Aerobic Fitness and Neurocognitive Function Scores in Young Faroese Adults and Potential Modification by Prenatal Methylmercury Exposure

Youssef Oulhote,1 Frodi Debes,2 Sonja Vestergaard,3 Pal Weihe,2 and Philippe Grandjean1,3

1Department of Environmental Health, Harvard T.H. Chan School of Public Health, Boston, Massachusetts, USA; 2Department of Occupational Medicine and Public Health, Faroese Hospital System, Torshavn, Faroe Islands; 3Institute of Public Health, University of Southern Denmark, Odense, Denmark

Background: Exposure to methylmercury was shown to decrease neural stem cell populations, whereas aerobic fitness has beneficial effects on the adult brain that relies on improved neurogenesis in the hippocampus.

Objectives: We examined the association between aerobic fitness and neurocognitive outcomes at young adult age, along with the potential moderating effect of prenatal exposure to methylmercury.

Methods: At age 22 years, 262 members of a Faroese birth cohort, established in 1986–1987, underwent a graded exercise test of aerobic fitness to measure maximal oxygen uptake (VO2Max). Their prenatal methylmercury exposure had been assessed from the mercury concentration in cord blood. We estimated cross-sectional associations between VO2Max and multiple measures of neurocognitive function. In addition, we compared groups with low and high prenatal methylmercury exposure.

Results: A 1 standard deviation (SD) increase in VO2Max was associated with better scores on short-term memory and cognitive processing speed by 0.21 SD (95% CI: −0.04, 0.46) and 0.28 SD (95% CI: 0.02, 0.54), respectively. In the group with lower prenatal methylmercury exposure, a 1 SD increase in VO2Max was associated with increased scores on cognitive processing speed by 0.45 SD (95% CI: 0.08, 0.81) and with a slightly lesser benefit in short-term memory. No such association was observed in the group with high prenatal methylmercury exposure.

Conclusions: Higher aerobic capacity was associated with better performance in short-term memory and processing speed. However, prenatal methylmercury exposure seemed to attenuate these positive associations.

Citation: Oulhote Y, Debes F, Vestergaard S, Weihe P, Grandjean P. 2017. Aerobic fitness and neurocognitive function scores in young Faroese adults and potential modification by prenatal methylmercury exposure. Environ Health Perspect 125:677–683; http://dx.doi.org/10.1289/EHP274

A subsample of 262 cohort members underwent fitness testing during a limited period, thus believed to be at random because it depended only on the examination schedule. All subjects responded to a questionnaire on past medical history, current health status, and lifestyle habits (see “Assessment of Participants’ physical activity (Questionnaire)” in the Supplemental Material). The ethical review committee covering the Faroe Islands as well as the U.S. institutional review board approved the study protocol, and all participants provided written, informed consent.

Aerobic Fitness Measurement

Participants underwent a progressive test performed on a mechanically braked cycle-ergometer (Monark AB, Vansbro, Sweden). They were asked to cycle for 5 min at a 75 Watt (W) load for females and 100W load for males, maintaining 60–70 rounds per minute throughout the test. After the first 5 min, loads were increased by 35W every 2 min until the participant was exhausted. Heart rate was monitored and an optimal test was defined as attained if the heart rate was ≥ 185 beats per minute and/or if the test leader evaluated the participant had reached exhaustion when the test was stopped by the participant. All participants received verbal encouragement from the test leader.
throughout the test. We estimated the maximum oxygen uptake (VO$_2$Max) from a maximal power output (MPO) as described by Andersen (1995). MPO was calculated as the Watts in the last completed workload, plus the increment in Watts of the last step divided by 120 sec, and multiplied by the number of seconds of the last step. Maximal oxygen uptake (VO$_2$Max) in L/min was estimated using the formula: VO$_2$Max (L/min) = 0.16 + (0.0117 × MPO), and then divided by weight and expressed in mL/kg/min.

**Prenatal Methylmercury Exposure**

Prenatal exposure to methylmercury was determined from mercury analysis of umbilical cord blood samples collected at birth (CB-Hg in μg/L). Briefly, blood samples from the umbilical cord were taken in 10-mL Abbott syringes equipped with Teflon-lined pistons, and kept deep-frozen during transport and storage until analysis (Grandjean et al. 1992). Mercury concentrations were determined using a UV-absorptiometer (Mercury Monitor 1235). The quality was secured by inclusion of quality controls and standard reference material samples from the National Institute of Standards and Technology (Gaithersburg, MD, USA) as previously described by Grandjean et al. (1992).

**Neurocognitive Functions**

A battery of neuropsychological tests was administered to ascertain a broad range of cognitive and learning abilities (Debes et al. 2016). We used several subtests from well-established clinical test batteries: the Wechsler Intelligence Scale for Children revised (WISC-R) (Wechsler 1974), the Wechsler Adult Intelligence Scale revised (WAIS-R) (Wechsler 1981), the Wechsler Memory Scale third edition (Wechsler 1997), the Woodcock–Johnson III test of cognitive abilities (Woodcock et al. 2001), the Boston Naming Test (BNT) (Kaplan et al. 1983), and the California Verbal Learning Test (CVLT) (Delis et al. 1987). The neurocognitive assessment took place at the same day, and just before the aerobic fitness testing. Detailed information about the tests administered has been recently published (Debes et al. 2016) and can also be found in “Neuropsychological tests (From Debes et al. 2016)” and can also be found in “Neuropsychological tests (From Debes et al. 2016)” to examine univariate associations between VO$_2$Max and participants’ characteristics.

