figure 3. The mean heart rate (HR) was 85.1 (IQR: 77.6-93.6),  
3 within the normal range for children aged 7 years [52]. Seventeen children (4.3%) were  
4 classified as pre-hypertensive and 9 (2.3%) as hypertensive.

5 HR was significantly correlated with almost all of the cardiac function parameters (S-Figure 3).  
6 Blood pressure was weakly correlated with HR ( $R=0.13$  and  $R=0.07$  respectively for DBP and  
7 SBP) and with the HRV and QT parameters (all  $R < 0.15$ ). Within the HRV parameters, SDNN,  
8 RMSSD, LF and HF were highly mutually correlated ( $R > 0.90$ ). Overall, QT intervals (QTc,  
9 QTpc) and T-wave amplitude were moderately correlated with HRV parameters (R coefficients  
10 around 0.20) (S-Figure 3)

11 Mean HR was significantly higher in girls (86.8 bpm (IQR: 79.7-94.6) vs. 82.5 bpm (IQR: 76.0-  
12 91.9) in boys, Wilcoxon test  $p=0.004$ ). SBP and DBP were similar in boys and girls, and  
13 significantly increased with child height, overweight and obesity, and maternal hypertension  
14 (S-Table 2). None of the HRV and QT-wave related parameters differed significantly between  
15 boys and girls, except the DFA2 and the T-wave amplitude (Tamp) which were significantly  
16 lower in girls. Associations between other maternal and child characteristics with HRV and  
17 QT-wave related parameters are presented in S-Table 3.

18

### 19 **3.3. Exposure**

20 The description of prenatal and child exposure to Hg and other metal contaminants is shown in  
21 Table 1; their correlations are shown in S-Table 4.

22 The median value of the Hg concentration was 6.6  $\mu\text{g/L}$  (Q1=4.4  $\mu\text{g/L}$ ; Q3=9.2  $\mu\text{g/L}$ ) in cord  
23 blood and 1.7  $\mu\text{g/L}$  (Q1=1.0  $\mu\text{g/L}$ ; Q3=2.8  $\mu\text{g/L}$ ) at 7 years. In cord blood, Hg was correlated  
24 with Se (Spearman  $R=0.41$ ) but not with Pb (Spearman  $R=0.02$ ). Hg concentrations in cord  
25 blood and in blood at 7 years of age were significantly (though modestly) correlated (Spearman

1 R=0.17). At 7 years, Hg levels were inversely correlated with Pb levels (Spearman R= -0.11).  
2 No significant differences between the sexes were observed in Hg concentrations in cord blood  
3 (t-test, p= 0.11) or in blood at 7 years (t-test, p=0.09). Children born to women having  
4 Caribbean ascendants had significantly higher levels of Hg in cord blood (Wilcoxon test,  
5 p=0.04), though not at 7 years of age (Wilcoxon test, p=0.62) (data not shown).  
6 Mothers reported a median seafood consumption frequency of 3.4 times a week (IQR= 1.9-  
7 5.6). Their median daily intake of fish was estimated at 73 g/day (IQR=32-159), including  
8 32g/day (IQR=0-80) of pelagic fish. The fish species most commonly consumed were snapper  
9 (55% of the women), dolphinfish (54%), parrotfish (35.4%), grouper (22.8%) and tuna (20.3%)  
10 (data not shown).

#### 11 **3.4. Association between cord blood mercury and cardiac function at 7 years**

12 The **mean HR** and **DBP** at 7 years were not significantly associated with mercury cord blood  
13 concentration. The **SBP** increased linearly for a doubling of cord mercury ( $\beta_{\log 2} = 1.02$  (0.10;  
14 1.93)), more markedly in girls ( $\beta_{\log 2} = 1.28$  (0.00; 2.57)) (Table 3, Figure 1). None of the **HRV**  
15 **indicators** were significantly linearly associated with cord blood mercury, when considering  
16 both sexes together, but interaction with sex was significant for RMSSD (p=0.02), SDNN  
17 (p=0.03), HF (p=0.03) and LF (p=0.09). After stratification by sex, RMSSD increased by 18%  
18 with doubling exposure in girls ( $\beta_{\log 2} = 1.18$  (1.00; 1.40)). A similar (non-significant) trend was  
19 observed for SDNN, HF, and LF. In boys, significant non-linear (U-shaped) associations were  
20 detected using GAM modeling for RMSSD, SDNN, and HF, indicating lower heart rate  
21 variability for intermediate levels of prenatal exposure (Table 3, Figure 1).

22 None of the **QT-wave related parameters** were linearly correlated with cord blood mercury  
23 when considering both sexes, but significant interactions with sex were found for QTc and  
24 QTpc ( $p_{\text{inter}}=0.06$  and 0.03, respectively). In boys, QTc and QTpc increased linearly with

1 doubling cord blood mercury ( $\beta_{\log 2} = 4.02$  (0.48; 7.56) for QTc and  $\beta_{\log 2} = 3.62$  (0.04; 7.21) for  
2 QTpc). In girls, no significant associations were found (Table 3, Figure 2).

3

### 4 **3.5. Association between blood mercury at 7 years and cardiac function at 7 years**

5 We found no association between **mean HR** and mercury blood concentration at 7 years of age  
6 ( $\beta_{\log 2} = -1.69$  (-3.57; 0.19), both sexes) (Table 4, Figure 3). **Blood pressure** decreased linearly  
7 with doubling mercury concentrations in girls ( $\beta_{\log 2} = -1.13$  (-2.22; -0.04) for DBP and  $\beta_{\log 2} = -$   
8  $1.73$  (-3.46; 0.01) for SBP). (Table 4, Figure 3). **HRV characteristics** were not significantly  
9 associated with childhood exposure (linearly or non-linearly), though some graphically  
10 significant (inverse U shaped) associations were observed for the LF/HF ratio and DFA1 in  
11 boys and AETP in girls (Figure 3). **QT-wave related parameters** were not associated with  
12 childhood exposure (Table 4, Figure 4).

13

### 14 **3.6. Sensitivity analyses**

15 Additional adjustment for co-exposures to lead, cadmium, selenium (S-Figures 4), and maternal  
16 seafood consumption (S-Figures 5), did not modify the previously observed associations  
17 between cord blood mercury and cardiac health. The sensitivity analyses including current  
18 mercury exposure and n-3 PUFAs, were conducted within sub-samples of children having  
19 available measurements of these co-exposures (N=310 children (77%) for current blood  
20 mercury, and N=141 (35%) for n-3 PUFAs measurements). Within these respective sub-  
21 samples, associations were similar with or without additional adjustment for n-3 PUFAs in cord  
22 blood (S-Figures 6) or for current mercury exposure at 7 years (S-Table 5 and S-Figures 7) or  
23 for. However, it is to note that some non-adjusted associations observed within these selected  
24 subsamples were modified, compared to the main analysis. Finally, results remained unchanged  
25 after excluding birth weight as a covariate or adjusting for sporting activity (S-Figures 8).

1

2 **4. DISCUSSION**

3 Our study explored the potential cardiotoxic effects of prenatal and childhood exposure to  
4 mercury within a Caribbean cohort of children at 7 years of age.

5 Higher prenatal exposure was associated with higher systolic blood pressure at 7 years of age,  
6 without significant differences in mean HR. Some sex-specific associations were observed:  
7 boys with intermediate prenatal exposure had lower global HRV and reduced parasympathetic  
8 activity (HF). Moreover, longer corrected QT and QT-peak durations were observed in boys  
9 having higher levels of prenatal mercury. In girls, HRV tended to increase linearly with prenatal  
10 exposure, and no association was observed with QT-wave related parameters. Hg exposure of  
11 the children at 7 years of age was associated with decreased blood pressure in girls only (without  
12 association with mean HR). In boys, a non-linear increase in the LF/HF ratio (shift toward a  
13 sympathetic dominance) and DFA1 was observed for intermediate levels of exposure. These  
14 associations were insensitive to additional adjustment for prenatal co-exposure to lead,  
15 selenium, n3-PUFAs and maternal seafood consumption, sporting activity or birth weight.

