Early life factors and structural brain network in children with overweight/obesity: The ActiveBrains project

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Category of study

Clinical

Impact statement

- Birth weight and birth length, but not breastfeeding, were associated with several structural brain networks involving the cerebellum, cingulate gyrus, occipital pole, and subcortical structures including hippocampus, caudate, putamen, pallidum, accumbens and amygdala in children with overweight/obesity, playing a role for a normal brain development.
- Despite no academic consequences, other behavioral consequences should be investigated. Interventions aimed at improving optimal intrauterine growth and development may be of importance to achieve a healthy brain later in life.

Abstract

BACKGROUND The aims of this study were to investigate the association of early life factors, including birth weight, birth length, and breastfeeding practices with structural brain networks; and to test whether structural brain networks associated with early life factors were also associated with academic performance in children with overweight/obesity (OW/OB).

METHOD 96 children with OW/OB aged 8-11 years (10.03±1.16) from the ActiveBrains project were included. Early life factors were collected from birth records and reported by parents as weight, height and months of breastfeeding. T1-weighted images were used to identify structural networks using a non-negative matrix factorization approach. Academic performance was evaluated by the Woodcock-Muñoz standardized test battery.

RESULTS Birth weight and birth length were associated with 7 networks involving the cerebellum, cingulate gyrus, occipital pole, and subcortical structures including hippocampus, caudate nucleus, putamen, pallidum, nucleus accumbens and amygdala. No associations were found for breastfeeding practices. None of the networks linked to birth weight and birth length were linked to academic performance.

CONCLUSIONS Birth weight and birth length, but not breastfeeding, were associated with brain structural networks in children with OW/OB. Thus, early life factors are related to brain networks, yet a link with academic performance was not observed.

Introduction

Early life experiences are predictive of brain health later in life [1]. Fetal and postnatal environments are crucial for brain maturation and development of brain structures with long lasting consequences. Early life factors such as anthropometric neonatal data (i.e., birth weight, birth length) as well as breastfeeding practices (i.e., exclusive and any breastfeeding) are indicators of the fetal environment and the first stage of life, and have been associated with brain health related outcomes at childhood such as gray matter volumes [2,3], white matter integrity[4,5], and structural and functional brain connectivity [6].

Using a whole brain voxel-wise gray matter approach, we previously demonstrated that birth weight and length were associated with gray matter volumes in childhood [2] in discrete regions of the brain, such as cerebellum, middle frontal and temporal gyrus or occipital regions [2,5]. This association was not restricted to specific or physically connected brain regions [7]; thus, it is relevant to examine whether these or other brain regions may be related and influenced in parallel by early life factors. Structural covariance analysis provides information about the relationship between brain regions that are not physically connected with one another, and models brain structure as a complex network [8,9] providing a more comprehensive approach to describe the interrelationship of brain regions [7].

Examining connectivity patterns across regions offers a wider approach to investigate changes in brain networks related to several behavioral factors. Studies related to structural covariance networks use different approaches including seed analysis, principal component analysis or graph analysis [8]. The downstream impact of early life factors on structural networks have recently started to be explored [7,10,11]. For example, prematurity at birth contributes to alterations in several gray matter structural networks among adolescents [10], and young adults, with

implications in language development [11]. However, breastfeeding practices, powerful early postnatal factors due to breast milk provide key nutrients necessary for myelinization as well as brain maturation, have been much less frequently studied. While few studies have investigated the relationship with white matter connectivity [12] or functional connectivity [6], no study has explored the relationship of breastfeeding practices with structural gray matter networks. Thus, further research related to structural gray matter networks is needed to provide a broad understanding of the influence of early life on gray matter, particularly in children with overweight or obesity.

