Title: Aberrant gyrification contributes to the link between gestational age and adult IQ after premature birth

Short title: Aberrant gyrification in prematurity

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Abbreviations

AMC:	Absolute mean curvature
ANOVA:	Analysis of variance
BLS:	Bavarian Longitudinal Study
BW:	Birth weight
CI:	Confidence interval
DARTEL:	Diffeomorphic Anatomical Registration Through Exponentiated Lie Algebra
FA:	Fractional anisotropy
FS-IQ:	Full-scale IQ
FT:	Full-term
FWE:	Family-wise error
FWHM:	Full-width at half maximum
GA:	Gestational age
INTI:	Intensity of neonatal treatment index
IP:	Intermediate progenitor
lGI:	Local gyrification index
MPRAGE:	Magnetization Prepared Rapid Acquisition Gradient Echo
MRI:	Magnetic resonance imaging
PMG:	Polymicrogyria
PoCC:	Postcentral Gyrus
RGC:	Radial glia cell
ROI:	Region of interest
SE:	Standard error
SES:	Socioeconomic status
LTC:	Lateral temporal cortex
TE:	Time to echo
TFCE:	Threshold-free cluster enhancement
TI:	Time to inversion
TR:	Time to repetition
VLBW:	Very low birth weight
VP:	Very preterm
VP/VLBW:	Very preterm/very low birth-weight
WAIS:	Wechsler Adults Intelligence Scale

Abstract

Gyrification is a hallmark of human brain development, starting in the second half of gestation in primary cortices, followed by unimodal and then transmodal associative cortices. Alterations in gyrification have been noted in premature-born newborns and children, suggesting abnormal cortical folding to be a permanent feature of prematurity. Furthermore, both gyrification and prematurity are tightly linked with cognitive performance, indicating a link between prematurity, gyrification, and cognitive performance. To investigate this triangular relation, we tested the following two hypotheses: firstly, gyrification is aberrant in premature-born adults, and secondly, aberrant gyrification contributes to the impact of prematurity on adult cognitive performance.

One hundred and one very premature-born adults (i.e., adults born before 32 weeks of gestation, and/or with birth weight below 1500g) and 111 mature-born adults were assessed by structural MRI and cognitive testing at 26 years of age. Gyrification was measured by local cortical absolute mean curvature (AMC), evaluated through structural MRI. Cognitive performance was assessed by the Wechsler-Adult-Intelligence-Scale, full-scale IQ test. Two-sample t-tests, regression and mediation analyses were used to assess AMC group differences and the relation between AMC, birth-related variables, and full-scale IQ.

Three key findings were identified. 1) Local AMC was widely increased in fronto-temporo-parietal primary and associative cortices of very premature-born adults. Increase of AMC was inversely associated with gestational age and birth weight and positively associated with medical complications at birth, respectively.

2) Increased AMC of temporal associative cortices specifically contributed to the association between prematurity and reduced adult IQ (two-path mediation), indicating that aberrant

gyrification of temporal associative cortices is critical for impaired cognitive performance after premature birth.

3) Further investigation of the relationship of gyrification between the early folding postcentral cortices and associative temporal cortices, folding later during neurodevelopment, revealed that the effect of gyrification abnormalities in associative temporal cortices on adult IQ is influenced itself by gyrification abnormalities occurring in the early folding postcentral cortices (three-path mediation). These results indicate that gyrification development across cortical areas in the brain conveys prematurity effects on adult IQ.

Overall, these results provide evidence that premature birth leads to permanently aberrant gyrification patterns suggesting an altered neurodevelopmental trajectory. Statistical mediation modelling suggests that both aberrant gyrification itself as well as its propagation across the cortex express aspects of impaired neurodevelopment after premature birth and lead to reduced cognitive performance in adulthood. Thus, markers of gyrification appear as potential candidates for prognosis and treatment of prematurity effects.

Introduction

Premature birth, i.e. birth before 37 weeks of gestation and/or birth weight below 2500g, has a worldwide prevalence of more than 10% (Volpe, 2009; Blencowe *et al.*, 2012). It is associated with an increased risk (i.e., 30-60% of premature-born individuals) for birth complications and adverse long-term outcomes including brain abnormalities and impaired cognitive functions (Volpe, 2009). Particularly, very premature individuals, who are born very preterm (VP; gestational age < 32 weeks) and/or have a very low birth weight (VLBW; < 1500 g), have a substantially increased risk (more than 50%) for long-term neurocognitive impairments, psychiatric disorders, and lower socio-economic status (Saigal and Doyle, 2008; Nosarti *et al.*, 2012; D'Onofrio *et al.*, 2013). The increased risk for cognitive impairments is thought to be caused by perinatal brain injury due to hypoxia, ischemia, haemorrhage, infections, and inflammatory processes as well as neonatal pain and stress and subsequent alterations in brain development (Volpe, 2009).

At the microscopic level, these processes primarily impair the development of pre-myelinating oligodendrocytes, GABA-ergic interneurons, and subplate neurons, which play a fundamental role in the development of cortical microstructure, morphology, and connectivity (Back *et al.*, 2002; Deng, 2010; Buser *et al.*, 2012; Kinney *et al.*, 2012; Ball *et al.*, 2013; Dean *et al.*, 2013; Salmaso *et al.*, 2014). For example, subplate neurons, which control the formation of cortical microcircuits mainly during gestational weeks 18 to 35, are highly vulnerable to transient hypoxia resulting in lasting subplate neuron dysfunction and consequently abnormal microcircuit formation (Kanold and Luhmann, 2010; Kostovic *et al.*, 2014; McClendon *et al.*, 2017). Corresponding with changes at the microscopic level, macroscopic alterations in both white matter and grey matter have been described after premature birth. Whereas impairments of white matter integrity where found to be widespread across the whole forebrain (Skranes *et al.*, 2007; Eikenes *et al.*, 2011; Ball *et al.*, 2012, 2014; Meng *et al.*, 2016), grey matter abnormalities, such as volume reduction, were found mainly

in medial and temporal lobes as well as subcortical structures, such as the thalamus, striatum, and basal forebrain (Pierson *et al.*, 2007; Nosarti *et al.*, 2008; Meng *et al.*, 2016; Grothe *et al.*, 2017; Karolis *et al.*, 2017). These structural brain changes are tightly associated with cognitive impairments (Northam *et al.*, 2011; Farajdokht *et al.*, 2017). For example, aberrant thalamo-cortical connectivity in premature newborns is associated with cognitive performance at two years of age (Ball *et al.*, 2015); impaired cognitive functioning was associated with grey and white matter abnormalities in the inferior frontal gyrus, bilateral temporal lobes and corpus callosum in preterm born subjects in young adulthood (Nosarti *et al.*, 2014). These neurocognitive changes suggest that basic processes of brain development are critically affected by premature birth leading to cognitive impairments and altered brain structure in adulthood.