16

17 ***Exposure levels***

18 With median cord blood mercury concentration at 6.6 µg/L (75<sup>th</sup> percentile=9.2 µg/L, 95<sup>th</sup>  
19 percentile=15 µg/L), the mercury exposure level in the Timoun cohort is lower than those  
20 reported in the Faroe Islands [14,46], Seychelles Islands [53–55] or among the Inuit in Canada  
21 [56], but higher than in inland European [57,58] or US populations [59,60]. Our insular  
22 population can therefore be characterized as moderately exposed to mercury compared to other  
23 populations in the world, with levels similar to those reported recently in a Hong-Kong  
24 population [61]. In the Guadeloupien population, no specific recommendations regarding  
25 seafood consumption were made in relation to mercury exposure. However, as in the French

1 population, the consumption of large wild predators (i.e. swordfish, marlin, shark, and lamprey)  
2 is highly discouraged because of their high levels of mercury contamination, the consumption  
3 of smaller wild predators has to be limited to 150g/week for pregnant and breastfeeding women  
4 (60g/week for children before 30 months), while maintaining a beneficial consumption of  
5 around two portions a week of non-predator fishes [62].

6

### 7 *Comparison with preceding studies*

8 The existing literature exploring the effect of early mercury exposure on children's **blood**  
9 **pressure** is inconclusive. A systematic review of 6 cohort studies that included children and  
10 adolescents from populations having various exposure levels to mercury reported mainly non-  
11 significant associations with prenatal and/or postnatal exposures [13]. Three more recent cohort  
12 studies [61,63,64] conducted in Europe, New-Hampshire and Hong-Kong found no significant  
13 associations with prenatal exposure. When significant, prenatal mercury exposure was  
14 associated with increased SBP and DBP at 7 years of age in the Faroe cohort [14], or with  
15 increased DBP in boys at 15 years of age in the Seychelles cohort [53]. Our results are consistent  
16 with this positive trend. Regarding postnatal exposure to mercury, only one of seven studies  
17 [46,56,60,61,64–66] reported increased DBP among boys at 5 years of age [64]. Conversely,  
18 Desai et al. (2021) [66] reported decreased DBP with higher exposure to a mixture of  
19 contaminants that included mercury (Pb, As, Hg, Cd) in the NHANES US cohort. They also  
20 found a non-linear association between DBP and mercury, though it was non-significant after  
21 adjustment for co-exposures. We also observed a tendency towards a negative association of  
22 current mercury exposure with BP (significantly decreased DBP in girls).

23 Two reviews have summarized the epidemiological evidence on the association between  
24 mercury exposure and **cardiac autonomic function**, assessed through HRV [11,12]. Between  
25 them, a total of 14 studies were covered, including children and/or adults, of which 7 assessed

1 cardiac function in children or adolescents after prenatal exposure and/or during current  
2 exposure to mercury [14,46,56,65,67,68] or at 19 years of age [54]. Two subsequent cohort  
3 studies: Zareba et al. 2019 (19-year-olds from the Seychelles cohort [55]) and Chan et al., 2021  
4 (7-8 year-olds from Hong-Kong [61]) completed the available literature. Of the 7 studies  
5 assessing prenatal exposure [14,46,54–56,61,67], 3 found no association [54–56] while 4  
6 showed negative associations with HRV indicators [14,46,61] and/or a positive association with  
7 a sympatho-dominant state (reduced HF, increased LF/HF ratio) [61,67]. As we also observed  
8 decreased HRV and HF and a trend toward increased LF/HF ratio, our results concur with these  
9 latter for boys. Unexpectedly, however, the associations with HRV tended toward the opposite  
10 direction for girls (borderline significant increases in RMSSD, SDNN and HF with exposure).  
11 Of the 9 studies assessing postnatal exposure, no clear evidence supported an association with  
12 HRV. When significant (in 4 studies), non-consistent associations were observed (decreasing  
13 LF with exposure in Sorensen et al. [14] and Valera et al. [56], but increasing in Valera et al.  
14 [65] and Murata et al. [67]). We found no association with LF, but a positive trend between HF  
15 and exposure that resulted in a significant higher LF/HF ratio for intermediate levels of  
16 exposure in boys.

17 The epidemiological literature analyzing **QT-wave related parameters** in association with  
18 mercury exposure is sparse, and almost inexistent when focusing on prenatal exposure. Of the  
19 HRV studies cited above, only one [67] also considered QT analysis, in Japanese children of  
20 similar age to those in our study. They did not find any association of prenatal or postnatal  
21 mercury exposure with the corrected QT (QTc), though they did not explore sex-specific  
22 associations. However, some cross-sectional epidemiological studies in various adult  
23 populations have produced consistent associations between mercury exposure and QT interval  
24 prolongation in farmers from Iraq who ingested contaminated bread [69], in Americans who  
25 were heavy consumers of fish [70], and in traffic enforcers from Manila [71]. Some



1 experimental studies have also shown increased QT durations during and/or after exposing rats  
2 to mercury: Santos-Ruybal et al. [72] showed that chronic MeHg treatment increased QTc and  
3 Tpeak–Tend interval duration; Carll et al. [73] found that exposure to diesel exhaust (which  
4 contains mercury) increased QT interval and decreased T-wave amplitude. In the Timoun  
5 cohort, we found a significant QTc prolongation with increasing prenatal mercury exposure in  
6 boys, and the same trend observed with current exposure (albeit non-significant). Associations  
7 of T-wave amplitude and TpTe with either prenatal or childhood exposures did not reach  
8 statistical significance in our study, although T-wave amplitude tended to decrease with  
9 prenatal and current exposure in girls and Tpeak–Tend interval duration tended to be higher in  
10 boys with higher current mercury exposure. Our results suggest that there is a need for further  
11 exploration of these QT-wave related parameters characterizing the repolarization phase in  
12 association with mercury in future studies.

#### 14 *Shape of the dose-response relationships*

15 Several of the significant associations we observed in our study with prenatal exposure were  
16 non-linear, in particular for HRV, for which spline modeling suggested several significant U-  
17 shaped (or inverted U-shaped) relationships. The non-linearity of the dose-response relationship  
18 between mercury exposure and cardiac health has been explored in only a few studies: Sorensen  
19 et al. [14] first suggested non-linear dose-response relationships between prenatal Hg exposure  
20 and the risk of impairments to the cardiac function (such as increasing SBP or decreasing HRV)  
21 in 7-year-olds, reporting a linear increase in mercury exposure at lower levels of exposure (<10  
22 µg/L cord blood), though not thereafter. This inflexion was not confirmed at a later age (14  
23 years of age) within the same cohort [46]. Other studies have also reported a non-linear dose-  
24 response relationship with BP: increasing DBP up to 8 µg/L prenatal Hg and flattening  
25 thereafter [53], with a linear increase in SBP starting after 4 µg/L prenatal Hg [60]. In our study,

1 in boys, HRV decreased with cord blood mercury until a concentration of around 8  $\mu\text{g/L}$ , at  
2 which point the slope reversed, resulting in non-significantly decreased HRV for concentrations  
3 above 20  $\mu\text{g/L}$ . The biological plausibility of a non-linear dose-response relationship with  
4 mercury exposure is questionable. The non-linear dose-response relations we observed in our  
5 study were complex to interpret because of log-transformations, and may result from certain  
6 unstable estimations of associations due to limited sample size and should be explored in future  
7 studies.