Obesity has been linked to structural brain networks abnormalities, coupled with cognitive implications[13,14]. For example, using covariance network analysis, obesity measures have been related to less gray matter volume in several networks, including fronto-temporal and fronto-limbic regions involved in memory and affective processing [15]. Further, obesity is linked to alterations in gray matter across regions implicated in executive function and higher-order cognitive processes [16]. A relevant real-world implication of these brain differences and cognitive processes during childhood is academic performance, which is critical for long-term wellbeing and productivity, and is diminished in children with overweight and obesity [17]. Thus, it is necessary to examine the relationship between early life factors, structural covariance network and academic performance in the context of childhood obesity.

Thus, the aims of this study were (i) to investigate the association of early life factors, namely anthropometric neonatal data (birth weight, birth length), and breastfeeding practices (exclusive and any breastfeeding) with structural brain networks; and (ii) to test whether structural brain networks associated with early life factors are also associated with academic performance in children with overweight/obesity.

Methods

Participants

In this cross-sectional analysis we used the baseline data from 96 children (aged 8-11 years) with overweight/obesity [18] from the ActiveBrains randomized clinical trial [19] (www.profith.ugr.es/activebrains) with valid data from early life factors, brain structural network and academic performance variables (see **Table 1** for n in each variable). Measurements were carried out from November 2014 to February 2016. In short, eligibility criteria were: (a) the absence of physical disabilities or neurological disorders, (b) (for girls) not having started menstruation at the time of initial assessment, c) being right-handed, and (d) not currently having Attention-Deficit Hyperactivity Disorder. Parents or legal guardians signed written informed parental consents were obtained and were informed about the purpose of the ActiveBrains randomized clinical trial. The ActiveBrains project was approved by the Ethics Committee on Human Research (CEIH) of the University of Granada and was registered in ClinicalTrials.gov (identifier: NCT02295072).

Early life factors

Birth weight (kg) and birth length (cm) were obtained from the health records and parents were asked about breastfeeding practices according to the protocols of the ActiveBrains project [2]. Briefly, parents reported the duration in months (as a continuous scale) of exclusive (neither formula or other liquid) and any (combined with other liquid, formula, or solid) breastfeeding.

Academic performance

Academic performance was assessed using the version III of the Woodcock-Muñoz Tests of Achievement (i.e., Spanish version of the Woodcock- Johnson III) [20]. Each administration lasted

between 100 and 120 min, and instructions were provided individually during the session to each child. Indicators of reading, writing, mathematics, and a total achievement standard score (including mathematics, reading and writing) were used as measures of academic performance.

Covariates

Covariates were chosen in alignment with previous studies in this sample [2,5,21]. Thus, biological sex, gestational age, peak height velocity (PHV), parental educational level and cardiorespiratory fitness (CRF) were included as covariates. Gestational age in weeks was collected from the birth records. PHV was calculated and used as pubertal maturity status [22]. The difference with chronological age was defined as a value of maturity offset. Both parents were asked to report their maximum completed level of education and answers were categorized as: (0) none of the parents had a university degree, (1) one of the parents had a university degree or (2) both parents had a university degree [23]. We included CRF (a measure of aerobic capacity) according to a previous volumetric analysis of this sample showing an association with gray matter volume [24]. Thus, CRF was obtained using 20-m shuttle-run test and maximal oxygen consumption (VO2max, mL/kg/min) and calculated by the Leger formulae [25]. Additionally, sensitivity analyses were performed excluding preterm (gestational age <37 weeks) children.

Magnetic resonance imaging (MRI)

Acquisition and processing

Details regarding MRI methods have been published previously [24]. Briefly, MRI data were obtained using a 3.0 Tesla Siemens Magnetom Tim Trio scanner (Siemens Medical Solutions, Erlangen, Germany). Whole-brain T1-weighted images were acquired using a magnetization-prepared rapid gradient- echo (MPRAGE) sequence (repetition time (TR) = 2300 ms, echo time

(TE) = 3.1 ms, inversion time (TI) = 900 ms, flip angle = 9º, Field of view (FOV) = 256 x 256, acquisition matrix = 320 x 320, 208 slices, resolution = 0.8 x 0.8 x 0.8 mm, scan duration 394 s.) The processing protocol included quality control, alignment and segmentation into gray matter tissue, white matter tissue and cerebrospinal fluid. In addition, Montreal Neurological Institute (MNI) space was used for spatial normalization and to create a template using Diffeomorphic Anatomical Registration Through Exponentiated Lie algebra (DARTEL). Finally, a normalization to the DARTEL template using a non-linear transformation and modulated with Jacobian determinants were performed; and images were smoothed by convolving them with an isotropic Gaussian kernel of 8 mm full width at half maximum (FWHM). Further detail can be found in our previous work [24].