We explored associations between VO$_2$Max and neurocognitive test scores using linear regressions adjusting only for sex. Furthermore, we used structural equations modeling (SEM) to assess the relationship between VO$_2$Max and groups of neurocognitive outcomes. In this approach, we considered test scores from specific neurocognitive tasks as indicators of six underlying latent functions in accordance with the Cartell–Horn–Carroll three-stratum theory (Schneider and McGrew 2012): Short-term Memory, Verbal Comprehension and Knowledge, Psychomotor Speed, Visual Processing, Long-term Storage and Retrieval, and Cognitive Processing Speed (Debes et al. 2016). Additional methodological aspects of the construction of latent functions are described in “Methodology for structural equations modeling” in the Supplemental Material. In a second approach, we allowed the six neurocognitive functions to be indicators of a broader neurocognitive function (g). In the first model, all the six neurocognitive functions were regressed on VO$_2$Max whereas in the second model, only the global g factor was regressed on VO$_2$Max adjusting for the same set of covariates. In a final model, we allowed the latent functions to be indicators of two broader neurocognitive functions, where Cognitive Processing Speed and Short-term Memory reflected Cognitive Efficiency, whereas Verbal Comprehension and Knowledge, Visual Processing, Long-term Storage and Retrieval, and participant’s Raven scores indicated the construct of General Thinking abilities (Debes et al. 2016; Woodcock et al. 2001). These two broader constructs were then regressed on VO$_2$Max while again adjusting for the same covariates. Predictors of neurocognitive outcomes—maternal and paternal employment, maternal and paternal education/training, and mother’s Raven scores—were also aggregated in a single latent variable to reflect family background. Some participants had missing data for physical activity (n = 1), smoking status (n = 2), maternal and paternal employment (n = 17), maternal education/training (n = 18), paternal education/training (n = 17), maternal Raven score (n = 21), and single neurocognitive test scores [n between 1 and 5, except for block designs (n = 48)]. We imputed missing data using multiple imputations by chained equations. We specified an adequate number of iterations (n = 10) according to the proportion of missing information (White et al. 2011). Mean estimates and variances were computed from the 10 imputed data sets using Rubin’s rules (Rubin 1987).

In additional analyses, we explored the differences in the association between VO$_2$Max and neurocognitive outcomes among two groups of higher and lower prenatal exposure to methylmercury. We assessed the differences in associations using two cut-offs. In the first analysis, we split CB-Hg at the median (23.5 μg/L), whereas in the second analysis we split the CB-Hg at the 67th percentile (35 μg/L) and ran the analysis across two groups of low (< 35 μg/L; lower two tertiles) and high (≥ 35 μg/L; highest tertile) prenatal exposure to methylmercury. To account for residual confounding within mercury exposure strata, we included mercury concentrations (log$_{10}$transformed) as a covariate. Differences in the associations in the two exposure groups were tested by comparing the value of d(SE$_d$) to the standard normal distribution, where d is the difference between the two estimates, and SE$_d$ = $\sqrt{SE_1^2 + SE_2^2}$ is the standard error of the difference (Altman and Bland 2003). An attempt to run SEM analyses across tertiles of CB-Hg failed to produce an acceptable model fit with lower numbers of observations.

In a sensitivity analysis, we attempted to adjust for potential selection bias due to incomplete participation or due to the exclusion of the individuals who did not perform optimally in the fitness test. We therefore applied stabilized inverse probability weights to all the models (Hernán et al. 2004) to conduct a weighted analysis. Because available SEM packages in R (version 3.2.3; R Project for Statistical Computing) do not allow for weighted analyses, we included these weights as sampling weights in a complex survey SEM analysis, while restricting clusters and strata to be the same for all individuals.

Because of the cross-sectional nature of the follow-up study and to ensure that potential significant associations between VO$_2$Max and neurocognitive functions were not due...
Aerobic fitness, prenatal mercury exposure, and cognition

significantly higher among men, nonsmokers, and physically active participants. Cognitive test scores were adequate indicating an excellent state of health. No association was observed regarding CB-Hg. Among the 262 participants who underwent the aerobic fitness test, 18 participants performed an optimal test were not different from those who did not in regard to CB-Hg, except for motor function for which the standardized coefficient was weaker (0.25). A 1-SD increase was associated with a marginally significant increase in short-term memory scores by 0.21 SD (95% CI: –0.04, 0.46; z = 1.66, p = 0.10).

In the SEM model assessing the association with general neurocognitive function (g), the six latent functions showed significant correlations to the g, with standardized coefficients between 0.61 and 0.76, except for motor function for which the standardized coefficient was weaker (0.25). A 1-SD increase of memory for words, CVLT scores on the learning trials 1–5, visual matching, and decision speed scores (see Table S2).

In a confirmatory factor analysis, neurocognitive test scores were adequate indicators of latent functions with significant standardized coefficients ranging between 0.25 and 0.93 (Table 2).

### Associations between Aerobic Fitness and Neurocognitive Functions

Final models included VO\(_{2\text{Max}}\), sex, physical activity, smoking status, BMI, family background, and prenatal methylmercury exposure (CB-Hg).