One such key developmental process influencing for adult brain morphology and function is cortex gyrification or cortical folding (Zilles *et al.*, 2013). Gyrification refers to the process of cortical folding (Zilles *et al.*, 2013), which is a main characteristic of brain development not only in humans, but also in other mammalian species of so-called gyrencephalic brains (Zilles *et al.*, 2013). In humans, gyrification is considered a neurodevelopmental milestone of the last trimester of gestation and the early postnatal period, leading to an increased cortical surface of the human brain relative to a limited skull capacity (Zilles *et al.*, 2013; Sun and Hevner, 2014). While recent studies provide evidence for a consistent developmental trajectory across brain regions starting in primary cortices followed by unimodal and then transmodal associative cortices (Hill *et al.*, 2010; Garcia *et al.*, 2018), the exact underlying mechanisms of gyrification are still unclear. Distinct hypotheses either emphasise the impact of asymmetric grey matter growth within the cortical plate (Armstrong *et al.*, 1991; Kriegstein *et al.*, 2006; Ronan *et al.*, 2014) or the effect of axonal tension by white matter fiber tracts connecting the cortex with distant cortical or subcortical areas (Van Essen, 1997; Mota and Herculano-Houzel, 2015). To quantify brain gyrification, several measures have been

developed over the last two decades (Luders *et al.*, 2006; Schaer *et al.*, 2008; Zilles *et al.*, 2013). The current gold-standard measure, gyrification index, was originally described on two-dimensional coronal brain slices, reflecting the ratio of the complete and outer (superficially exposed) contours (Zilles *et al.*, 1988). In the last decade, tools to measure gyrification in-vivo at each vertex of a three-dimensional brain image were developed, either by a surface-based approach, called local gyrification index (Schaer *et al.*, 2008) or by computation of absolute mean curvature (AMC) as an estimation of cortical convolution (Luders *et al.*, 2006). In the latter method, which is used in this paper, change in normal direction along the cortical surface is measured and expressed as mean curvature. Large negative values correspond to sulci and large positive values correspond to gyri. Absolute values of mean curvature express the local amount of gyrification and can be considered a measure of the sharpness of gyri and sulci (Luders *et al.*, 2006). A very strong positive correlation has been shown between AMC-based gyrification and total surface area (for an explanatory figure, please see (Luders *et al.*, 2006)). Based on these measures, both high regional and inter-subject variability have been demonstrated for gyrification (Zilles *et al.*, 2013), with cortical folding having highest levels in 'associative' parieto-occipito-temporal and prefrontal cortices (Zilles *et al.*, 1988) and consistent differences between males and females (Luders *et al.*, 2008). This likely reflects brain morphology as a consequence of different genetic, developmental, environmental, and functional influences (Zilles *et al.*, 2013).

Prior studies have demonstrated that prematurity affects gyrification, showing aberrant cortical folding in premature-born newborns (Kersbergen *et al.*, 2016; Lefèvre *et al.*, 2016) and young children (Zhang *et al.*, 2015). Although gyrification is a highly dynamic process that can be altered across the lifespan (Hogstrom *et al.*, 2013) or during shorter periods of environmental stress such as periods of malnutrition (Bernardoni *et al.*, 2018), it is unknown whether aberrant gyrification in premature-born infants results in permanent alterations.

Regional variance in gyrification may explain cognitive impairments associated with premature birth. It has been shown that variance in local gyrification of fronto-parietal cortices accounts for more than 10% of variance in general intelligence scores in normal healthy young persons (Gregory *et al.*, 2016). In line with this association, aberrant gyrification was described in several pathological neurodevelopmental conditions. For example, a decrease in gyrification was found in attention deficit hyperactivity disorder (Wolosin *et al.*, 2009) and dyslexia (Casanova *et al.*, 2004), while increased gyrification was described in Williams syndrome (Gaser *et al.*, 2006), autism (Jou *et al.*, 2010), and schizophrenia (Palaniyappan and Liddle, 2012; Schultz *et al.*, 2013). However, it is unknown whether abnormal gyrification is relevant for reduced long-term outcomes of prematurity.

Thus, gyrification is a hallmark of brain development during the second half of gestation and shows a sequential development starting in unimodal cortices and then extending to temporal associative cortices. It interferes with prematurity regarding both, its developmental starting period and its association with impaired general cognitive capacities. We hypothesised that premature birth leads to long-lasting cortical folding anomalies. Furthermore, these aberrations may express the effect of altered neurodevelopment after prematurity on adult cognitive performance. We tested our hypotheses through calculation of both curvature-based estimations of gyrification derived from structural MRI and cognitive ability as evaluated by intelligence assessments. The two parameters were subsequently investigated using canonical statistical testing and mediation analyses in a large cohort of very premature-born young adults (VP/VLBW) and age-matched controls born at full-term (FT).

Material and methods

Participants

The participants examined in this study are part of the Bavarian Longitudinal Study (BLS), a geographically defined, whole-population sample of neonatal at-risk children and healthy full term controls who were followed from birth into adulthood (Riegel *et al.*, 1995; Wolke and Meyer, 1999). Of the initial 682 infants born very preterm (<32 weeks) and/or with very low birth weight (< 1500g), 411 were eligible for the 26-year follow-up assessment, and 260 (63.3%) participated in psychological assessments (Breeman *et al.*, 2015). Of the initial 916 full term born infants from the same obstetric hospitals that were alive at 6 years, 350 were randomly selected as control subjects within the stratification variables of sex and family socioeconomic status in order to be comparable with the VP/VLBW sample. Of these, 308 were eligible for the 26-year follow-up assessment, and 229 (74.4%) participated in psychological assessments. All of the 260 subjects from the VP/VLBW group underwent an initial screening for MR-related exclusion criteria, which included: (self-reported) claustrophobia, inability to lie still for > 30 minutes, unstable medical conditions (e.g. severe asthma), epilepsy, tinnitus, pregnancy, non-removable, MRI incompatible metal implants and a history of severe CNS trauma or disease that would impair further analysis of the data. Also, patients that could not participate at the functional MRI part of the examination due to restricted motor function of the hands, impaired vision, or incapacity to understand the functional MRI paradigm. The most frequent reason not to perform the MRI exam, however, was a lack of motivation. The remaining eligible, 101 VP/VLBW and 111 FT individuals underwent MRI at 26 years of age. The distribution of gestational age and birth weight in the VP/VLBW group is depicted in the supplemental information (Figure S1).