Non-negative Matrix Factorization analysis

Non-negative Matrix Factorization (NNMF) analysis was used to identify structural networks. NNMF is a method for extracting structural networks where volume covaries across all participants [9]. NNMF is not constrained by a limited set of predetermined anatomical regions, it offers a more extensive representation of covariance networks. Networks obtained through NNMF display exceptional reproducibility, and enhance statistical power by constraining the need for numerous comparisons. This means that we can identify not only individual regions but also how they interact and form networks, shedding light on the underlying mechanisms of the observed relationships between clinical variables and brain structure [26]. An extended function of NNMF was used corresponding to the orthonormal projective non-negative matrix factorization (OPNMF), which was run using "*opnmf_mem*" in MATLAB with code available in https://github.com/asotiras/brainparts. To build a more consistent network, available data from

both time points (pre- and post) smoothed structural gray matter images for each subject were reshaped into a matrix (dimensions: 198 participants x 2122945 voxels) [27]. The local grey matter volumes with a threshold of 0.2 (i.e., to eliminate the voxels with partial volume effect) were then extracted in a whole-brain grey matter mask. Then, we used it as input for OPNMF (dimensions: 198 participants x 470556 voxels) and approximated this matrix as a product of two matrices with non-negative elements.

The data were represented denoting the corresponding sparse components (W) and the subject-specific loading coefficients (H). The first matrix, W, is of size V × K and contains the estimated non-negative networks and their respective loadings on each of the V voxels; and K is the specified number of networks. The W matrix, or "Network Components," is composed of coefficients that denote the relative contribution of each voxel in the network. The second matrix, H, is of size K × N and contains subject-specific loading coefficients for each network. These subject-specific coefficients indicate the contribution of each network in reconstructing the original gray matter map. To obtain a range of possible solutions for comparison [9,26], we ran multiple NMF solutions requesting a K from 6 to 24 networks in steps of two (i.e., K=6:2:24). We then calculated the reconstruction error for each solution as the Frobenius norm and plotted the reconstruction error for all solutions. The solution resulted in 20 networks is shown in the **supplemental Figure S1.**

Statistical analysis

Descriptive characteristics are presented as mean and standard deviation (SD) for continuous variables, and absolute (n) and relative frequencies (%) for categorical variables. In a first step, we conducted multiple linear regressions using available data for each early life factor such as anthropometric neonatal data (i.e., birth weight, birth length), and breastfeeding practices

(i.e. exclusive and any breastfeeding -in a continuous manner in months-) as explicative variables and each structural network solution (specific loading coefficient -H- for each network) as the explained variable. Covariates used in all models were sex, gestational age, PHV, parental education level and CRF. Because of the number of comparisons (20 networks for each explicatory variable), a false discovery rate (FDR) [28] correction was applied to the results across each early life factor using the "p.adjust" function in R. In a second step, we conducted multiple linear regressions within those networks previously associated with early life factors after FDR correction ($p < .05$) and academic performance using sex, PHV, parental education level and CRF as covariates. Additional analysis including BMI were conducted. A false discovery rate (FDR) correction was also applied to the association between networks and academic performance (corresponding to 28 comparisons). All statistical analyses were performed in R (version 3.6.1; R Foundation for Statistical Computing, Vienna, Austria), and statistical significance was set at $p < .05$.

Results

Table 1 displays participants characteristics. The regions corresponding to the 20 structural covariance solutions are shown in **supplemental Figure S1**.