### 8 *Sex-specific effects of mercury*

9 Several associations reported in our study are consistently specific to boys (lower HRV, longer  
10 QT, higher BP associated with medium or high exposure). This is in accordance with previous  
11 studies, which found boy-specific associations with lower HRV [14,61] or higher BP [53,58].  
12 Another study reported a boy-specific inverse association (higher HRV with exposure) [54]  
13 while others found no interaction with sex [56,60]. Sex-specific susceptibility to prenatal  
14 mercury neurotoxicity has been reported in both epidemiological studies of human infants and  
15 children and in experimental animal studies [4]. Overall, these observed greater developmental  
16 effects on cognition in males. The mechanism underlying such sex-related differences may be  
17 related to a differential MeHg metabolism (faster body clearance of mercury in female rats than  
18 in males) and/or differences in antioxidant defenses (in the rat brain, mitochondria from female  
19 rats exhibit higher expression of antioxidant enzymes and lower production of reactive oxygen  
20 species). Moreover, estrogens were shown to have anti-oxidant effects [4]. These experimental  
21 results tend to support the boy-specific associations we observed in our study.

### 22 *Mechanisms*

23 Because of the well-established developmental neurotoxicity of mercury in humans,  
24 epidemiological studies historically investigated the effect of prenatal exposure to mercury on  
25 cardiac functions regulated by the ANS (HRV, BP). The specific mechanisms by which the

1 development of the ANS in utero is affected by mercury exposure are not elucidated. The  
2 mercury-induced neurotoxicity was partially explained by glutamate distribution and calcium  
3 homeostasis, as well as reactive oxygen species (ROS) generation, all of them activating  
4 neuronal death [74]. However we cannot exclude that the modifications in BP, HRV and QT  
5 we observed may also (partly or fully) involve direct cardiotoxicity of mercury exposure during  
6 the development of the heart. In humans exposed to high levels of mercury, craniofacial  
7 malformations, undescended testis and enlarged colon have been reported, while no indication  
8 of an excess of heart malformations was found [2]. However, perturbations of cardiac cellular  
9 maturation and tissue hypertrophy following mercury exposure have been reported in  
10 preweaning stage rats [75]. The mechanism of action of mercury on cardiac function is not yet  
11 fully understood but oxidative stress and deregulation of the calcium homeostasis have been  
12 identified in the literature as plausible modes of action [76–78].

13

#### 14 *Limitations*

15 The main limitation of our study relates to the measurement conditions of the cardiac function  
16 outcomes. During the ECG recording, children were in a sitting position (near their mother as  
17 she was interviewed) rather than in the standard supine position. The sitting vs. supine position  
18 may induce some systematic differences in the absolute measurement of cardiac function  
19 indicators – in particular an increase in LF power and a decrease in HF power in the sitting  
20 position [79], but our primary interest was the relative values of outcome between children  
21 exposed to contrasting levels of mercury. However, because the measurement conditions of the  
22 ECG- derived indicators was not fully controlled, the derived cardiac function indicators may  
23 be subject to measurement errors. This variability in measurement conditions can reasonably  
24 be considered independent of the exposure level, so that measurement errors are expected to be  
25 non-differential. Consequently, this did not bias our results – although it does limit the power

1 of our study as well as our ability to establish significant associations. BP was measured twice,  
2 but within a single day and in the context of a medical examination, and may not represent the  
3 child's usual BP level.

4 Multiple testing is another important issue in our study: we considered numerous outcome  
5 indicators, stratified the analyses, and tested linear and non-linear associations for both prenatal  
6 and childhood mercury exposures. The number of statistically significant associations is low  
7 compared to the number of tests performed, and random associations may be suspected. Our  
8 study may also suffer from a lack of power, particularly once sex stratification. This may have  
9 prevented us from achieving statistical significance of associations with some parameters,  
10 despite concordant trends with other indicators. The sensitivity analyses within sub-samples  
11 (children with blood sample at 7 years, children with n3-PUFAs measures in cord blood)  
12 suggested that some associations may be sensitive to sample selection. Within the sub-sample  
13 with n3-PUFAs information, variations in the associations compared to the main analysis may  
14 be due to the exclusion of both highest mercury concentrations and children with cardiovascular  
15 health risk factors (less premature births, less maternal hypertension or diabetes) in this sub-  
16 sample.

17 These limitations prevented us from taking full advantage of a comprehensive set of cardiac  
18 function indicators in order to better understand the mechanisms involved in suspected  
19 developmental mercury toxicity.

20

## 21 *Strengths*

### 22 *Assessment of cardiac function through multiple indicators*

23 The main strength of our study is the broad set of measurements used to characterize the  
24 children's overall cardiac health through different indicators (BP, HRV and QT-wave related  
25 parameters). The HRV assessment included 9 time-domain and frequency-domain

1 measurements commonly used in HRV studies, as well as 4 non-linear measurements  
2 considered independent predictors of mortality in patients with post myocardial infarction [80].  
3 In addition to blood pressure and HRV, ours is the second study to analyze the delayed effect  
4 of prenatal exposure to mercury on children's ECG morphological characteristics, after Murata  
5 et al. who also examined QT duration along with HRV, and found no association [67]. We also  
6 analyzed several QT-wave related parameters: QT and QT peak duration (which reflect  
7 ventricular depolarization and repolarization), but also the TpTe interval and the T-wave  
8 amplitude; this allowed us to explore potential specific modifications of the repolarization  
9 phase. In [81], the authors suggest that T-wave amplitude is significantly different between  
10 hypertensive patients and healthy controls, and could be used as an early marker of  
11 repolarization abnormalities in a hypertensive population. A recent study reports that  
12 morphological T-wave variation is the only ECG marker associated with life-threatening  
13 ventricular arrhythmias in individuals free of cardiovascular disease, independent of QT  
14 interval [82]. In this study, we failed to identify any significant associations between exposure  
15 and repolarization phase characteristics – though some borderline graphical trends between  
16 reduced T-wave amplitude and prolonged TpTe and mercury exposure (concordant with  
17 experimental studies) suggest that it might be of interest to include them (or more recent index  
18 quantifying T-wave morphologic variation) [82] in future studies.

### 19 *Quality of exposure measurement*

20 In populations featuring frequent fish and seafood consumption, blood mercury concentration  
21 is considered a good marker for mercury exposure, reflecting exposure during the preceding 2-  
22 3 months [2]. At 7 years of age, 66% of the Timoun cohort children reported eating oily or  
23 white fish one to three times a week. We can thus consider that despite a single measurement,  
24 the concentration accurately reflects the average exposure of the child at 7 years via diet. Other  
25 sources of exposure to mercury, including non-organic Hg, are unlikely to be present in the

1 Guadeloupean context, and negligible. It is therefore reasonable to consider total mercury blood  
2 concentrations measured at birth in cord blood, and at 7 years of age as reliable estimates of  
3 exposure to methylmercury during pregnancy and at 7 years, respectively. Because dietary  
4 habits, in particular fish consumption, evolves during childhood, mercury concentrations  
5 measured once at 7 years may not represent the whole childhood exposure. The associations  
6 with mercury exposure at 7 years should thus be interpreted as transversal.