The MRI examinations took place at two sites: The Department of Neuroradiology, Klinikum rechts der Isar, Technische Universität München, (n=145) and the Department of Radiology, University Hospital of Bonn (n=67). The study was carried out in accordance with the

Declaration of Helsinki and was approved by the local institutional review boards. Written consent was obtained from all participants. All study participants received travel expenses and a small payment for attendance. A more detailed description of participants, including incidental brain MRI findings can be found in previous publications (Bauml *et al.*, 2015; Grothe *et al.*, 2017) and the supplementary material.

Birth-related variables:

Gestational age (GA) was estimated from maternal reports on the first day of the last menstrual period and serial ultrasounds during pregnancy. In cases where the two measures differed by more than two weeks, clinical assessment at birth with the Dubowitz method was applied (Dubowitz *et al.*, 1970). Maternal age, birth weight (BW), duration of hospitalization, and Intensity of Neonatal Treatment Index (INTI), which reflects the duration and intensity of medical treatment after birth, were obtained from obstetric records (Riegel *et al.*, 1995; Gutbrod *et al.*, 2000). Family socioeconomic status (SES) was assessed through structured parental interviews within 10 days of childbirth. SES was computed as a weighted composite score based on the profession of the self-identified head of each family together with the highest educational qualification held by either parent (Bauer, 1988). Daily assessments of care level, respiratory support, feeding dependency and neurological status (mobility, muscle tone, and neurological excitability) were performed. Each of the six variables was scored on a 4-point rating scale (0-3). The INTI was computed as the mean score of daily ratings during the first 10 days of life or until a stable clinical state was reached, depending on which occurred first, ranging from 0 (best state) to 18 (worst state).

Cognitive assessment in adulthood

At 26 years of age, study participants were assessed using the short version of the German Wechsler Adults Intelligence Scale, Third edition (WAIS-III) (von Aster *et al.*, 2006). The

assessment took place prior to and independent of the MRI scan and was carried out by trained psychologists who were blinded to group membership. Consecutively, full-scale intelligence quotient (FS-IQ) performance was computed.

MRI data acquisition

MRI examinations were performed at both sites on either a Philips Achieva 3T or a Philips Ingenia 3T system using an 8-channel SENSE head-coils. Subject distribution among scanner was as follows: Bonn Achieva 3T: 5 VP/VLBW, 12 FT, Bonn Ingenia 3 T: 33 VP/VLBW, 17 FT, Munich Achieva 3T: 60 VP/VLBW, 65 FT, Munich Ingenia 3T: 3 VP/VLBW, 17 FT. To account for possible confounds by the scanner-specific differences, all statistical analyses included categorical dummy regressors for scanner identity as covariates of no interest. Sequence parameters were kept identical across all scanners. Scanners were checked regularly to provide optimal scanning conditions. MRI physicists at the University Hospital Bonn and Klinikum rechts der Isar regularly scanned imaging phantoms, to ensure within-scanner signal stability over time. Signal-to-noise ratio (SNR) was not significantly different between scanners (one-way ANOVA with factor "scanner-ID" [Bonn 1, Bonn 2, Munich 1, Munich 2]; $F(3,182) = 1.84$, $p = 0.11$). The image protocol included a high-resolution T1-weighted, 3D-MPRAGE sequence (TI=1300 ms, TR = 7.7 ms, TE = 3.9 ms, flip angle 15°; field of view: 256 mm x 256 mm) with a reconstructed isotropic voxel size of 1 mm^3. All images were visually inspected for artifacts and passed homogeneity control implemented in the CAT12 toolbox (Gaser and Dahnke, 2016).

Surface-based morphometry analysis

First, all images saved as DICOMs were transformed to Nifti-format using dcm2nii (Li *et al.*, 2016). The CAT12 toolbox comprises a processing pipeline for surface-based morphometry, which includes an established novel algorithm for extracting the cortical surface (Dahnke *et*

al., 2013), which then allows for the computation of multiple morphometric parameters, including gyrification based on the absolute mean curvature (AMC) approach (Luders *et al.*, 2006).

In brief, T1-weighted images underwent tissue segmentation into grey matter, white matter and cerebrospinal fluid. Topological correction was performed through an approach based on spherical harmonics (Yotter *et al.*, 2011). An adapted volume-based diffeomorphic DARTEL algorithm was then applied to the surface for spherical registration. Local curvature-based gyrification index, AMC, was extracted based on absolute mean curvature (Luders *et al.*, 2006). Central cortical surfaces were created for both hemispheres separately. Finally, all scans were re-sampled and smoothed with a Gaussian kernel of 20 mm (FWHM).

Statistical analysis

To determine differences in AMC between groups, a two-sample t-test was performed using the batch-mode implemented in SPM12, adjusting for sex and scanner as covariates of no interest. Contrasts were processed using threshold-free cluster enhancement (TFCE) (Smith and Nichols, 2009) and statistical significance was defined as $p<0.05$, family-wise error (FWE) corrected. The pattern of between group difference of AMC (VP/VLBW > FT) was saved as binary cluster and introduced as explicit mask into multiple regression analyses. In three separate multiple regression analyses, either GA, BW, or INTI served as regressors of interest, and residuals of the other two covariates were introduced as regressors of no interest, in addition to sex and scanner.

Linking prematurity, AMC, and IQ; mediation analysis

To determine the association between gyrification and FS-IQ in premature-born adults, multiple regression analysis was performed with AMC as dependent variable, FS-IQ as

regressor of interest, and sex and scanner as regressors of no interest in the VP/VLBW group only. All results were obtained using TFCE at $p<0.05$, FWE corrected.