Although VO\(_{2\text{Max}}\) was positively correlated with all neurocognitive domains, the association was statistically significant only for cognitive processing speed (Table 3). We found that a 1-SD increase in VO\(_{2\text{Max}}\) was significantly associated with an increase in cognitive processing speed by 0.28 SD [95% confidence interval (CI): 0.02, 0.54; z = 2.14, p = 0.04]. In addition, a 1-SD increase in VO\(_{2\text{Max}}\) was associated with a marginally significant increase in short-term memory scores by 0.21 SD (95% CI: –0.04, 0.46; z = 1.66, p = 0.10).

### Results

Among the 262 participants who underwent the aerobic fitness test, 18 participants failed the test, and 47 performed suboptimally. Thus, only 197 performed optimally. Women, nonsmokers, and physically active participants were significantly more likely to have an optimal aerobic fitness test. Participants who performed an optimal test were not different from those who did not in regard to CB-Hg, BMI, maternal and paternal employment and education/training, and maternal Raven score.

In univariate associations, VO\(_{2\text{Max}}\) was significantly higher among men, participants who had never smoked, individuals with normal BMI, and participants who reported being physically active and having an excellent state of health. No association with VO\(_{2\text{Max}}\) was observed regarding CB-Hg and maternal and paternal employment and education/training, nor with maternal Raven scores (Table 1).

All neurocognitive test scores exhibited appropriate variability, and were therefore adequate for subsequent analyses (see Table S1). Adjusting for sex, higher VO\(_{2\text{Max}}\) was significantly associated with higher scores of memory for words, CVLT scores on the learning trials 1–5, visual matching, and decision speed scores (see Table S2).

In a confirmatory factor analysis, neurocognitive test scores were adequate indicators of latent functions with significant standardized coefficients ranging between 0.25 and 0.93 (Table 2).

### Table 1. Levels of maximal oxygen uptake in relation to important covariates.

| Participant characteristics | n(%) | VO\(_{2\text{Max}}\) (ml/kg/min) (mean ± SD) | p Value |
|----------------------------|------|------------------------------------------|---------|
| Sex                        |      |                                          |         |
| Women                      | 77 (39) | 31.2 ± 6.2                              | < 0.001 |
| Men                        | 120 (61) | 37.6 ± 7.1                              |         |
| Smoking                    |      |                                          | 0.001   |
| Never                      | 113 (68) | 36.7 ± 7.2                              |         |
| Occasional                 | 30 (15) | 33.6 ± 6.5                              |         |
| Regular                    | 52 (27) | 32.4 ± 7.7                              |         |
| Missing                    | 2     |                                          |         |
| Prenatal mercury exposure  |      |                                          | 0.44    |
| Low (< 23.5 μg/L)          | 99 (50) | 34.7 ± 6.4                              |         |
| High (> 23.5 μg/L)         | 98 (50) | 35.5 ± 8.3                              |         |
| BMI                        |      |                                          | < 0.001 |
| < 25                       | 111 (66) | 37.7 ± 6.3                              |         |
| 25–29                      | 60 (31) | 34.3 ± 6.4                              |         |
| > 29                       | 26 (13) | 25.8 ± 5.9                              |         |
| Self-reported physical activity |      |                                          | < 0.001 |
| Practicing hard or competitive sports | 46 (24) | 40.5 ± 7.6                              |         |
| Doing sports some hours a week | 48 (24) | 36.1 ± 5.1                              |         |
| Walking or riding bicycle few hours a week | 75 (39) | 33.1 ± 8.3                              |         |
| Inactive                   | 27 (14) | 29.6 ± 7.4                              |         |
| Missing                    | 1     |                                          |         |
| Self-reported health status |      |                                          | < 0.001 |
| Excellent                  | 41 (21) | 37.7 ± 6.5                              |         |
| Very good                  | 78 (40) | 37.2 ± 6.3                              |         |
| Good                       | 71 (38) | 31.7 ± 7.5                              |         |
| Not especially good        | 6 (3)  | 30.0 ± 10.1                             |         |
| Missing                    | 1     |                                          |         |
| Maternal education/training|      |                                          | 0.88    |
| No                         | 71 (40) | 35.4 ± 7.6                              |         |
| Yes                        | 108 (60) | 35.2 ± 7.6                              |         |
| Missing                    | 18    |                                          |         |
| Paternal education/training|      |                                          | 0.88    |
| No                         | 43 (24) | 35.4 ± 8.4                              |         |
| Yes                        | 137 (76) | 35.2 ± 7.3                             |         |
| Missing                    | 17    |                                          |         |
| Maternal employment        |      |                                          | 0.23    |
| No                         | 83 (46) | 34.5 ± 7.7                              |         |
| Yes                        | 97 (54) | 35.9 ± 7.4                              |         |
| Missing                    | 17    |                                          |         |
| Paternal employment        |      |                                          | 0.70    |
| No                         | 28 (16) | 34.7 ± 7.4                              |         |
| Yes                        | 152 (84) | 35.3 ± 7.6                             |         |
| Missing                    | 17    |                                          |         |
| Maternal Raven score       |      |                                          | 0.76    |
| ≤ 45                       | 61 (35) | 34.8 ± 8.0                              |         |
| > 45                       | 62 (35) | 35.8 ± 7.1                              |         |
| > 51                       | 53 (30) | 35.1 ± 7.3                              |         |
| Missing                    | 21    |                                          |         |
| Total                      | 197 (100) | 35.1 ± 7.4                           |         |
in VO$_{2\text{Max}}$ was associated with an increased neurocognitive g score of 0.24 SD (95% CI: –0.02, 0.50; $z = 1.81$, $p = 0.07$). In the final model using two broad neurocognitive functions, aerobic fitness was significantly associated with cognitive efficiency. Here, a 1-SD increase in VO$_{2\text{Max}}$ was associated with a score increase of 0.32 SD (95% CI: 0.01, 0.62; $z = 2.10$, $p = 0.04$), although not in regard to general thinking abilities (Table 3).