### 7 *Controlling for co-exposures*

8 One strength of our study is that we controlled for the beneficial effects of seafood consumption  
9 and its associated nutrients (selenium, n3-PUFAs) in sensitivity analyses. In our population,  
10 maternal seafood consumption and selenium concentrations were significantly correlated with  
11 mercury concentrations in cord blood ( $R=0.39$  and  $0.41$ ,  $p<0.001$ , respectively). Mutual  
12 adjustment for mercury and selenium in cord blood or maternal seafood consumption did not  
13 modify the associations observed with mercury alone. The n3-PUFAs concentrations were  
14 available only for a small sub-sample of the children (those children without health  
15 complications during pregnancy or birth). In this selected sub-sample, the correlation between  
16 cord blood mercury and n3-PUFAs was low ( $R=0.10$ ,  $p=0.23$ ,  $N=141$ ) and co-adjustment did  
17 not modify the associations. These three sensitivity analyses suggest that negative confounding  
18 by beneficial nutrients present in consumed fish (which could cause underestimation of the true  
19 effect of mercury exposure) is unlikely in our study. This might be explained by the fact that in  
20 the Timoun cohort, mothers with the largest seafood consumptions presented higher mercury  
21 cord blood concentrations, but no increase in selenium or n3-PUFAs levels concentrations  
22 compared to the low consumers, probably due to the type of seafood they ate (data not shown).  
23 Chemical co-exposures in the Timoun cohort were also considered in our study. Cadmium was  
24 detected in only 53% of the neonates (median value =  $0.05 \mu\text{g/L}$ , maximum =  $1.6 \mu\text{g/L}$ ) and was  
25 not correlated with mercury. As expected, co-adjustment for Cd levels did not impact the

1 associations with mercury. Although Hg and Pb concentrations were not correlated in cord  
2 blood, and moderately negatively correlated in the children's blood at 7 years of age, co-  
3 exposure to lead was taken into account in a sensitivity analysis. Additional adjustment for Pb  
4 did not modify the associations and nor was prenatal Pb associated with any of the cardiac  
5 health indicators (data not shown). Since our study focused a priori on mercury exposure and  
6 addressed co-exposures as potential confounders rather than mixtures, we did not explore more  
7 complex co-exposure patterns.

8 All our analyses were controlled for child adiposity (BMI). Cardiovascular and cardiac  
9 functions are intricately linked within overall cardiometabolic health, and the causality direction  
10 between them is not clearly established. Within the Timoun cohort, SBP and HRV were found  
11 to be associated with adiposity in an earlier study [24]. Adjustment for child BMI allowed us  
12 to infer the association between mercury exposure and the part of the cardiac function not  
13 directly associated with adiposity. In a sensitivity analysis, we also controlled our analyses for  
14 each child's level of physical activity, as an established determinant of HRV and cardiovascular  
15 health, without any effect on the results.

16

## 17 **5. CONCLUSION**

18 Our study suggests associations between prenatal mercury exposure and disruption of some  
19 cardiac health parameters of children from an insular fish-consuming population, at 7 years of  
20 age. Associations with BP were unstable and discordant between prenatal exposure (positive)  
21 and exposure at 7 years (negative). Decreased HRV and a trend to sympatho-vagal dominance  
22 observed in boys was concordant with previous literature. Linear associations observed with  
23 prolonged QT were concordant between prenatal and childhood exposures and should be  
24 replicated.

1 However, the sex-specificity and non-linearity of the associations observed must be interpreted  
2 with caution in light of potential issues present in the study, in terms of statistical power,  
3 sampling variations and multiple testing. The persistence of such a pattern of associations needs  
4 to be assessed in a further follow-up at a later age before interpreting them as early signs of a  
5 long-term disruption of cardiac function following early exposure to mercury.

6

7

8

9

Journal Pre-proof



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**Table 1. Characteristics of the children included in the analyses**

Characteristics	BP analysis (N=399) <sup>a</sup>		HRV analysis (N=300) <sup>a</sup>		QT analysis (N=266) <sup>a</sup>	
	N	% or mean (std) or median (Q1; Q3)	N	% or mean (std) or median (Q1; Q3)	N	% or mean (std) or median (Q1; Q3)
<b>Mother at inclusion<sup>b</sup></b>						
Maternal age	399	31.6 (6.7)	300	31.6 (6.7)	266	31.3 (6.6)
<b>Geographical origin (mother)</b>						
French West Indies/Caribbean Islands	350	87.7	261	87.0	231	86.8
Europe	49	12.3	39	13.0	35	13.2
<b>Geographical origin (grandparents)</b>						
French West Indies/Caribbean Islands	382	95.7	288	96.0	256	96.2
Europe	17	4.3	12	4.0	10	3.8
<b>Maternal education (years)</b>						
< 5	17	4.3	13	4.3	12	4.5
5–12	276	69.2	203	67.7	177	66.5
> 12	106	26.6	84	28.0	77	29.0
<b>Pre-pregnancy aternal BMI</b>						
Underweight	26	6.6	18	6.1	15	5.7
Normal	211	53.8	159	53.7	142	54.2
Overweight	89	22.7	69	23.3	65	24.8
Obese	66	16.8	50	16.9	40	15.3
Smoking during pregnancy (Yes)	13	3.3	14	4.7	13	4.9
Alcohol during pregnancy (Yes)	10	2.6	8	2.7	6	2.3
Hypertension during pregnancy (Yes)	46	12.0	37	12.3	33	12.4
Seafood consumption (times/week)	263	3.1 (1.9; 5.6)	344	3.4 (1.9; 5.6)	233	3.1 (1.9; 5.6)
<b>Child at birth</b>						
Sex (Male)	187	47.0	149	49.7	136	51.1
Preterm birth (Yes)	53	13.3	38	12.7	34	12.8
Birth weight	399	3114 (516)	300	3148 (498)	266	3153 (512)
Cord blood n-3 PUFAs (%total FA)	141	5.0 (1.8)	110	5.2 (1.8)	97	5.1 (1.8)
Cord blood Hg (µg/l)	399	6.6 (4.4; 9.2)	300	6.5 (4.4; 9.2)	266	6.4 (4.4; 9.2)
Cord blood Pb (µg/l)	399	13.2 (10.2; 18.0)	300	13.1 (10.2; 17.6)	266	13.1 (10.2; 17.6)
Cord blood Cd (µg/l)	399	0.05 (<LOD; 0.17)	300	0.05 (<LOD; 0.11)	266	0.05 (<LOD; 0.16)
Cord blood Se (µg/l)	399	118.4 (100.0; 134.2)	300	110.5 (100.0; 134.2)	266	110.5 (100.0; 130.0)
<b>Child at 7 years</b>						
Age at examination (years)	399	7.7 (0.2)	300	7.7 (0.2)	266	7.7 (0.2)
Height (cm)	399	130.6 (5.9)	300	130.9 (6.0)	266	130.9 (6.0)
z-BMI	397	0.3 (1.3)	299	0.3 (1.3)	265	0.3 (1.3)
<b>Child BMI</b>						
Underweight	8	2.0	5	1.7	6	2.2
Normal	281	70.4	211	70.3	188	70.7
Overweight	64	16.1	55	18.3	46	17.3
Obese	46	11.5	29	9.7	26	9.8
Sport practice (Yes)	181	45.2	137	44.7	120	45.1
Sport time among practicing (hrs/wks)		2.8 (1.7)		2.8 (1.8)		2.8 (1.8)
Maternal smoking (Yes)	82	20.7	69	23.1	62	23.4
Passive smoking in childhood (Yes)	69	20.2	56	21.8	49	21.7
Blood Hg (µg/l)	310	1.7 (1.0; 2.8)	231	1.8 (1.0; 2.9)	204	1.7 (1.0; 2.6)
Blood Pb (µg/l)	310	22.0 (16.2; 27.1)	231	22.2 (16.4; 27.4)	204	22.2 (16.5; 27.0)
Blood Cd (µg/l)	310	0.10 (<LOD; 0.18)	231	0.10 (<LOD; 0.18)	204	0.10 (<LOD; 0.18)

<b>Blood Se (<math>\mu\text{g/l}</math>)</b>	310	110.0 (70.0, 120.0)	251	110.0 (70.0, 120.0)	204	100.0 (70.0, 110.0)
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<sup>a</sup> N=259 children (65%) included in the three analyses

<sup>b</sup> Maternal characteristics were collected at inclusion at a median term of 29 weeks of amenorrhea (IQR=(27-32))

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**Table 2. Cardiac function indicators in 7-year-old children included in the Timoun study**