Figure 1 and supplemental table S1 shows significant associations between anthropometric neonatal data and structural networks after FDR correction. Results showed that birth weight and birth length were positively associated with seven different structural networks. Specifically, both indicators had overlapping significant associations with five different networks involving principally the cerebellum, cingulate gyrus, occipital pole, and subcortical structures including hippocampus, caudate nucleus, putamen, pallidum, nucleus accumbens and amygdala,

and unique associations with network 4 (for birth weight) and network 7 (for birth length), involving frontal medial cortex, paracingulate and anterior cingulate gyrus; and occipital, calcarine cortex and lingual gyrus**, respectively (Figure 1)**. Both early life factors were associated with cerebellar, striatal and limbic networks. Birth weight specifically correlated with a midline frontal network, and birth length with a posterior cortical network. The association with the 20 structural networks can be found in **supplemental Table S2.** We did not find significant associations between breastfeeding practices and structural networks after FDR correction. The association of the breastfeeding practices with the 20 structural networks can be found in **supplemental table S3**. The findings remained consistent after excluding preterm-born children (n=17) from the analysis and including BMI as covariate (data not shown).

Table 2 shows the association between the networks previously related to anthropometric neonatal data (after FDR correction) and academic performance. Only an association between the network 12 (Posterior cingulate gyrus and anterior precuneus cortex) and reading skills ($\beta = 0.245$; $p = 0.025$) was found, although this association disappeared after FDR correction.

Discussion

In this cross-sectional study, we computed the associations of early life factors with structural covariance networks in a sample of children with overweight or obesity. Our main finding was that both birth weight and birth length were consistently associated with structural brain networks involving the cerebellum, cingulate gyrus, occipital pole, and subcortical structures including hippocampus, caudate nucleus, putamen, pallidum, nucleus accumbens and amygdala. However, we did not find associations between breastfeeding practices and brain structural networks. Moreover, the implication on academic outcomes remains inconclusive.

Previous research including our own, support the selective vulnerability of cerebellum (networks 1 and 3) to variables of the early life stage [2,29,30]. Of note, the cerebellum also is a region linked to vulnerability in an obese population [30-32]. Thus, this suggests that early life factors contribute to structural cerebellar-related dysfunctions in children with obesity, which may have implications for motor and cognitive development [30]. On the other hand, regions implicated in the network 4 (Frontal medial cortex, paracingulate and anterior cingulate gyrus) have been recently highlighted as being associated with reduced gray matter volume in obesity [33], and with previous evidence related to early life factors [34]. Hence, regions of network 4 (frontal medial cortex, paracingulate and anterior cingulate gyrus) are implicated in emotional processes including social behaviors, decision-making and emotional responses [35,36]. Interestingly, the regions included in network 7 (occipital pole, supracalcarine cortex, intracalcarine cortex and lingual gyrus) were also linked to birth length in our prior work, reinforcing the idea of the long-term consequence of fetal environment on regions supporting perceptual processing [2,37-39]. The network 9 involved subcortical structures (caudate nucleus, putamen, pallidum, nucleus accumbens and amygdala) implicated in supporting sensorimotor, limbic, and cognitive information processing [40]. Accordingly, prematurity has been associated with a similar set of subcortical / forebrain structures, suggesting downstream effects of prematurity on striatal and thalamic integrity in childhood. Finally, the network including hippocampus and parahippocampal gyrus (network 19) are characterized as memory-related regions and are also known as specific areas disrupted by early life stress [41].

To our knowledge, only one previous study has examined structural covariance analysis in relation to early life factors, although the analyses were focused on gestational age and cortical thickness during adolescence [7]. Nassar et al., found, using our same approach of non-negative

matrix factorization analysis, that gestational age was related to 11 (of 26) structural networks in regions including orbitofrontal, parietal, temporal as well as hippocampus, amygdala and caudate, among others. As the low birth weight and short birth length are consequences of poor fetal nutrition and other stressors during pregnancy, the mechanisms that could account for the association of this with structural gray matter networks are diverse. Ischemia, inflammation, excitotoxicity due to poor nutrition (producing intrauterine growth restriction) or stress during gestation may affect neuron production, differentiation as well as consolidation of brain gray matter regions and networks, and its consequences may reach even later ages [7,41].