**Effect Modification by Prenatal Methylmercury Exposure**

In multiple group SEMs comparing the two groups with low (< 23.5 μg/L) and high (≥ 23.5 μg/L) prenatal methylmercury exposure, we found a significant positive association between aerobic fitness and cognitive processing speed in the group with lower prenatal methylmercury exposure, with a 1-SD increase in VO$_{2\text{Max}}$ significantly associated with increased cognitive processing speed by 0.45 SD (95% CI: 0.08, 0.81; $z = 2.45$, $p = 0.01$). No clear association was observed for the other neurocognitive functions (Table 4). In the group with higher prenatal methylmercury exposure, we observed no association between VO$_{2\text{Max}}$ and neurocognitive functions (Table 4). Although the association between VO$_{2\text{Max}}$ and scores of cognitive processing speed was stronger among individuals with low prenatal methylmercury exposure compared to those with high exposure, the two groups did not differ significantly for any of the neurocognitive functions.

No significant association was found in the model assessing the association between VO$_{2\text{Max}}$ and the general neurocognitive function (g), and the associations were similar in the two groups of exposure, although it was marginally significant in the lower exposure group (0.27 SD; 95% CI: −0.05, 0.59; $z = 1.65$, $p = 0.10$) (Table 4). However, we found a significant positive association between VO$_{2\text{Max}}$ and cognitive efficiency in the group of low prenatal exposure, with a 1-SD increase in VO$_{2\text{Max}}$ significantly associated with increased cognitive efficiency score increase by 0.43 SD (95% CI: 0.07, 0.80; $z = 2.28$, $p = 0.02$). Again, no clear association was found in the group of higher prenatal exposure (0.24 SD; 95% CI: −0.20, 0.68; $z = 1.07$, $p = 0.28$). No significant association was observed between VO$_{2\text{Max}}$ and general thinking abilities in the two groups of lower and higher prenatal exposures (Table 4).

In the multiple group SEM analyses with a cut-off of prenatal methylmercury exposure of 35 μg/L (67% percentile), we observed the same trend as for the first cut-off at the median. Thus, we found a positive association between aerobic fitness and specific neurocognitive outcomes in the group with lower prenatal methylmercury exposure, with a 1-SD increase in VO$_{2\text{Max}}$ associated with increased short-term memory and cognitive processing speed by 0.28 SD (95% CI: 0.00, 0.56; $z = 1.96$, $p = 0.05$) and 0.46 SD (95% CI: 0.19, 0.74; $z = 3.22$, $p = 0.001$). No significant association was observed for other neurocognitive functions, and none in the group with higher prenatal methylmercury exposure. However, in contrast to the analyses with a median cut-off, the associations differed significantly between the two groups of low and high prenatal exposure for cognitive processing speed ($p = 0.007$), though not for the other neurocognitive outcomes (Table 5).

When assessing the general neurocognitive function (g), the associations were similar in the two groups of exposure, although marginally better scores were seen in the lower exposure group (0.27 SD; 95% CI: −0.02, 0.55; $z = 1.89$, $p = 0.06$) (Table 5). Again, we observed the same trend when comparing results in the tertile groups of prenatal exposure in regard to the cognitive efficiency and general thinking abilities. Thus, we found a positive association in subjects with prenatal methylmercury exposure within the two lower tertiles, where a 1-SD increase in VO$_{2\text{Max}}$ associated with increased cognitive thinking abilities in the two groups of lower exposure (0.24 SD; 95% CI: −0.02, 0.55; $z = 1.89$, $p = 0.06$) (Table 5). Again, we observed the same trend when comparing results in the tertile groups of prenatal exposure in regard to the cognitive efficiency and general thinking abilities. Thus, we found a positive association in subjects with prenatal methylmercury exposure within the two lower tertiles, where a 1-SD increase in VO$_{2\text{Max}}$ associated with increased cognitive thinking abilities in the two groups of lower exposure (0.24 SD; 95% CI: −0.02, 0.55; $z = 1.89$, $p = 0.06$).