		All	Boys	Girls		
Cardiac Function indicators	N <sup>a</sup>	Median (Q1; Q3)	Median (Q1; Q3)	Median (Q1; Q3)	<i>p</i> -value <sup>b</sup>	
<b>Blood Pressure</b>	SBP (mm Hg)	399	99.0 (93.0; 106.0)	99.0 (95.0; 106.0)	98.0 (93.0; 105.0)	0.33
	DBP (mm Hg)	399	57.0 (53.0; 61.0)	57.0 (53.0; 60.0)	57.0 (54.0; 61.0)	0.64
<b>Heart Rate</b>	Mean (bpm)	300	85.1 (77.6; 93.6)	82.5 (76.0; 91.9)	86.8 (79.7; 94.6)	<b>0.004</b>
<b>HRV</b>	RMSSD (ms)	300	72.6 (48.2; 105.4)	76.0 (48.7; 111.1)	69.22 (46.6; 100.5)	0.20
	SDNN (ms)	300	73.1 (55.8; 97.5)	75.2 (55.4; 99.4)	71.2 (55.9; 94.8)	0.19
	LF (10 <sup>3</sup> .ms <sup>2</sup> )	300	0.10 (0.05; 0.16)	0.11 (0.05; 0.17)	0.09 (0.05; 0.15)	0.26
	HF (10 <sup>3</sup> .ms <sup>2</sup> )	300	0.14 (0.07; 0.29)	0.15 (0.07; 0.30)	0.13 (0.07; 0.29)	0.36
	LF/HF	300	0.63 (0.42; 0.98)	0.63 (0.44; 0.99)	0.65 (0.42; 0.94)	0.84
	SETP	300	1.80 (1.63; 1.95)	1.81 (1.65; 1.95)	1.79 (1.60; 1.97)	0.43
	AETP	300	1.24 (1.20; 1.28)	1.24 (1.20; 1.27)	1.24 (1.21; 1.28)	0.57
	DFA1	300	0.86 (0.75; 1.02)	0.85 (0.75; 1.02)	0.88 (0.74; 1.02)	0.34
	DFA2	300	0.73 (0.61; 0.85)	0.75 (0.61; 0.86)	0.70 (0.61; 0.84)	0.14
<b>QT-wave analysis</b>	QTc (ms)	264	420.0 (408.6; 432.7)	417.8 (407.6; 431.2)	420.6 (409.5; 435.2)	0.32
	QTpc (ms)	264	330.7 (317.9; 341.7)	330.1 (319.2; 339.9)	331.3 (316.8; 343.0)	0.86
	T amplitude (μV)	266	634.1 (469.1; 749.1)	688.2 (531.5; 776.4)	558.3 (400.8; 701.0)	<b>&lt;0.0001</b>
	TpTe (ms)	266	74.7 (71.9; 78.4)	74.5 (71.8; 77.9)	74.9 (71.9; 79.2)	0.45

DBP, Diastolic Blood Pressure; SBP, Systolic Blood Pressure; RMSSD, Root Mean Square of the Successive Differences of RR intervals; SDNN, Standard Deviation of the N-N intervals; HF, High Frequency; LF, Low Frequency; LF/HF, LF/HF ratio; SETP, Sample Entropy; AETP, Approximate Entropy; DFA1, Detrended short term Fluctuations; DFA2, Detrended long term Fluctuations; QTc, corrected (Bazett) QT duration; QTpc, corrected (Bazett) QT-peak duration; T amp, T-Wave amplitude; TpTe, interval from T-wave peak to end.

<sup>a</sup> N=259 children (65%) included in the three analyses

<sup>b</sup> *p*-value : Wilcoxon test comparing boys and girls

**Table 3. Association of mercury cord blood concentrations with cardiovascular function at 7 years of age**

		Linear modeling				GAM modeling <sup>c</sup>		
		Both sexes	Boys	Girls	Sex interaction <sup>b</sup>	Both sexes	Boys	Girls
		$\beta$ or $10^\beta$ <sup>a</sup> (95% CI)	$\beta$ or $10^\beta$ <sup>a</sup> (95% CI)	$\beta$ or $10^\beta$ <sup>a</sup> (95% CI)		edf ( <i>p-val</i> )	edf ( <i>p-val</i> )	edf ( <i>p-val</i> )
<b>Heart Rate</b>	HR	0.72 (-0.80; 2.24)	0.56 (-1.68; 2.80)	0.97 (-1.24; 3.19)	0.80	2.6 (0.18)	1.4 (0.68)	2.7 (0.18)
<b>Blood Pressure</b>	DBP	0.59 (-0.07; 1.25)	1.08 (-0.04; 2.19)	0.31 (-0.48; 1.1)	0.29	1.5 (0.16)	2.2 (0.08)	1 (0.44)
	SBP	<b>1.02 (0.10; 1.93)</b>	0.77 (-0.56; 2.09)	<b>1.28 (0.00; 2.57)</b>	0.54	1 (0.03)	1.5 (0.40)	1.1 (0.07)
<b>Heart Rate Variability</b>	RMSSD (log)	1.04 (0.93 ;1.16)	0.93 (0.80 ;1.08)	<b>1.18 (1.00 ;1.40)</b>	<b>0.02</b>	1 (0.49)	<b>3.6 (0.04)*</b>	1 (0.05)
	SDNN (log)	1.01 (0.92 ;1.10)	0.92 (0.81 ;1.04)	1.11 (0.97 ;1.27)	<b>0.03</b>	1.1 (0.94)	<b>3.4 (0.03)*</b>	1 (0.11)
	HF (log)	1.05 (0.83 ;1.34)	0.86 (0.62 ;1.20)	1.36 (0.95 ;1.95)	<b>0.03</b>	2 (0.56)	<b>3.6 (0.02) *</b>	1 (0.09)
	LF (log)	1.02 (0.83 ;1.26)	0.84 (0.64 ;1.11)	1.24 (0.91 ;1.69)	<b>0.09</b>	1 (0.87)	2.2 (0.29)	1 (0.16)
	LF/HF (log)	0.97 (0.81 ;1.15)	0.97 (0.76 ;1.25)	0.91 (0.72;1.16)	0.37	2.4 (0.49)	2.8 (0.23)	1 (0.46)
	SETP	0.01 (-0.02; 0.04)	0.003 (-0.04; 0.04)	0.01 (-0.03; 0.05)	0.80	1 (0.52)	2.8 (0.52)	1.3 (0.71)
	AETP (log)	1.00 (0.98 ;1.02)	1.00 (0.97 ;1.03)	1 (0.973 ;1.03)	0.92	1.8 (0.74)	2.6 (0.31)	1 (0.95)
	DFA1	-0.01 (-0.03; 0.01)	-0.01 (-0.04; 0.03)	-0.02 (-0.05; 0.01)	0.34	1 (0.39)	2.2 (0.58)	1 (0.26)
	DFA2	0.000 (-0.02; 0.02)	0.01 (-0.03; 0.04)	-0.02 (-0.05; 0.01)	0.21	1 (0.98)	3.4 (0.36)	1 (0.28)
<b>QT-wave analysis</b>	QTc	1.30 (-1.27; 3.87)	<b>4.02 (0.48; 7.56)</b>	-1.63 (-5.55; 2.29)	<b>0.06</b>	1 (0.32)	1 (0.03)	1 (0.41)
	QTpc	0.82 (-1.74; 3.37)	<b>3.62 (0.04; 7.21)</b>	-2.25 (-6.09; 1.59)	<b>0.03</b>	1 (0.53)	1 (0.05)	1 (0.25)
	Tamp	-13.24 (-43.97; 17.49)	-2.52 (-44.3; 39.25)	-29.73 (-76.85; 17.4)	0.22	1 (0.40)	1 (0.92)	1.5 (0.34)
	TpTe (log)	1.01 (0.98 ;1.04)	1.00 (0.97 ;1.03)	1.016 (0.964 ;1.074)	0.44	1 (0.60)	1 (0.92)	1 (0.54)

HR, Heart Rate; DBP, Diastolic Blood Pressure; SBP, Systolic Blood Pressure; RMSSD, Root Mean Square of the Successive Differences of RR intervals; SDNN, Standard Deviation of the N-N intervals; HF, High Frequency; LF, Low Frequency; LF/HF, LF/HF ratio; SETP, Sample Entropy; AETP, Approximate Entropy; DFA1, Detrended short term Fluctuations; DFA2, Detrended long term Fluctuations; QTc, corrected (Bazett) QT duration; QTpc, corrected (Bazett) QT-peak duration; Tamp, T-Wave amplitude; TpTe, interval from T-wave peak to end. Models were adjusted for the child sex, exact age at measurement (in months), height, BMI, and birth weight, the maternal hypertension and smoking during pregnancy, maternal educational level and geographical origin of the maternal grand-parents of the child (a proxy for ethnic origin). For all HRV parameters, models were additionally adjusted for mean heart rate.