In order to test whether AMC mediates the influence of premature birth on FS-IQ, a mediation analysis was performed using the PROCESS toolbox (version 3.0) (Hayes, 2017). Mean AMC values were extracted subject-wise from between-group differences in AMC, as depicted in Figure 1. Firstly, the influence of prematurity as a group effect on FS-IQ was investigated using multiple regression analysis (dependent variable: FS-IQ, covariate of interest: premature birth, covariate of no interest: sex). Secondly, to investigate whether differences in gyrification have a mediating effect on the association between prematurity and FS-IQ, mediation analysis was performed. In the mediation model, prematurity at birth was entered as causal variable, FS-IQ as the outcome variable, mean AMC within the group difference cluster as the mediator variable; and MRI scanner and sex as covariates of no interest. Path coefficients were estimated using (unstandardised) regression coefficients from multiple regression analyses, and statistical significance of the indirect effect was tested using a nonparametric bootstrap approach (with 5000 repetitions) to obtain 95% confidence intervals. Thirdly, in order to get more specific spatial information about the mediation effect of gyrification of prematurity on IQ, we used distinct parts of the widespread group difference cluster of AMC and tested them separately for mediation effects. Particularly, mean AMC per subject was extracted in a ROI-based manner using the Desikan-Killiany atlas (Desikan *et al.*, 2006) from both the postcentral (PoCC, mean AMC of postcentral gyrus ROI) and the lateral temporal cortices (LTC, mean AMC of middle and superior temporal gyrus ROIs) bilaterally. These primary and associative cortices have been used for two reasons: 1) the gyrification of primary and associative cortices are distinctively relevant for IQ, with associative cortex gyrification being more tightly linked with IQ in healthy young adults (Gregory *et al.*, 2016); 2) primary cortex and associative cortex gyrification start in a sequential manner, with earlier gyrification of primary cortices (Hill *et al.*, 2010; Garcia *et al.*, 2018), suggesting that

prematurity has distinct effects on primary and associative cortex gyrification. The ROI-based averaging approach based on a given brain atlas was chosen due to technical limitations for vertex-wise mediation analysis. Detailed schemes of mediation analyses are shown in Figure 3. Path coefficients are calculated as previously described.

Finally, we tested whether gyrification development across the brain – primary cortices first, associative cortices later – is relevant for the impact of prematurity on IQ. Only LTC gyrification was a significant mediation variable between prematurity and IQ in a parallel mediation analysis of LTC and PoCC gyrification. No significant mediation was seen between PoCC, gyrification, and IQ in this model. However, primary cortex gyrification precedes folding of associative cortices and thus may be more affected by prematurity and associated hypoxic/hypoxemic adverse events. We therefore predicted that primary cortex gyrification acts as a mediator of cortical folding of association cortices. A subsequent three-path 'sequential' mediation analysis was performed, testing whether aberrant cortical folding in the PoCC has an indirect effect on FS-IQ in VP/VLBW subjects via aberrant cortical folding in the LTC and whether this mediates the impact of prematurity at birth (or its different aspects GA, BW, INTI, mediation analysis in the VP/VLBW group only) on adult FS-IQ. For the detailed path-model see figure 4 (path coefficients are calculated as described before).

Data Availability Statement

Patient data used in this study are not publicly available but stored by the principal investigators of the Bavarian Longitudinal Study.

Results

Sample characteristics

Group demographic and clinical background variables are shown in Table 1. There were no significant differences between the VP/VLBW and FT group regarding age at scanning ($p=0.765$), sex ($p=0.167$), SES at birth ($p=0.492$), and maternal age ($p=0.889$). By design, VP/VLBW subjects had significantly lower GA ($p<0.001$) and lower BW ($p<0.001$). They were hospitalised for a longer time after birth ($p<0.001$). VP/VLBW subjects had significantly lower FS-IQ scores ($p<0.001$).

Widespread increases of AMC in premature-born adults, being associated with birth-related variables

In order to test for group differences in gyrification between VP/VLBW and FT subjects, we conducted a vertex-wise two-sample t-test for AMC, correcting for sex and scanner (Figure 1, Table 2). We found significantly increased AMC in both hemispheres with predominance in the right hemisphere, namely in bilateral lateral temporal, right lateral frontal and right parietal cortices as well as in bilateral pre- and postcentral cortices. We did not find significantly decreased AMC clusters in the VP/VLBW group.

In order to test whether AMC increases in premature-born adults were indeed linked with premature birth, we performed vertex-wise multiple regression analyses of the relationship between AMC and variables related to premature birth, namely GA, BW, and INTI, in the VP/VLBW group only, restricted to the AMC group difference cluster (Figure 2; Tables S2.1-3). Due to the high collinearity between GA, BW, and INTI as determined by Pearson correlation (GA and BW: $r = 0.316$ ($p = 0.001$); GA and INTI: $r = -0.483$ ($p < 0.001$); BW and INTI: $r = -0.269$ ($p=0.007$)), we chose one parameter as covariate of interest, respectively, and residualised the other two parameters before including them as covariates of no interest. For GA, we found a negative association with AMC, predominantly in the right lateral

temporal lobe and the bilateral lateral frontal lobe, as well as in the central region, the right temporoparietal junction, and left medial temporal lobe (Figure 2A). For BW, we found a negative association with AMC in lateral temporal and frontal cortices, the left temporoparietal junction, and left medial temporal lobe (Figure 2B). For INTI, we found a positive association with AMC in bilateral frontal cortices (Figure 2C). These results suggest that AMC increases in premature-born adults are indeed linked to the degree of prematurity mostly in frontoparietal, pre- and postcentral cortices, and that different aspects of prematurity affect regionally distinct cortices.

Increased AMC of temporal associative cortices contributes to the association between prematurity and IQ

Cognitive performance has been linked to gyrification (Gregory *et al.*, 2016) and premature birth is associated with reduced cognitive performance in adult life (Saigal and Doyle, 2008). To test whether aberrant gyrification in premature-born adults, measured as increased AMC, is associated with aberrant cognitive impairment, defined by reduced FS-IQ, we conducted a multiple regression analyses in the VP/VLBW group only with AMC as dependent variable and FS-IQ as independent variable. Deviations due to sex and scanner were controlled for. We observed a significant negative association between AMC and FS-IQ in bilateral lateral and anterior temporal cortices as well as in the occipitotemporal junction (Figure 3, Table S3). The negative association between FS-IQ and AMC was most evident for anterolateral temporal cortical areas, which are also significantly negatively correlated with GA. No significant positive association was seen between AMC and FS-IQ.