### Table 2. Factor loadings and estimated correlation of measured test scores to the neurocognitive latent functions as previously defined (Debes et al. 2016).

| Domain/neurocognitive test | Factor loading | SE | p-Value | Standardized coefficient |
|----------------------------|----------------|----|---------|-------------------------|
| Gm Numbers reversed        | 1              | 0  | NA      | 0.85                    |
| Gm Memory for words        | 0.35           | 0.08| < 0.001 | 0.59                    |
| Gm Spatial span forward    | 0.25           | 0.08| 0.002   | 0.28                    |
| Gm Spatial span backward   | 0.24           | 0.08| < 0.001 | 0.34                    |
| Gc Boston Naming Test, correct without cues | 1          | 0  | NA      | 0.78                    |
| Gc Boston Naming Test, correct with cues | 0.86         | 0.03| < 0.001 | 0.79                    |
| Gc Synonyms                | 0.43           | 0.05| < 0.001 | 0.78                    |
| Gc Antonyms                | 0.25           | 0.04| < 0.001 | 0.62                    |
| Gc Verbal analogies        | 0.32           | 0.06| < 0.001 | 0.67                    |
| Gps Finger tapping, dominant hand | 1          | 0  | NA      | 0.93                    |
| Gps Finger tapping, non-dominant hand | 0.86         | 0.11| < 0.001 | 0.70                    |
| Gps Finger tapping, alternate hands | 0.99         | 0.22| < 0.001 | 0.55                    |
| Gv Block Design, WISC-R    | 1.66           | 0.20| < 0.001 | 0.66                    |
| Gv Spatial Relations       | 0.74           | 0.17| < 0.001 | 0.70                    |
| Glr CVLT, trial 1, number correct | 1          | 0  | NA      | 0.59                    |
| Glr CVLT, learning trials 1–5, total number correct | 6.92          | 0.72| < 0.001 | 0.83                    |
| Glr CVLT, list B, number correct | 0.72         | 0.13| < 0.001 | 0.38                    |
| Glr CVLT, short delay, free recall, correct | 1.72          | 0.28| 0.001   | 0.91                    |
| Glr CVLT, long delay, free recall, correct | 1.78          | 0.30| < 0.001 | 0.92                    |
| Glr CVLT, long delay, recognition, correct | 0.45          | 0.11| < 0.001 | 0.42                    |
| Glr Incidental Memory      | 0.83           | 0.24| 0.001   | 0.25                    |
| Gs Block Design, WISC-R    | 1              | 0  | NA      | 0.83                    |
| Gs Decision Speed          | 0.56           | 0.09| < 0.001 | 0.73                    |

*For each neurocognitive function, the latent variable is constructed on the scale of the first component.

### Table 3. Adjusted associations between VO$_{2\text{Max}}$ and neurocognitive functions.

| Neurocognitive domain | B (95% CI) | p-Value |
|-----------------------|------------|---------|
| Short-term memory     | 0.21 (–0.04, 0.46) | 0.10    |
| Verbal comprehension and knowledge | 0.12 (–0.11, 0.34) | 0.31    |
| Psychomotor speed     | 0.03 (–0.18, 0.24) | 0.78    |
| Visual processing     | 0.07 (–0.24, 0.38) | 0.64    |
| Long-term storage and retrieval | 0.16 (–0.06, 0.38) | 0.16    |
| Cognitive processing speed | 0.28 (0.02, 0.54)** | 0.04    |
| Cognitive efficiency  | 0.32 (0.01, 0.62)** | 0.04    |
| General thinking abilities | 0.16 (–0.10, 0.42) | 0.23    |
| General function (g)  | 0.24 (–0.02, 0.50)* | 0.07    |

Models were adjusted for sex, physical activity, smoking status, BMI, family background, and prenatal methylmercury exposure.

*Change in the standard deviation of the neurocognitive function associated with a 1-SD increase in VO$_{2\text{Max}}$.
efficiency (0.49 SD; 95% CI: 0.20, 0.79; \( z = 3.20, p = 0.001 \)), but not general thinking abilities. The associations between VO_{2\text{Max}} and cognitive efficiency differed significantly between the highest tertile and the two lower tertiles of prenatal exposure (\( p = 0.03 \)).

In the sensitivity analysis, estimates of associations adjusting for inverse probability weights led to similar conclusions as those drawn from main analyses, although the estimates were slightly attenuated (see Tables S3, S4, and S5).

**Associations between Neurocognitive Functions at Age 14 and Aerobic Fitness at Age 22 Years**

The general neurocognitive score at age 14 was significantly correlated with the six latent cognitive functions at 22 years with correlations ranging from 0.24 (motor function) to 0.91 (short-term memory). Analyses assessing reverse causation showed no association between neurocognitive scores at 14 years of age and aerobic fitness at 22 years, with changes of 0.09, –0.04, and 0.06 SD in VO_{2\text{Max}} at 22 years for a 1 SD increase in age 14 neurocognitive factors 1, 2, and 3 (\( p = 0.25, 0.54, \) and 0.57, respectively). Likewise, a 1-SD increase in the general neurocognitive score at age 14 was not associated with VO_{2\text{Max}} at age 22 years (standardized \( \beta = 0.11 \) SD; 95% CI: –0.11, 0.34).

**Discussion**

In the present study, aerobic fitness showed positive associations with scores of cognitive processing speed at age 22 years, but not in regard to other neurocognitive functions. The results also exhibited marginally significant associations for short-term memory and general neurocognitive function. Thus, a 1-SD increase in maximal oxygen uptake (mL/kg/min) was associated with increases in scores by 24%, 28%, and 21% of the SD for general neurocognitive function, cognitive processing speed and short-term memory, respectively. These results extend findings from previous studies on associations between aerobic fitness and neurocognitive functions (Chaddock-Heyman et al. 2014; Colcombe and Kramer 2003; Colcombe et al. 2006; Desai et al. 2015; Erickson and Kramer 2009; Erickson et al. 2011; Sardinha et al. 2014). Of particular interest, the positive association of aerobic fitness and cognitive processing speed and short-term memory appeared to be constrained to individuals with lower prenatal exposure to methylmercury. Furthermore, higher aerobic fitness in adulthood did not appear to be driven by better cognitive functioning during adolescence, thus arguing against reverse causation. Our study is the first to address the modifying effect of prenatal exposure to neurotoxins on the potential positive relationship between aerobic fitness and cognitive functions.