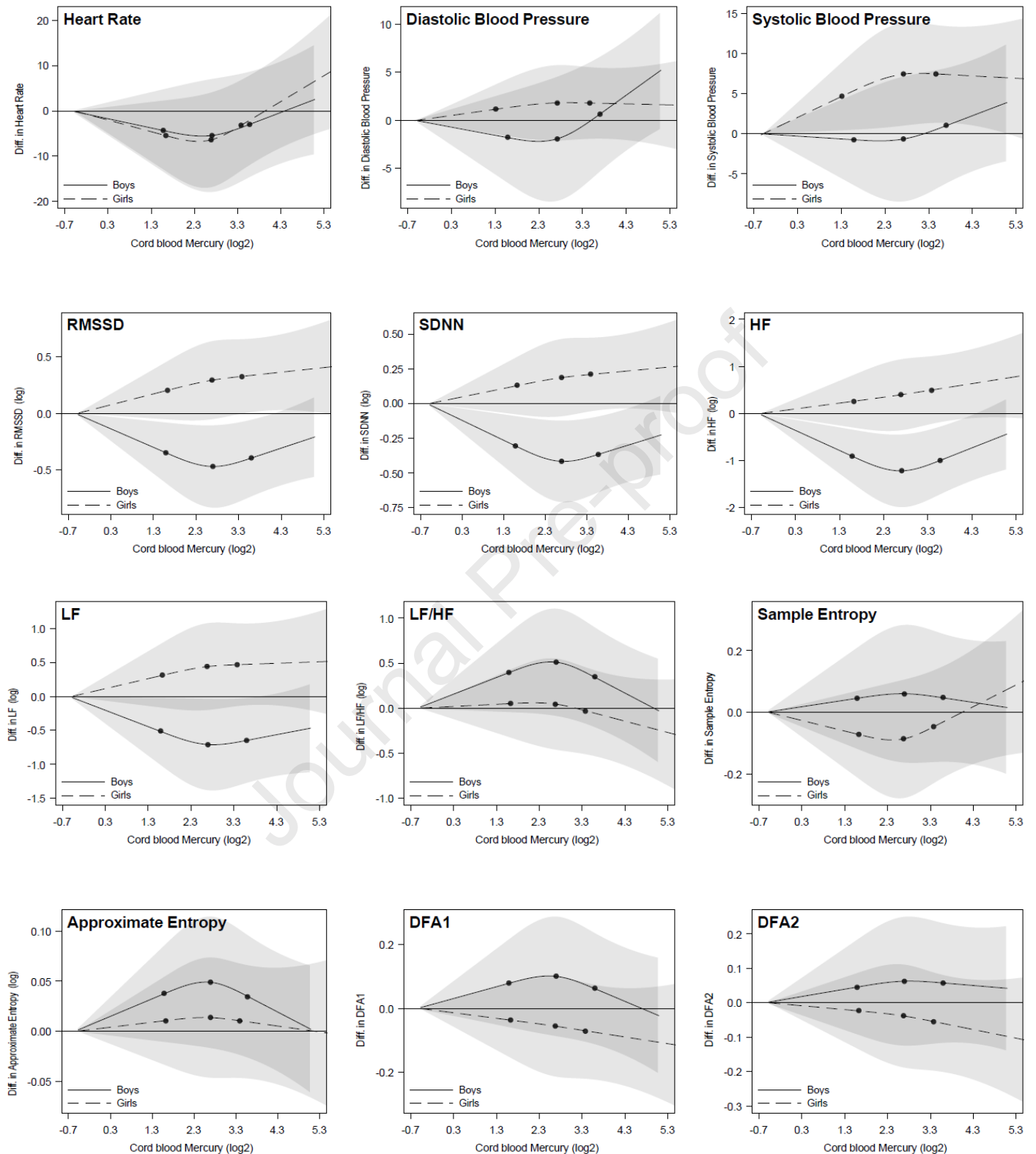
<sup>a</sup>  $\beta$  represents variation in outcome for a doubling of the exposure. When the outcome was not transformed, variation in outcome is absolute (outcome +  $\beta$ ). When the outcome was log transformed, variation in outcome is multiplicative (outcome x  $10^\beta$ ) ( $10^\beta$  is indicated in the table).

<sup>b</sup> *p*-value of the test of significance of interaction between cord blood mercury concentration and sex of the child in adjusted linear models

° Generalized Additive Models: Automatic smoothness estimation by cross-validation (*mgcv* R package). edf indicates the estimated degree of freedom. Significance ( $p$ -value) of the smooth term of the GAM analysis (restricted cubic splines of cord blood mercury log-concentrations) is given in parentheses. \* indicates a significant Likelihood Ratio test comparing the fit (AIC) of linear modeling versus GAM (K $\chi^2$  test,  $p < 0.10$ ).

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**Figure 1. Associations of mercury concentrations in cord blood with heart rate, blood pressure and HRV at 7 years of age (GAM, stratified by sex) <sup>a,b</sup>**

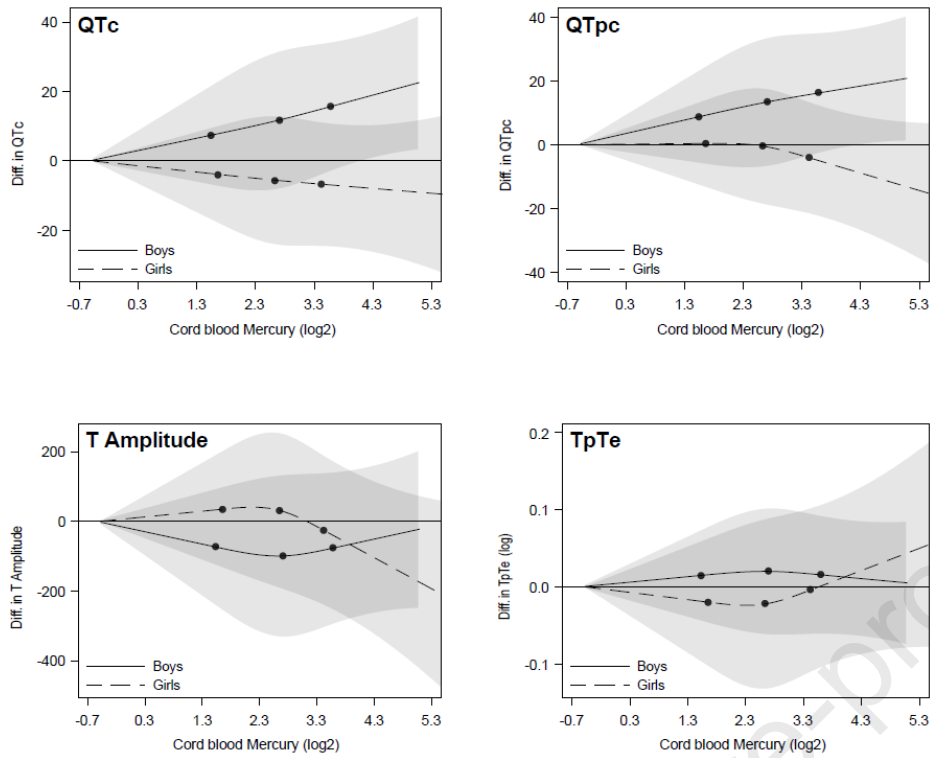


HR, Heart Rate; DBP, Diastolic Blood Pressure; SBP, Systolic Blood Pressure; RMSSD, Root Mean Square of the Successive Differences of RR intervals; SDNN, Standard Deviation of the N-N intervals; HF, High Frequency; LF, Low Frequency; LF/HF, LF/HF ratio; SETP, Sample Entropy; AETP, Approximate Entropy; DFA1, Detrended short term Fluctuations; DFA2, Detrended long term Fluctuations.