Regarding the academic implications of these results, structural brain networks associated with early life factors were not associated with academic performance in children with overweight or obesity. Only a network corresponding to the posterior cingulate gyrus and anterior precuneus (network 12) was related to reading, which is in line previous studies [42,43], but this association disappeared after FDR correction. Thus, the academic, clinical, and public health implications of the present results remain unknown but, based on previous studies, they might be related to emotion regulation, sensory motor support, social support or decision making [2,5], behaviors that were not assessed in this study and should be confirmed in future work.

Limitations

The present study has both strengths and limitations. An intrinsic limitation is that causal relations cannot be determined with this type of design. Also, breastfeeding practices were retrospectively recorded, and misunderstanding of the meaning of "exclusiveness" of breastfeeding, as well as the lack of information about complementary feeding may explain, in part, the null association related to breastfeeding practices [2,44]. Additionally, although we included

potentially confounding variables such as PHV and gestational age in our statistical models, other potentially key variables of fetal environment such as nutrition indicators, maternal stress or smoke exposure which may impact brain outcomes were not available [45,46]. A specific strength of our work is the use of the integrative approach to study gray matter volume to demonstrate alterations in the gray matter covariance patterns in this special population of children with overweight/obesity, with demonstrated alterations in gray matter structures.

Conclusion

In summary, these results suggest that birth weight and birth length, but not breastfeeding practices, are significantly associated with structural brain networks in children with overweight or obesity. The structural networks associated to birth weight or birth length were not linked to academic performance. Altogether, our findings suggest that early life factors are related to several structural brain networks assessed later during childhood, contributing to normal brain development.

Data availability

The datasets generated during and/or analyzed during the current study are not publicly available due to the participant consent form but are available from the corresponding author upon reasonable request.

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Author contributions

P.S.-U. was involved in the conception and design of the paper, analysis, interpretation of data, and drafting the manuscript. I.E.-C and F.B.O. were involved in the conception and design of the paper/project, interpretation of data, investigation (experiments), and critical revision. M.R.-A., J.V.-R., K.I.E., A.V.-G. and A.C. were involved in critical revision. All authors have approved the final version of the manuscript.

Competing interests

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Patient consent

The study was approved by the Committee for Research Involving Human Subjects at the University of Granada (Reference: 848, February 2014). All parents were informed about the study objective and written informed consent following the Declaration of Helsinki.

Figure legends

Figure 1. Structural brain network associated to birth weight and birth length after FDR correction. **Network 1:** Cerebellum I-IV, VIIIa, VIIb, crus II, and vermis VIIIb and IX; **Network 3:** Cerebellum V, VI, crus I, and vermis VI and VIIIa: **Network 4:** Frontal medial cortex, paracingulate and anterior cingulate gyrus; **Network 7:** Occipital pole, supracalcarine cortex, intracalcarine cortex and lingual gyrus; **Network 9:** Caudate, putamen, pallidum, accumbens and amygdala; **Network 12:** Posterior cingulate gyrus and anterior precuneus cortex; **Network 19:** Hippocampus and parahippocampal gyrus. Values into grid are standardized regression coefficients (β).

Table 1. Characteristics of study sample.