Aerobic fitness was associated significantly only with cognitive efficiency scores indicated by cognitive processing speed and short-term memory, thereby suggesting that the influence of aerobic fitness on neurocognitive function might be specific for certain functional domains. Although test batteries vary widely between different studies, and domain-representation differs between tests, previous studies suggest that tests involving response speed may be the most sensitive to aerobic exercise and fitness (Etien et al. 2006; Hillman et al. 2005; Smith et al. 2010, 2013). Spatial learning and memory may benefit more than verbal learning and delayed memory recall (Herting and Nagel 2012). These findings appear to be in accordance with the notion that exercise benefits memory encoding in the hippocampus, but not retrieval involving other brain regions (Rugg et al. 2002).

Several mechanisms might explain the link between aerobic fitness and neurocognitive functioning (Hötting and Röder 2013). Aerobic fitness has been shown to affect the hippocampus through increases in cerebral blood volume (Pereira et al. 2007) and densities of neuronal synapses (Kramer et al. 2002), in addition to the stimulation of neurogenesis (van Praag 2008; van Praag et al. 1999). After inhibition of neurogenesis, one study in mice showed no exercise-related enhancement in memory performance and spatial learning (Clark et al. 2008). Because neurogenesis must arise from a local neural stem cell population (Christian et al. 2014), the plasticity of the hippocampal neuronal circuit can be negatively affected by a decreased availability of neuronal stem cells.

The positive association between aerobic fitness and neurocognitive functioning appeared to be constrained mainly to individuals with lower prenatal methylmercury exposure, and no clear association was observed in individuals highly exposed to methylmercury in the womb. Because aerobic fitness is likely to exert its effect through neurogenesis, it is possible that prenatal exposure to methylmercury affected the cellular basis for this process. In vitro studies suggest that neural stem cells are exceedingly sensitive to adverse effects from mercury exposure (Guindacker et al. 2012; Tamm et al. 2006).

In animal models, exposure to methylmercury during the perinatal period resulted in decreased neural stem cell populations (Sokolowski et al. 2013), inhibited hippocampal DNA synthesis, degradation of cyclin E, and reduced cyclin D1 expression (Christian et al. 2014). The plasticity of the hippocampal neuronal circuit can be negatively affected by a decreased availability of neuronal stem cells.

**Table 4. Adjusted associations between VO_{2\text{Max}} and neurocognitive functions in regard to prenatal mercury exposure split at the median (23.5 μg/L).**

| Neurocognitive domain                  | Low prenatal exposure (\(< 23.5 \mu g/L\)) | High prenatal exposure (\(\geq 23.5 \mu g/L\)) | Difference (95% CI) |
|--------------------------------------|------------------------------------------|-----------------------------------------------|---------------------|
| Short-term memory                    | 0.18 (–0.14, 0.50)                       | 0.18 (–0.23, 0.60)                            | 0.98                |
| Verbal comprehension and knowledge   | 0.08 (–0.20, 0.35)                       | 0.11 (–0.31, 0.52)                            | 0.91                |
| Psychomotor speed                    | 0.12 (–0.16, 0.40)                       | –0.07 (–0.38, 0.25)                           | 0.24                |
| Visual processing                    | 0.11 (–0.21, 0.43)                       | –0.02 (–0.53, 0.49)                           | 0.60                |
| Long-term storage and retrieval      | 0.16 (–0.13, 0.43)                       | 0.19 (–0.15, 0.52)                            | 0.90                |
| Cognitive processing speed           | 0.45 (0.06, 0.81)**                      | 0.16 (–0.24, 0.56)                            | 0.20                |
| Cognitive efficiency                 | 0.43 (0.07, 0.80)**                      | 0.24 (–0.20, 0.68)                            | 0.53                |
| General thinking abilities           | 0.15 (–0.17, 0.47)                       | 0.15 (–0.22, 0.52)                            | 0.97                |
| General function (g)                 | 0.27 (–0.05, 0.59)                       | 0.18 (–0.18, 0.54)                            | 0.69                |

Models were adjusted for sex, physical activity, smoking status, BMI, family background, and prenatal methylmercury exposure. *Change in the standard deviation of the neurocognitive function associated with a 1 SD increase in VO_{2\text{Max}}.* ** \( p < 0.05 \).