<sup>a</sup> N=149 boys and N=151 girls for HR and HRV analyses; N=149 boys and N=212 girls for BP analyses

<sup>b</sup> Generalized Additive Models with restricted cubic splines of cord blood mercury log<sub>2</sub>-concentrations (nodes at the 10<sup>th</sup>, 50<sup>th</sup>, and 90<sup>th</sup> percentiles of the distribution of exposure)

**Figure 2. Associations of mercury concentrations in cord blood with QT-wave related parameters at 7 years of age (GAM, stratified by sex) <sup>a,b</sup>**



QTc, corrected (Bazett) QT duration; QTpc, corrected (Bazett) QT-peak duration; Tamp, T-Wave amplitude; TpTe, interval from T-wave peak to end.

<sup>a</sup> N=136 boys and N=128 girls

<sup>b</sup> Generalized Additive Models with restricted cubic splines of cord blood mercury log<sub>2</sub>-concentrations (nodes at the 10<sup>th</sup>, 50<sup>th</sup>, and 90<sup>th</sup> percentiles of the distribution of exposure)

**Table 4. Association of mercury blood concentrations at 7 years with cardiovascular function at 7 years of age**

		Linear modeling			GAM modeling <sup>c</sup>			
		Both sexes	Boys	Girls	Sex	Both sexes	Boys	Girls
		$\beta$ or $10^{\beta}$ <sup>a</sup> (95% CI)	$\beta$ or $10^{\beta}$ <sup>a</sup> (95% CI)	$\beta$ or $10^{\beta}$ <sup>a</sup> (95% CI)	interaction <sup>b</sup>	edf ( <i>p-val</i> )	edf ( <i>p-val</i> )	edf ( <i>p-val</i> )
<b>Heart Rate</b>	HR	-1.17 (-2.47; 0.13)	-0.6 (-2.43; 1.23)	-1.29 (-3.33; 0.76)	0.96	1 (0.08)	1 (0.52)	1 (0.21)
<b>Blood Pressure</b>	DBP	-0.33 (-0.92; 0.26)	-0.07 (-1.03; 0.9)	-0.78 (-1.54; -0.03)	0.13	2.5 (0.20)	2.4 (0.43)	1.1 (0.05)
	SBP	-0.57 (-1.4; 0.25)	-0.14 (-1.29; 1.01)	-1.2 (-2.4; 0.01)	0.16	1.5 (0.29)	1 (0.87)	1 (0.05)
<b>Heart Rate Variability</b>	RMSSD	0.98 (0.89; 1.08)	0.94 (0.83; 1.08)	0.99 (0.86; 1.15)	0.84	1.6 (0.73)	1.8 (0.46)	1 (0.91)
	SDNN	0.99 (0.91; 1.07)	0.97 (0.88; 1.08)	0.98 (0.87; 1.11)	0.53	1 (0.80)	1 (0.59)	1 (0.77)
	HF	0.95 (0.77; 1.18)	0.92 (0.7; 1.22)	0.92 (0.66; 1.27)	0.45	1.7 (0.73)	2.8 (0.30)	1 (0.61)
	LF	1.06 (0.88; 1.28)	1.03 (0.81; 1.31)	1.06 (0.79; 1.42)	0.73	1 (0.51)	1 (0.80)	1 (0.71)
	LF_HF	1.12 (0.96; 1.3)	1.12 (0.91; 1.37)	1.15 (0.93; 1.43)	0.52	2.1 (0.18)	2.6 (0.10)	1 (0.21)
	SETP	0.01 (-0.01; 0.04)	0.02 (-0.02; 0.05)	0.01 (-0.03; 0.04)	0.75	1 (0.34)	1 (0.31)	1.2 (0.88)
	AETP	1.01 (1; 1.03)	1.01 (0.99; 1.04)	1.02 (0.99; 1.05)	0.60	1 (0.10)	3.6 (0.09)	2.1 (0.16)
	DFA1	0.01 (-0.01; 0.03)	0.01 (-0.01; 0.04)	0 (-0.03; 0.03)	0.93	1.9 (0.39)	2.4 (0.14)	1 (0.82)
<b>QT-wave analysis</b>	DFA2	-0.01 (-0.02; 0.01)	0 (-0.03; 0.02)	-0.01 (-0.04; 0.02)	0.89	1.9 (0.45)	1.5 (0.71)	1 (0.60)
	QTc	0.9 (-1.35; 3.15)	1.53 (-1.43; 4.5)	0.33 (-3.43; 4.08)	0.86	1 (0.45)	1 (0.31)	1 (0.87)
	QTpc	1.23 (-0.98; 3.43)	0.87 (-2.12; 3.87)	2.15 (-1.43; 5.74)	0.35	1 (0.27)	1.2 (0.71)	1 (0.24)
	Tamp	-11.41 (-38.57; 15.75)	-5.34 (-43.01; 32.33)	-24.02 (-66.95; 18.91)	0.52	1 (0.41)	1.3 (0.81)	1.8 (0.35)
	TpTe	1.01 (0.98; 1.03)	1.02 (0.99; 1.05)	0.97 (0.93; 1.02)	<b>0.03</b>	1 (0.78)	3.4 (0.09)	1 (0.26)

HR, Heart Rate; DBP, Diastolic Blood Pressure; SBP, Systolic Blood Pressure; RMSSD, Root Mean Square of the Successive Differences of RR intervals; SDNN, Standard Deviation of the N-N intervals; HF, High Frequency; LF, Low Frequency; LF/HF, LF/HF ratio; SETP, Sample Entropy; AETP, Approximate Entropy; DFA1, Detrended short term Fluctuations; DFA2, Detrended long term Fluctuations; QTc, corrected (Bazett) QT duration; QTpc, corrected (Bazett) QT-peak duration; Tamp, T-Wave amplitude; TpTe, interval from T-wave peak to end.

Models were adjusted for the child sex, exact age at measurement (in months), height, BMI, and birth weight, the maternal hypertension and smoking during pregnancy, maternal educational level and geographical origin of the maternal grand-parents of the child (a proxy for ethnic origin). For all HRV parameters, models were additionally adjusted for mean heart rate.