We further tested whether aberrant gyrification, i.e. increased AMC, mediates the association between prematurity and IQ. Prematurity was significantly associated with lower FS-IQ (regression coefficient: 8.347, SE: 1.714, standardised coefficient beta: 0.322; 95%-CI: 4.97-11.73 $p<0.001$). In a first mediation analysis using averaged mean AMC across the group

difference cluster as mediator, the direct effect of premature birth as a group factor on adult FS-IQ was not significant (c_1'=3.75 ± 2.12; p=0.079). However, the bootstrapped 95%-CI determined that the indirect effect mediated by mean AMC across groups was significantly different from zero (b_1=5.09±1.39; 95%-CI: 2.55-7.96), indicating the mediating effect of mean AMC on the association between prematurity and IQ (Figure 3B).

In order to get more detailed spatial information about the mediation effect of gyrification on FS-IQ we used distinct ROI-based clusters from the postcentral cortices, PoCC, and lateral temporal cortices, LTC, serving as surrogate for the primary and secondary folding cortices respectively as mediators. We included AMC of both PoCC and LTC as mediators in a parallel analysis to test for mediation effects on adult FS-IQ in premature-born adults (Figure 3C). In this mediation analysis, the direct effect of prematurity on FS-IQ remained significant (c_2=4.75 ± 1.98; p=0.018, 95%-CI: 0.84-8.67). The indirect mediation effect of the PoCC was not significant (a_2=1.51 ± 0.96; 95%-CI: -0.26-3.53) while mean AMC in the LTC did show a significant mediation effect between prematurity and FS-IQ (b_2=2.72 ± 1.56; 95%-CI: 0.52-5.09). In conclusion, we observed a statistical mediation effect for gyrification in temporal association cortices but not for primary postcentral cortices.

Gyrification development across the brain contributes to the association between prematurity and IQ

Finally, we asked whether gyrification development across the brain – starting with primary cortices, followed by associative cortices – is relevant for the impact of prematurity on IQ. We performed a three-path mediation analysis, which extends the previous two-path mediation model (Figure 3B and 3C). Specifically, we expected gyrification in primary postcentral cortices to propagate prematurity effects on the gyrification of temporal cortices. In more detail (Figure 4A), setting prematurity as dichotomous causal variable and adult FS-IQ as outcome variable with mean AMC in the bilateral PoCC being the first mediator and

mean AMC in bilateral LTC being the second mediator, the direct effect of prematurity on adult FS-IQ did remain significant (c_3=4.75 ± 1.96; p=0.018). The indirect effect mediated by mean AMC in the PoCC was not significant (a_3 =1.46±0.96; 95%-CI: -0.31-3.48) while the indirect effect mediated by mean AMC in the LTC was significant (b_3 =1.41±0.71; 95%-CI: 0.23-3.00) (Figure 4A). Critically, mean AMC in the PoCC had a significant indirect mediation effect via mediation of mean AMC in LTC on FS-IQ (ab_3 =1.22±0.56; 95%-CI: 0.23-2.39), indicating that prematurity effects on the mediation effect of lateral temporal cortices gyrification is translated by postcentral cortices gyrification.

In order to address in more detail which aspects of prematurity modulate postcentral cortices gyrification, we restricted the last mediation analysis to premature-born adults and performed again a 'sequential' three-path mediation analysis. In more detail, we investigated whether the effect of GA on FS-IQ is sequentially mediated by AMC in the PoCC and the LTC (Figure 4B). We used the same models also for BW and INTI, respectively, instead of GA (see below). Partial correlation analysis, controlled for sex and scanner, revealed a significant association of FS-IQ with GA (r=0.276; p=0.007), BW (r=0.306; p=0.003), and INTI (r=-0.220; p=0.032), respectively. The direct effect of GA on FS-IQ was not significant (c_4=0.79±0.63; p=0.212) in the mediation model. The indirect effect mediated by mean AMC in the PoCC was not significant (a_4=0.09±0.21; 95%-CI: -0.30-0.58) while the indirect effect mediated by mean AMC in the LTC was significant (b_4=0.62±0.27; 95%-CI: 0.15-1.20). Interestingly, mean AMC in the PoCC had a significant mediation effect via mediation of mean AMC in LTC on FS-IQ (ab_4=0.32±0.17; 95%-CI: 0.05-0.70). This result indicates that gestational age impacts IQ through mediatory effects of temporal associative cortices gyrification, which in turn are mediated by gyrification changes in primary postcentral cortices.

Analogous results were obtained when inserting BW or INTI as causal variables. For BW, the mediating effect of PoCC was not significant (a_5=0.0003±0.001; 95%-CI: -0.003-0.003) while

the mediating effects of LTC (b_5=0.0024±0.0014; 95%-CI: 0.0001-0.0055) and of PoCC via LTC (ab_5=0.0025±0.0011; 95%-CI: 0.0007-0.0049) were significant. For INTI, the mediating effect of PoCC was not significant (a_6=-0.048±0.105; 95%-CI: -0.291-0.133), while the mediating effects of LTC (b_6=-0.191±0.123; 95%-CI: -0.491- -0.004) and of PoCC via LTC (ab_6=-0.179±0.084; 95%-CI: -0.363- -0.038) were significant. Both results indicate that effects of birth weight and medical complications at birth on the mediation effect of temporal associative cortex gyrification are itself influenced by changes in primary postcentral cortex gyrification. When considering the final results and the fact that primary cortices are distinctively affected by prematurity as they fold earlier than associative cortices, these results suggest that the development of gyrification across the brain contributes to the impact of gestational age on IQ. In particular, 'early' effects on primary postcentral cortices gyrification seem to convey prematurity effects on subsequent gyrification processes in temporal association cortices and on their relevance for cognitive performance.

Discussion

Using structural MRI and intelligence assessments, we demonstrated for the first time, that widespread increases of gyrification in very premature-born adults contribute to the association between prematurity and general IQ reduction. Results suggest that impaired gyrification is an important marker of prematurity effects on the development of both brain morphology and cognitive performance.