**Table 5. Adjusted associations between VO_{2\text{Max}} and neurocognitive functions in regard to prenatal mercury exposure split at the 67th percentile (35 μg/L).**

| Neurocognitive domain                  | Low prenatal exposure (\(< 35 \mu g/L\)) | High prenatal exposure (\(\geq 35 \mu g/L\)) | Difference (95% CI) |
|--------------------------------------|------------------------------------------|-----------------------------------------------|---------------------|
| Short-term memory                    | 0.27 (0.00, 0.56)**                      | –0.08 (–0.55, 0.40)                           | 0.21                |
| Verbal comprehension and knowledge   | 0.01 (–0.27, 0.29)                       | 0.14 (–0.25, 0.53)                            | 0.57                |
| Psychomotor speed                    | 0.10 (–0.04, 0.23)                       | –0.15 (–0.44, 0.14)                           | 0.12                |
| Visual processing                    | 0.25 (–0.06, 0.56)                       | –0.17 (–0.57, 0.22)                           | 0.09                |
| Long-term storage and retrieval      | 0.09 (–0.16, 0.34)                       | 0.17 (–0.13, 0.47)                            | 0.69                |
| Cognitive processing speed           | 0.47 (0.19, 0.74)**                      | –0.12 (–0.44, 0.21)                           | 0.007               |
| Cognitive efficiency                 | 0.49 (0.20, 0.79)**                      | –0.12 (–0.58, 0.33)                           | 0.03                |
| General thinking abilities           | 0.08 (–0.19, 0.35)                       | 0.27 (–0.17, 0.71)                            | 0.47                |
| General function (g)                 | 0.27 (–0.02, 0.55)**                     | 0.13 (–0.21, 0.47)                            | 0.54                |

Models were adjusted for sex, physical activity, smoking status, BMI, family background, and prenatal methylmercury exposure. *Change in the standard deviation of the neurocognitive function associated with a 1 SD increase in VO_{2\text{Max}}.* ** \( p < 0.10 \). *** \( p < 0.05 \).
and D3 during postnatal development in which neurogenesis is highly active in the hippocampus (Tyler and Allan 2013). It also induced apoptotic cell death (Falluel-Morel et al. 2007). All of these effects may result in subsequent deficits in hippocampal structure and function. Additionally, exposure to methylmercury has been reported to disturb the serotonergic system (Oudar et al. 1989), which plays a direct regulatory role in exercise-dependent hippocampal neurogenesis (Klempin et al. 2013).

Due to the limited number of cohort members included in this study, details on the exposure dependence of such effects cannot be estimated. Significant differences were seen only when comparing the highest exposures to the rest of the cohort. Within this cohort, about 90% of the subjects were prenatally exposed to methylmercury at levels exceeding the reference dose recommended by the U.S. Environmental Protection Agency (5.8 μg/L), but the study size did not allow meaningful comparisons between subjects with low background exposures to those with elevated exposures. Despite the nonsignificant effect modification reported when comparing participants above the median prenatal methylmercury exposure to those below the median, we speculate that stem cell–mediated toxicity may occur well below the median level of prenatal methylmercury exposure in the present study, given the overall tendencies and the well-documented and extreme sensitivity of neural stem cells to mercury exposure.

This study has some limitations. The cross-sectional design at the age 22 years follow-up examination does not allow causal inference, but we were able to exclude an impact of better cognitive outcomes at age 14 on aerobic capacity 8 years later. However, because fewer tests were administered at age 14 years, we were not able to directly compare the same neurocognitive functions, so reverse causation could not be completely ruled out. Exposure to methylmercury was assessed from the total mercury concentration in cord blood as a measure of the exposure assessed from the total mercury concentration in cord blood as a measure of the exposure.
Aerobic fitness, prenatal mercury exposure, and cognition

Herting MM, Nagel BJ. 2012. Aerobic fitness relates to prenatal mercury exposure. PLoS One 10:e0122487, doi: 10.1371/journal.pone.0122487.

Erickson KI, Kramer AF. 2009. Aerobic exercise effects on cognitive and neural plasticity in older adults. Br J Sports Med 43:22–24.

Erickson KI, Voss MW, Prakash RS, Basak C, Szabo A, Chaddock L, et al. 2011. Exercise training increases size of hippocampus and improves memory. Proc Natl Acad Sci U S A 108:2017–2022.

Enlert JL, Nowell PM, Landers DM, Sibley BA. 2006. A meta-regression to examine the relationship between aerobic fitness and cognitive performance. Brain Res Rev 52:119–130.

Falluel-Morel A, Sokolowski K, Sisti HM, Zhou X, Shors TJ, Dicicco-Bloom E. 2007. Developmental mercury exposure elicits acute hippocampal cell death, reductions in neurogenesis, and severe learning deficits during puberty. J Neurochem 103:1988–1981.

Hernán MA, Hernández-Díaz S, Robins JM. 2004. A structural approach to selection bias. Epidemiology 15(5):615–625.

Grandjean P, Budtz-Jørgensen E. 2014. Neurotoxicity from prenatal and postnatal exposure to methylmercury. Neurotoxicol Teratol 43:39–44.

Grandjean P, Weihe P, Jorgensen E. 2007. Total impregnation of exposure biomarkers: implications for calculating exposure limits. Am J Ind Med 50:712–719.

Grandjean P, Landrigan PJ. 2014. Neurobehavioural effects of developmental toxicity. Lancet Neurol 13:330–338.

Grandjean P, Weihe P, Debes F, Choi AL, Budtz-Jørgensen E. 2014. Neurotoxicity from prenatal and postnatal exposure to methylmercury. Neurotoxicol Teratol 43:39–44.

Herting MM, Nagel BJ. 2012. Aerobic fitness relates to learning on a virtual Morris Water Task and hippocampal volume in adolescents. Behav Brain Res 233:517–525.

Hillman CH, Castelli DM, Buck SM. 2005. Aerobic fitness and neurocognitive function in healthy preadolescent children. Med Sci Sports Exerc 37:1967–1974.

Hindin SB, Zelinski EM. 2012. Extended practice and aerobic exercise interventions benefit untrained cognitive outcomes in older adults: a meta-analysis. J Am Geriatr Soc 60:138–141.