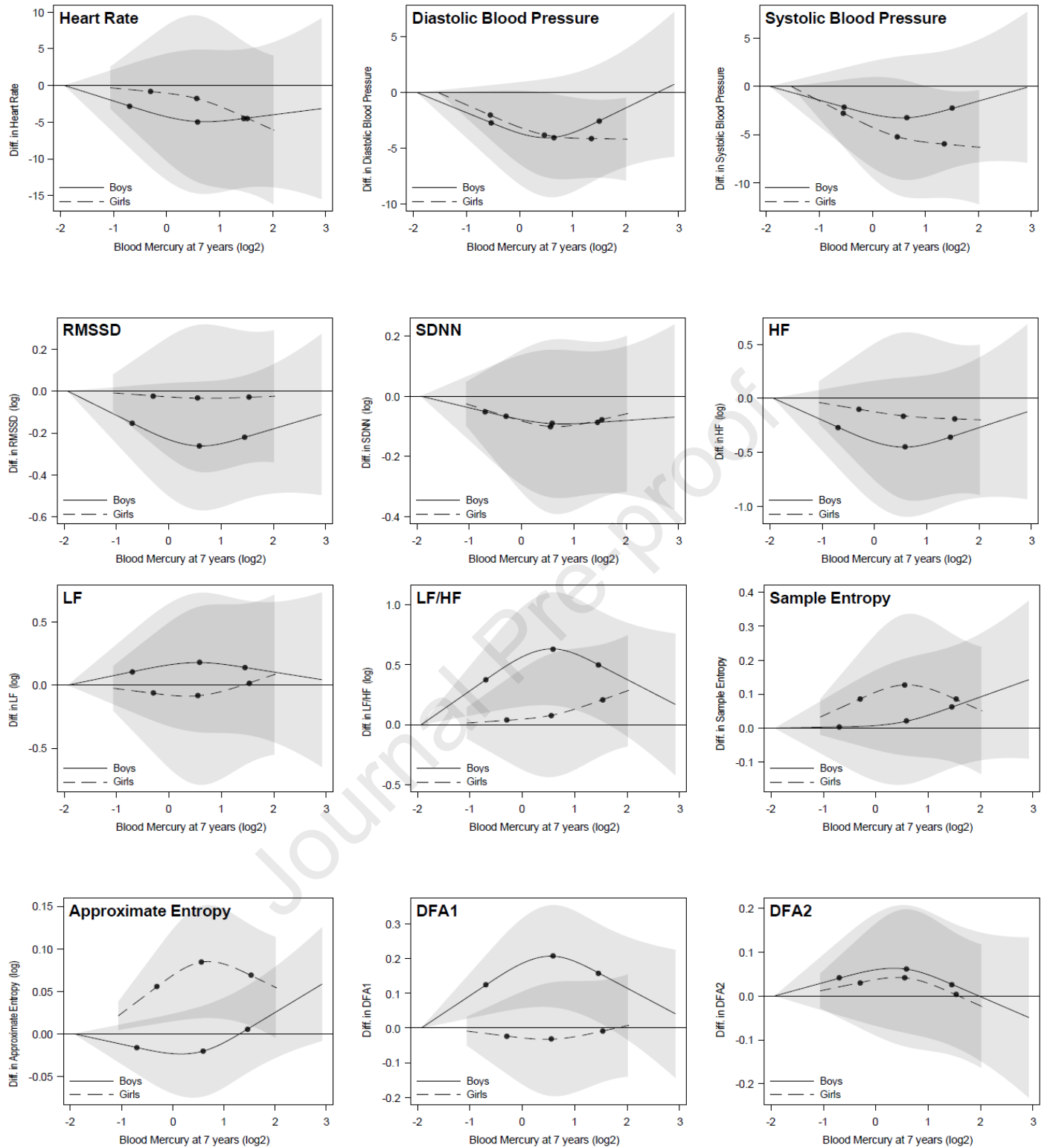
<sup>a</sup>  $\beta$  represents variation in outcome for a doubling of the exposure. When the outcome was not transformed, variation in outcome is absolute (outcome +  $\beta$ ). When the outcome was log transformed, variation in outcome is multiplicative (outcome x  $10^{\beta}$ ) ( $10^{\beta}$  is indicated in the table).

<sup>b</sup> p-value of the test of significance of interaction between cord blood mercury concentration and sex of the child in adjusted linear models

<sup>c</sup> Generalized Additive Models: Automatic smoothness estimation by cross-validation (*mgcv* R package). edf indicates the estimated degree of freedom. Significance (*p*-value) of the smooth term of the GAM analysis (restricted cubic splines of cord blood mercury log-concentrations) is given in parentheses. \* indicates a significant Likelihood Ratio test comparing the fit (AIC) of linear versus GAM modeling (K $\chi^2$  test,  $p < 0.10$ ).

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**Figure 3. Associations of mercury concentrations in blood at 7 years of age with heart rate, blood pressure and HRV at 7 (GAM, stratified by sex) <sup>a,b</sup>**



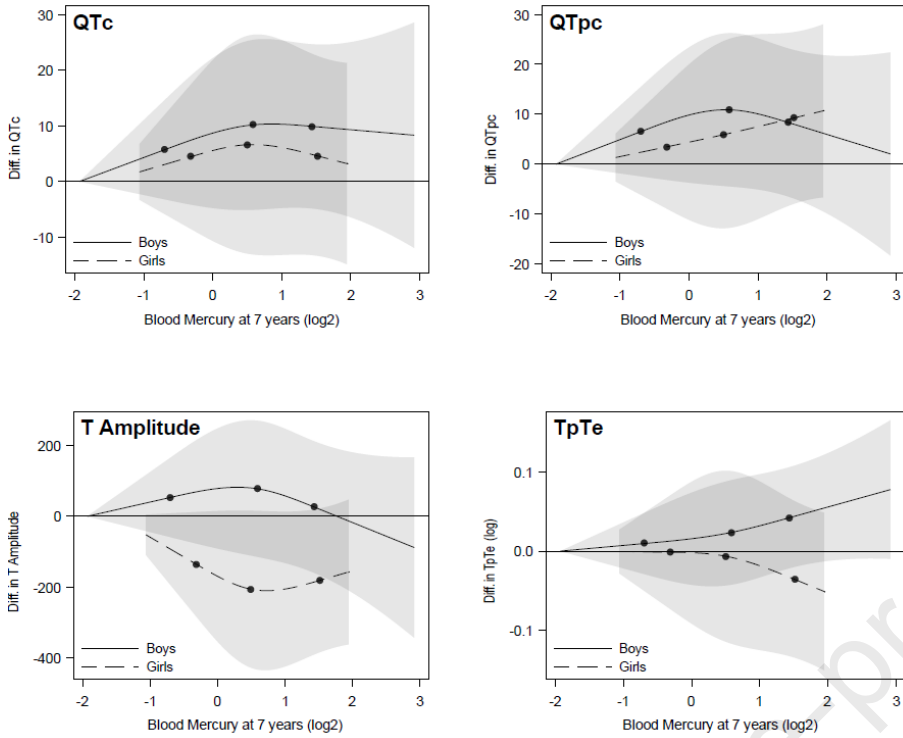
HR, Heart Rate; DBP, Diastolic Blood Pressure; SBP, Systolic Blood Pressure; RMSSD, Root Mean Square of the Successive Differences of RR intervals; SDNN, Standard Deviation of the N-N intervals; HF, High Frequency; LF, Low Frequency; LF/HF, LF/HF ratio; SETP, Sample Entropy; AETP, Approximate Entropy; DFA1, Detrended short term Fluctuations; DFA2, Detrended long term Fluctuations

<sup>a</sup> N=113 boys and N=118 girls for HR and HRV analyses; N=143 boys and N=167 girls for BP analyses

<sup>b</sup> Generalized Additive Models with restricted cubic splines of blood mercury log<sub>2</sub>-concentrations (nodes at the 10<sup>th</sup>, 50<sup>th</sup>, and 90<sup>th</sup> percentiles of the distribution of exposure)



**Figure 4. Associations of mercury concentrations in blood at 7 years of age with QT-wave related parameters at 7 (GAM, stratified by sex) <sup>a,b</sup>**



QTc, corrected (Bazett) QT duration; QTpc, corrected (Bazett) QT-peak duration; T Amp, T-Wave amplitude; TpTe, interval from T-wave peak to end.

<sup>a</sup> N=104 boys and N=100 girls

<sup>b</sup> Generalized Additive Models with restricted cubic splines of blood mercury log<sub>2</sub>-concentrations (nodes at the 10<sup>th</sup>, 50<sup>th</sup>, and 90<sup>th</sup> percentiles of the distribution of exposure)

## Highlights

- Insular populations in the French Caribbeans are exposed to mercury from high seafood consumption
- Resulting prenatal exposure may impair the development of the cardiac function
- Reduced heart rate variability and longer QT were observed in boys with higher prenatal mercury exposure
- Higher blood pressure at age 7 was observed after higher prenatal mercury exposure

**Declaration of interests**

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests:

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