Prematurity and gyrification

Gyrification is permanently altered after premature birth

We found that AMC-based gyrification was increased in VP/VLBW adults, namely in bilateral fronto-temporo-parietal cortices with an emphasis on the right hemisphere (Figure 1). This result was not influenced by gender and/or scanner differences, as we controlled for these factors. The AMC value computes mean cortical curvature by equating an increase in amplitude and frequency of cortical folding to an increase in absolute mean curvature. Furthermore, AMC-based gyrification is strongly correlated with total surface area which signifies that an increase of cortical curvature, termed gyrification translates to increased total cortical surface area (Luders *et al.*, 2006). AMC increases were correlated to premature birth variables (Figure 2), indicating that they were indeed linked to prematurity. In particular, AMC was associated with GA, after correction for BW and INTI, in bilateral lateral frontal cortices and right-sided lateral temporo-parietal cortices (Figure 2A). Furthermore, AMC was associated with BW, independent from GA and INTI, in mostly left-hemispherical lateral frontal and lateral temporal areas (Figure 2B). We found an additional covariance between medical complications at birth and AMC in these areas, indicating that gyrification is a key process affected by premature birth and its complications (Figure 3B). Interestingly, all variables related to premature birth consistently showed an association with early folding pre- and postcentral cortices (White *et al.*, 2010; Zilles *et al.*, 2013; Sun and Hevner, 2014).

Associations with associative cortices that are known to fold later during pregnancy and in the postnatal period (Hill *et al.*, 2010; Garcia *et al.*, 2018) were also present, however less consistent.

In a former study of 19-year-old premature-born adults, surface area was used as a measure of cortical expansion and was found to be decreased in premature-born individuals in lateral fronto-temporo-parietal cortices (Skranes *et al.*, 2013). The pattern of aberrant cortical surface alterations is well in line with our results indicating that prematurity consistently affects distinct areas of the brain. However, since cortical surface area and AMC are different measures of cortical structure they have to be compared with caution. More directly related to our findings, Zhang et al. investigated sulcus depth and surface area in preterm-born, seven-year-old children and found shallower anterior superior temporal sulci. Surface area was shown to be partly decreased (in the posterior superior temporal cortex) and partly increased (in medial frontoparietal cortex) (Zhang *et al.*, 2015). The authors state that while the global gyrification index was decreased in very preterm-born children, the cortex appeared visibly more convoluted with most obvious cortical folding abnormalities found in areas of increased relative surface are, such as the medial frontoparietal cortex.

In conclusion, the widespread pattern of aberrant gyrification in both hemispheres and the direction of altered gyrification towards an increase in AMC are consistent with previous reports and can be considered long lasting effects of prematurity on brain morphology.

Possible causes of aberrant cortical folding in prematurity

To date, the forces and mechanisms that drive the process of cortical folding during brain development in late pregnancy and early postnatal life are not completely understood. Two main hypotheses exist that explain gyrification as a physical process of folding, underpinned by distinct biological mechanisms. These are: the axonal-tension model (Van Essen, 1997;

Mota and Herculano-Houzel, 2012) and the different-tangential expansion model (Ronan *et al.*, 2014). According to the latter theory, the balanced proliferation of neuronal progenitors that reside in the ventricular and subventricular zone is important for the formation of cortical folds through their influence on cortical expansion and architecture (Sun and Hevner, 2014). In particular, subplate neurons constitute an important cell population that has been shown to influence these processes of cortical growth and architecture formation (Kostovic and Rakic, 1990; Hoerder-Suabedissen *et al.*, 2013).

The subplate is a transient cell compartment in the developing brain that is important for guiding thalamocortical and corticocortical connections of the cortical plate and thus for establishing cortical microarchitecture (Kostovic and Rakic, 1990; Judaš *et al.*, 2010; Hoerder-Suabedissen and Molnár, 2015). The subplate, which is most prominent in late maturing and developing association cortices has been shown to be selectively vulnerable to transient hypoxia and hypoxia-ischemia in premature birth resulting in impaired dendritic arborization and altered cortical microstructure (Kanold and Shatz, 2006; Dean *et al.*, 2013; Zilles *et al.*, 2013; McClendon *et al.*, 2017). As this is a basic mechanism of neurodevelopment, it can be hypothesised that subplate damage in prematurity has an impact on cortical folding by altering cortical growth and microstructure (Zilles *et al.*, 2013; Sun and Hevner, 2014).

Prematurity, gyrification, and cognitive performance

Gyrification in temporal association cortices specifically contributes to the impact of prematurity on reduced IQ.

We have shown that gyrification in temporal association cortices, but not in earlier folding primary cortices, statistically mediates the association between prematurity and reduced IQ (Figure 3). This underscores the importance of later-developing, associative cortices for cognitive development and is in line with a recent study on healthy controls investigating

associations between gyrification and cognitive ability (Gregory *et al.*, 2016). Gregory et al. found the highest correlation between gyrification and IQ in bihemispheric medial and lateral fronto-parietal cortices and the highest variability of cortical folding in the superolateral temporal lobe. In contrast, we found the lateral temporal association cortices to be most tightly linked to FS-IQ in prematurely born adults. This may be explained by the high variability of cortical folding in these regions that may be influenced by the cognitive development of preterm infants from birth until adulthood.

This raises the question whether aberrant cortical folding is rather the consequence or the cause of impaired cognitive development leading to reduced FS-IQ. In earlier post-mortem studies, the pattern of cortical folding was thought to remain nearly constant after one year of age (Armstrong *et al.*, 1995). However, recent studies suggest that behavioural or environmental factors do have an impact on gyrification later in life. For example, changes in cortical folding have been shown in professional keyboard players and meditation practitioners (Amunts *et al.*, 1997; Luders *et al.*, 2012). Also, gyrification has been shown to decrease with aging, independently from changes in cortical thickness, surface area and volume (Hogstrom *et al.*, 2013; Klein *et al.*, 2014) and to be different in male and female subjects (Luders *et al.*, 2008). More recently, a study on anorexic patients demonstrated that nutritional factors have a rather short-term impact on cortex morphology: in anorexic patients, gyrification was significantly decreased but changed back to normal after body weight restoration (Bernardoni *et al.*, 2018). These studies suggest plasticity of cortical folding to some extent across the lifespan.