Hötting K, Rieder B. 2013. Beneficial effects of physical exercise on neuroplasticity and cognition. Neurosci Biobehav Rev 37(8 pt B):2245–2257.

Kaplan E, Goodglass H, Weintraub S. 1983. Boston Naming Test. Philadelphia, PA: Lea & Febiger.

Karagas MR, Choi AL, Oken E, Horvat M, Schoeny R, Kamai E, et al. 2012. Evidence on the human health effects of low-level methylmercury exposure. Environ Health Perspect 120:789–806.

Kipling F, Beis D, Mosienko V, Kempermann G, Bader M, Alenina N. 2013. Serotonin is required for exercise-induced adult hippocampal neurogenesis. J Neurosci 33:8270–8275.

Kramer AF, Colcombe S, Erickson K, Belopolsky A, McAuley E, Cohen NJ, et al. 2002. Effects of aerobic exercise training on human cortical function: a proposal. J Mol Neurosci 19:227–231.

Oberski D. 2014. lavaan.survey: an R package for complex survey analysis of structural equation models. J Stat Softw 57(1):1–27.

Oudar D, Caillard L, Fillion G. 1989. In vivo effect of organic and inorganic mercury on the serotonergic system. Pharmacol Toxicol 65:245–248.

Pereira AC, Huddleston DE, Brickman AM, Sosnowy AA, Hen R, McKhann GM, et al. 2007. An in vivo correlate of exercise-induced neurogenesis in the adult dentate gyrus. Proc Natl Acad Sci U S A 104:5638–5643.

Pescatello LS, American College of Sports Medicine. 2014. ACSM’s Guidelines for Exercise Testing and Prescription. Philadelphia, PA: Wolters Kluwer/Lippincott Williams & Wilkins Health.

Rosselé V. 2012. lavaan: an R package for structural equation modeling. J Stat Softw 48:1–36.

Rubin DB. 1987. Multiple Imputation for Nonresponse in Surveys. New York: Wiley.

Rugg MD, Otten LJ, Henson RNA. 2002. The neural basis of episodic memory: evidence from functional imaging. Philos Trans R Soc Lond B Biol Sci 357:1097–1110.

Sardinha LB, Marques A, Martins S, Palmeira A, Minderico C. 2014. Fitness, fatness, and academic performance in seventh-grade elementary school students. BMC Pediatr 14:176, doi: 10.1186/1471-2431-14-176.

Schneider WJ, McGrew K. 2012. The Cattell-Horn-Carroll model of intelligence. In: Contemporary Intellectual Assessment: Theories, Tests, and Issues. 3rd ed. Flanagan DP, Harrison PL, eds. New York: Guilford Press, 99–144.

Shepherd RJ. 1984. Tests of maximum oxygen intake. A critical review. Sports Med 1:99–124.

Smith PJ, Blumenthal JA, Hoffman BM, Cooper H, Strauman TA, Welsh-Bohmer K, et al. 2010. Aerobic exercise and neurocognitive performance: a meta-analytic review of randomized controlled trials. Psychosom Med 72:239–252.

Smith PJ, Potter GG, McLaren ME, Blumenthal JA. 2013. Impact of aerobic exercise on neurobehavioral outcomes. Ment Health Phys Act 6:139–153.

Sokolowski K, Obiorah M, Robinson K, McCandlish E, Buckley B, DiCicco-Bloom E. 2012. Neural stem cell apoptosis after low-methylmercury exposures in postnatal hippocampus produce persistent cell loss and adolescent memory deficits. Dev Neurobiol 73:936–949.

Tamm C, Duckworth J, Hermanson O, Cecattelli S. 2006. High susceptibility of newborn stem cells to methylmercury toxicity: effects on cell survival and neuronal differentiation. J Neurochem 97:69–78.

Tyler CR, Allan AM. 2013. Adult hippocampal neurogenesis and mRNA expression are altered by perinatal arsenic exposure in mice and restored by brief exposure to enrichment. PLoS One 8:e73720, doi: 10.1371/journal.pone.0073720.

van Buuren S, Groothuis-Oudshoorn K. 2011. mice: multivariate imputation by chained equations in R. J Stat Softw 45:1–67.

Van der Wal WM, Geskus RB. 2011. ipw: an R package for inverse probability weighting. J Stat Softw 43(1):1–23.

van Praag H. 2008. Neurogenesis and exercise: past and future directions. Neuromolecular Med 10:128–140.

van Praag H, Kempermann G, Gage FH. 1999. Running increases cell proliferation and neurogenesis in the adult mouse dentate gyrus. Nat Neurosci 2:266–270.

Wechsler D. 1974. Wechsler Intelligence Scale for Children–Revised. New York: Psychological Corp.

Wechsler D. 1981. Manual of the Wechsler Adult Intelligence Scale–Revised. San Antonio, TX: Psychological Corp.

Wechsler D. 1997. Wechsler Memory Scale–3rd Edition (WMS-III). San Antonio, TX: Psychological Corp.

White IR, Royston P, Wood AM. 2011. Multiple imputation using chained equations: issues and guidance for practice. Stat Med 30:377–399.

Woodcock RW, McGrew KS, Mather N. 2001. Woodcock Johnson III Tests of Cognitive Abilities. Itasca, IL: Riverside Publishing.