		All		*Boys*		*Girls*	*p-value*
N	n		n		n		
Physical characteristics	96		60		36		
Age (yr)		10.01 ± 1.14		10.16 ± 1.14		9.78 ± 1.13	0.114
Weight (kg)		55.65 ± 11.15		56.66 ± 10.69		53.97 ± 11.85	0.255
Height (cm)		143.81 ± 8.32		144.69 ± 7.37		142.34 ± 9.64	0.182
Peak height velocity offset (yr)		-2.33 ± 0.96		-2.65 ± 0.78		-1.80 ± 1.01	<0.001
Cardiorespiratory fitness (mL/kg/min)*		40.86 ± 2.77		40.84 ± 2.77		40.90 ± 2.82	0.927
Body mass index (kg/m^2)		26.70 ± 3.69		26.90 ± 3.79		26.36 ± 3.53	0.496
Body mass index category (n,%)	96		60		36		0.720
Overweight		25 (26.0)		16 (26.7)		9 (25.0)	
Obesity type I		41 (42.7)		27 (45.0)		14 (38.9)	
Obesity type II		30 (31.2)		17 (28.3)		13 (36.1)	
Parental education university level (n,%)	96		60		36		0.316
None of the parents		64 (66.7)		43 (71.7)		21 (58.3)	
One of the two parents		17 (17.7)		10 (16.7)		7 (19.4)	
Both parents		15 (15.6)		7 (11.7)		8 (22.2)	
Neonatal characteristics							
Birth weight (g)	94	3343.72 ± 542.25	59	3358.98 ± 579.32	35	3318.00 ± 480.28	0.725
Birth length (cm)	85	50.69 ± 2.68	57	50.61 ± 3.02	28	50.86 ± 1.84	0.686
Gestational age (week)	96	38.62 ± 2.59	60	38.57 ± 2.59	36	38.70 ± 2.63	0.812
Breastfeeding practices (months)	92						
Exclusive breastfeeding† [0-16]		3.19 ± 3.24	59	3.53 ± 3.53	33	2.58 ± 2.59	0.175
Any breastfeeding‡ [0-43]		7.06 ± 8.03	59	6.81 ± 7.18	33	7.50 ± 9.44	0.690
Academic performance (standard score)**	96		60		36		
Mathematics		102.02 ± 10.68		102.37 ± 11.23		101.44 ± 9.82	0.684
Reading		108.55 ± 12.92		108.33 ± 11.08		108.92 ± 15.67	0.832
Writing		113.99 ± 11.99		112.55 ± 11.91		116.39 ± 11.91	0.130
Total Achievement		109.49 ± 11.66		108.98 ± 10.67		110.33 ± 13.25	0.585

Values are mean ± SD or percentage. *Measured by the 20-m shuttle run test; †Months the child received only breast milk. ‡Months the child received breast milk combined with other liquid, or solid. **Measured by the Battery III Woodcock-Muñoz Tests of Achievement.

Supplemental table S1. Association of birth weight and birth length with structural network that survived correction

Network	Birth weight		Birth length	
	β	p	*β*	p
Network 1: Cerebellum I-IV, VIIIa, VIIb, crus II, and vermis VIIIb and IX	0.309	0.001	0.359	<0.001
Network 3: Cerebellum V, VI, crus I, and vermis VI and VIIIa	0.260	0.009	0.314	0.003
Network 4: Frontal medial cortex, paracingulate and anterior cingulate gyrus	0.245	0.010	-	-
Network 7: Occipital pole, supracalcarine cortex, intracalcarine cortex and lingual gyrus	-	-	0.288	0.009
Network 9: Caudate, putamen, pallidum, accumbens and amygdala	0.252	0.008	0.304	0.004
Network 12: Posterior cingulate gyrus and anterior precuneus cortex	0.240	0.014	0.266	0.013
Network 19: Hippocampus and parahippocampal gyrus	0.252	0.011	0.327	0.002

Analyses were adjusted for sex, peak height velocity offset (years), parent education university level (neither/one/both), gestational age (weeks) and cardiorespiratory fitness (mL/kg/min). Network are depicted grapically in Figure 1.