Development of the cortical folding pattern expresses the impact of prematurity on reduced IQ

In our sequential mediation analysis, we observed an indirect statistical mediation effect of postcentral cortices gyrification on full-scale IQ after premature birth via lateral temporal cortices gyrification (Figure 4).

When interpreting this result, one has to reflect the difference concepts of mediation in a statistical and biological context. Statistical mediation is used to identify and to further characterise mechanisms underlying an observed relationship via introducing one or more variables as mediators (Hayes and Rockwood, 2017). The term 'mediation' reflects mainly two concepts in a biological context: A biological mediator either influences an observed relationship across different scales (e.g. local brain circuits generating a certain kind of behaviour) or across different scales and time points (e.g. the progression of cortex development influences cognitive capacities). Thus, caution is advised when transferring results from statistical mediation to biological models, because in a statistical pathway mediation analysis alternative pathways, which are not included in the model, are ignored, irrespective of their biological significance. In conclusion, while statistical mediation can support or oppose a factor's contribution to a given relationship between two variables, it ignores alternative pathways and thus falls short of explaining complex and concurrent relationships in a biological model.

The sequential mediation effect we have observed for PoCC and LTC is interesting because these distinct cortical areas show different timelines of gyrification during gestational or postnatal brain development (Garcia *et al.*, 2018). The postcentral cortices have been shown to develop rather early during gestation as do other primary sensory and motor regions of the brain starting in the second trimester (Dubois *et al.*, 2008). In contrast, cortical folding in associative cortices has been shown to start later during gestation and to extend into the postnatal period (Hill *et al.*, 2010; Zilles *et al.*, 2013). A recent study on cortical folding in preterm infants postulated a distinct developmental trajectory based on MR imaging between gestational week 28 and 38 (Garcia *et al.*, 2018). The described trajectory of preterm cortical

growth starts around the central sulcus and then migrates peripherally towards the associative cortices (Garcia *et al.*, 2018). This model suggests that gyrification should not be treated as a local phenomenon during cortical development but that it is important to perceive gyrification as a global and interrelated developmental step. A study investigating diffusion properties of the cortex in premature-born and term-born individuals between 27 and 47 weeks post conception showed the highest decrease of cortical fractional anisotropy (FA) as a marker of development in frontal, temporal and parietal association cortices (Ball *et al.*, 2013). This cortical development may in part be related to the dynamics of cortical folding in these areas during the last trimester. Our results show the involvement of primary and associative cortices in permanent gyrification aberrancies. Furthermore, we have shown that the variance of AMC in primary cortices is sequentially associated with the variance of AMC in associative cortices. Given the temporal coincidence of gyrification in primary cortices and premature birth we interpret our results in the following way: Adverse events during premature birth, such as hypoxia, lead to perturbation of cortical folding in early folding cortices which in turn influences aberrant cortical folding in primary cortices on the one hand by a shifted trajectory. Thus, we hypothesize that gyrification in primary cortices might be a prognostic marker for the disruption of neurodevelopment at preterm birth, which should be further investigated in other cohorts of premature-born individuals. Furthermore, we propose the hypothesis that gyrification in secondary, associative cortices might be suitable as a treatment target after premature birth. For the latter point, one should keep in mind the aforementioned dynamics of post-natal gyrification including altered gyrification of professionals in keyboard playing and mediation practice (Amunts *et al.*, 1997; Luders *et al.*, 2012) .

Strengths and limitations

Some points should be carefully considered when interpreting our results. First, the current sample is biased to VP/VLBW adults with less severe neonatal complications, less functional

impairments, and higher IQ. Individuals with stronger birth complications and/or severe lasting impairments in the initial BLS sample were more likely both to be excluded in initial screening for MRI due to exclusion criteria for MRI (for example infantile cerebral palsy). Thus, differences in gyrification between VP/VLBW and term control adults reported here are conservative estimates of true differences. Second, the study sample was limited by MRI- and study-related contraindications including a history of severe neurological disorders (e.g. epilepsy, multiple sclerosis, cerebral haemorrhage, traumatic brain injury, tinnitus), severe back problems, (potential) pregnancy, severely impaired vision, as well as non-removable ferromagnetic implants (e.g. pacemakers). Third, the current sample has the strength of large size (101 VP/VLBW and 111 FT adults), enhancing the generalizability of our findings. A strength of our study is that a relevant impact of patient age on gyrification measures at the time of the MRI scan is excluded due to the inclusion of preterm and term subjects who had approximately the same age of 26 years. When interpreting our results about the significant mediation of prematurity effects on adult full-scale IQ by gyrification measures one has to keep in mind that the statistical model is inherently limited as explained in the 'Discussion' section. We have identified aberrant AMC-based gyrification in different, primary and associative cortical areas serving as markers of impaired neurodevelopmental processes which contribute to prematurity effects on adult full-scale IQ in a sequential statistical mediation model. However, complementary effects as well as markers and mechanisms of impaired neurodevelopment, for example as assessed by structural or functional connectivity may also have a mediating effect on adult cognitive performance. These markers have to be taken into account in further studies in order to create a more general understanding of cognitive impairments after premature birth. One technical limitation of our study includes the measure of local gyrification, which varies among studies and is heavily dependent on the software used, limiting comparability. In our study, we used a curvature-based approach to locally measure gyrification of the brain via AMC. This measure cannot be used interchangeably

with surface-based lGI which has been used in most studies so far (Luders *et al.*, 2006; Schaer *et al.*, 2008). A recent study examining anorexic patients compared these two measures and demonstrated that the direction of alterations (increased vs. decreased gyrification) is generally opposite for AMC and lGI while the cortical areas identified as abnormal corresponded well between the two measures (Ronan *et al.*, 2014; Bernardoni *et al.*, 2018).

Conclusion

We have shown widespread differences in gyrification between premature-born adults and full-term controls suggesting that disturbances of gyrification in the last trimester alter brain morphology permanently. This alteration in brain morphology is functionally relevant since it is associated with FS-IQ and contributes to the effect of prematurity on reduced cognitive performance. While there may be various other potential measures of brain structure, structural and functional connectivity that could potentially mediate preterm effects on cognitive performance, gyrification seems to be an important morphological characteristic and expression of aberrant brain development in this respect. Moreover, we showed that this effect is specific for the lateral temporal associative cortex. Finally, we suggested the importance of the development of gyrification from early-folding, postcentral cortices towards more lateral, associative cortices through our sequential mediation model. We propose that gyrification in early folding cortices might potentially serve as a prognostic marker after premature birth while gyrification in cortices that fold later during development might serve as treatment target for specific therapeutic interventions aiming at improving the neurocognitive outcomes of premature birth.