Supplemental table S2. Association of birth weight and birth length with structural network

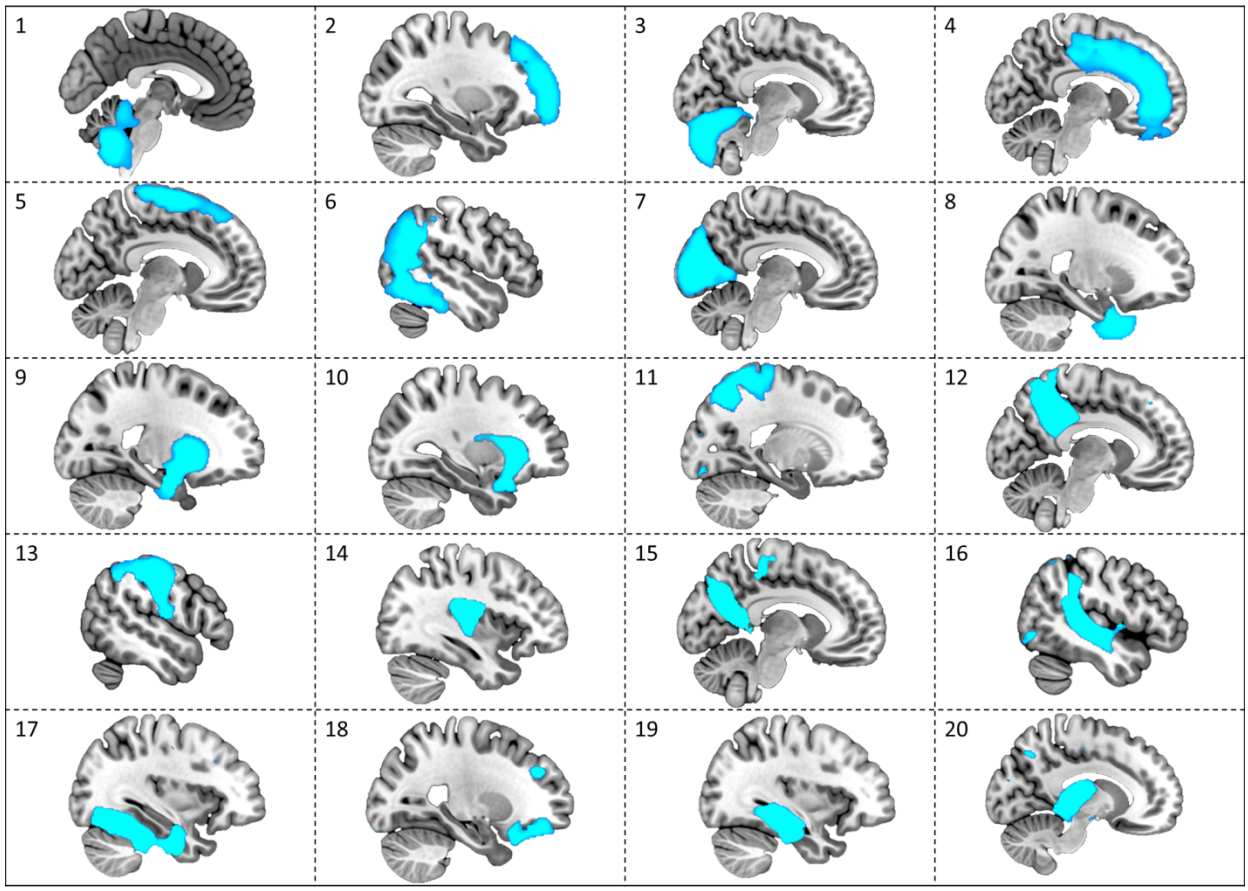
Network	Birth weight		Birth length	
	β	*p*	*β*	*p*
Network 1	**0,309**	**0,001**	**0,359**	**< 0,001**
Network 2	0,158	0,103	0,117	0,281
Network 3	**0,260**	**0,009**	**0,314**	**0,003**
Network 4	**0,245**	**0,010**	**0,237**	**0,023**
Network 5	0,094	0,318	0,071	0,499
Network 6	**0,207**	**0,033**	**0,225**	**0,035**
Network 7	**0,212**	**0,037**	**0,288**	**0,009**
Network 8	**0,194**	**0,043**	0,145	0,169
Network 9	**0,252**	**0,008**	**0,304**	**0,004**
Network 10	**0,199**	**0,031**	0,196	0,058
Network 11	0,150	0,149	0,133	0,240
Network 12	**0,240**	**0,014**	**0,266**	**0,013**
Network 13	0,127	0,229	0,131	0,256
Network 14	0,126	0,190	0,149	0,157
Network 15	**0,198**	**0,040**	**0,237**	**0,025**
Network 16	**0,212**	**0,032**	**0,242**	**0,021**
Network 17	0,095	0,307	0,187	0,069
Network 18	0,155	0,096	0,109	0,293
Network 19	**0,252**	**0,011**	**0,327**	**0,002**
Network 20	0,189	0,062	0,174	0,118

Analyses were adjusted for sex, peak height velocity offset (years), parent education university level (neither/one/both), gestational age (weeks) and cardiorespiratory fitness (mL/kg/min). Statistically significant values are shown in bold. Networks are depicted in Figure 1.

Supplemental table S3. Association of any and exclusive breastfeeding with structural network

Network	Any breastfeeding‡		Exclusive breastfeeding†	
	β	*p*	*β*	*p*
Network 1	0,154	0,087	0,161	0,076
Network 2	0,106	0,262	0,066	0,489
Network 3	0,113	0,239	0,153	0,114
Network 4	0,074	0,426	0,030	0,751
Network 5	0,105	0,251	0,070	0,451
Network 6	0,124	0,189	0,111	0,246
Network 7	0,165	0,092	**0,197**	**0,046**
Network 8	0,120	0,199	0,011	0,903
Network 9	0,066	0,483	-0,017	0,855
Network 10	0,080	0,376	0,018	0,848
Network 11	0,110	0,275	0,041	0,688
Network 12	0,095	0,323	0,041	0,673
Network 13	0,166	0,100	0,043	0,672
Network 14	0,120	0,191	0,044	0,640
Network 15	0,121	0,195	0,076	0,425
Network 16	0,094	0,329	0,131	0,178
Network 17	0,155	0,084	0,095	0,296
Network 18	0,167	0,062	0,044	0,630
Network 19	0,092	0,339	0,051	0,602
Network 20	0,092	0,353	0,031	0,758

Analyses were adjusted for sex, gestational age, peak height velocity offset (years), parent education university level (neither/one/both) and cardiorespiratory fitness (mL/kg/min). †Months the child received only breast milk. ‡Months the child received breast milk combined with other. Statistically significant values are shown in bold. liquid, or solid. Network are depicted grapically in Figure 1.



Supplemental figure S1. Structural covariance networks delineated by Non-negative Matrix Factorization analysis. Structural covariance networks are shown for the 20-network solution. The blue color represents the spatial distribution of each network. For each network, we show the sagittal view (left hemisphere) that best captures the main areas of coverage. The anatomical coverage of each structural covariance network was a follows:(1) cerebellum I-IV, VIIIa, VIIb , crus II, vermis VIIIb to vermis IX; (2) frontal pole; (3) cerebellum V, VI, crus I, vermis VI to vermis VIIIa; (4) frontal medial cortex, paracingulate gyrus to anterior cingulate gyrus; (5) superior frontal gyrus, supplementary motor cortex to precentral gyrus; (6) lateral occipital cortex, angular gyrus to temporooccipital parts of middle and inferior temporal gyri; (7) occipital pole, supracalcarine cortex, intracalcarine cortex to lingual gyrus; (8) temporal pole to temporal fusiform cortex; (9) caudate, putamen ,pallidum, accumbens to amygdala; (10) frontal operculum cortex to insular cortex; (11) superior poscentral gyrus to superior lateral occipital cortex; (12) posterior cingulate gyrus to anterior precuneous cortex; (13) inferior poscentral gyrus to central opercular cortex; (14) anterior supramarginal gyrus to parietal operculum cortex; (15) posterior precuneus cortex to cuneal cortex; (16) posterior supramarginal gyrus to posterior superior temporal gyrus; (17) occipital fusiform gyrus to temporal occipital fusiform cortex; (18) frontal orbital cortex; (19) hippocampus to parahippocampal gyrus; and (20) thalamus.