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Tables:

Table 1: Demographical, clinical, and cognitive data.

	VP/VLBW (n=101)			FT (n=111)			
	M	SD	Range	M	SD	Range	p value
Sex (male/female)	58/43			66/45			0.167
Age (years)	27.71	± 0.61	25.7 – 28.3	26.84	± 0.74	25.5 – 28.9	0.765
GA (weeks)	30.5	± 2.1	25 - 36	39.7	± 1.1	37 - 42	<0.001
BW (g)	1325	± 313	630 - 2070	3398	± 444	2120 - 4670	<0.001
Hospitalization (days)	72.2	± 26.4	24 - 170	6.9	± 3.0	2 - 26	<0.001
INTI (a.u.)	11.6	± 3.8	3 – 18	-	-	-	n.a.
SES[a] (a.u.)	29/44/28		1-3	35/50/26		1-3	0.760
Maternal age (years)	29.5	± 4.8	16 - 41	29.4	± 5.2	18 - 42	0.889
Full-scale IQ[b] (a.u.)	94.1	± 12.7	64 - 131	102.5	± 11.9	77 - 130	<0.001

Statistical comparisons: sex, SES with χ^2 statistics; age, GA, BW, Hospital, maternal age, IQ with two sample t-tests.

Abbreviations: GA, gestational age; BW, birth weight; Hospital, duration of hospitalization; INTI, Intensity of Neonatal Treatment (Morbidity) Index; SES, socioeconomic status at birth; maternal age, maternal age at birth; IQ intelligence quotient.

[a]1=upper class, 2=middle class, 3=lower class

[b]Data are based on 97 VP/VLBW and 108 FT born persons.

Table 2: Cortical areas of increased absolute mean curvature index in prematurely born adults compared to term-born controls.

p (FWE-corrected)	Cluster size (number of significant vertices)	Overlap of atlas region	
Left hemisphere			
0.00900	11604	12%	postcentral
		10%	supramarginal
		9%	superiortemporal
		9%	precentral
		8%	lateraloccipital
		8%	inferiorparietal
		7%	middletemporal
		6%	precuneus
		5%	superiorparietal
		5%	inferiortemporal
		4%	fusiform
		4%	parsopercularis
		3%	rostralmiddlefrontal
		2%	parstriangularis
		1%	parahippocampal
		1%	entorhinal
		1%	parsorbitalis
		1%	lingual
Right hemisphere			
0.00640	14509	10%	postcentral
		9%	inferiorparietal
		9%	precentral
		7%	rostralmiddlefrontal
		7%	lateraloccipital
		6%	supramarginal
		5%	middletemporal
		5%	precuneus
		5%	inferiortemporal
		4%	superiortemporal
		4%	superiorfrontal
		4%	superiorparietal
		3%	parstriangularis
		3%	medialorbitofrontal
		3%	fusiform
		3%	caudalmiddlefrontal
		2%	parsopercularis
		2%	posteriorcingulate
		2%	lateralorbitofrontal
		2%	cuneus
		1%	rostralanteriorcingulate
		1%	parsorbitalis
		1%	paracentral
		1%	caudalanteriorcingulate

A two-sample t-test was performed between prematurely born adults (VP/VLBW, n=101) and full-term controls (FT, n=101). Sex and scanner served as covariates of no interest. Results are thresholded at $p<0.05$, FWE-corrected, threshold-free cluster enhancement was used. Atlas labeling was performed according to the Desikan-Killiany atlas (Desikan *et al.*, 2006).Abbreviations: VP/VLBW: very preterm and/or very low birth weight; FT: full-term; FWE: family-wise error.

Figure legends:

Figure 1. Increased absolute mean curvature in very premature-born adults. Statistical parametric map of group comparison for AMC between VP/VLBW and FT adults. Bihemispheric lateral and medial views are shown. Two-sample t-test, $p<0.05$, FWE-corrected, threshold-free cluster enhancement was used. Color bars indicate p-values for increased AMC in the VP/VLBW group. Warm colors represent lower p-values. Abbreviations: AMC: absolute mean curvature, FT: full-term, VP/VLBW: very preterm and/or very low birth weight.

Figure 2. Associations of increased absolute mean curvature with variables of premature birth.

A) Statistical parametric map of negative associations between AMC and GA within the VP/VLBW group, controlled for residuals of BW and INTI, sex and scanner.

B) Statistical parametric map of negative associations between AMC and BW within the VP/VLBW group, controlled for residuals of GA and INTI, sex and scanner.

C) Statistical parametric map of positive associations between AMC and INTI within the VP/VLBW group, controlled for residuals of GA and BW, sex and scanner.

Bihemispheric lateral and medial views are shown. Multiple regression analyses, $p<0.05$, FWE-corrected, threshold-free cluster enhancement was used. Between-group difference in AMC were used as explicit mask. Color bars indicate p-values for associations between AMC and GA (A), BW (B), and INTI (C), respectively, in the VP/VLBW group. Warm colors represent lower p-values.

Abbreviations: AMC: absolute mean curvature, FT: full-term, VP/VLBW: very preterm and/or very low birth weight, GA: gestational age; BW: birth weight; INTI: intensity of neonatal treatment index.

Figure 3. Linking increased absolute mean curvature, reduced full-scale IQ, and prematurity.

A) Statistical parametric map of negative associations between FS-IQ and AMC within the VP/VLBW group. Bihemispheric lateral and medial views are shown. Multiple regression analyses, $p<0.05$, FWE-corrected, threshold-free cluster enhancement was used. Between-group difference in AMC were used as explicit mask. Color bars indicate p-values for negative associations between AMC and FS-IQ in the VP/VLBW group. Warm colors represent lower p-values.

Two-way path diagrams are shown to illustrate results of mediation analyses:

B) Scheme of mediation analysis for mean AMC of the group difference cluster of Figure 1.

C) Scheme of mediation analysis for mean AMC restricted to bilateral postcentral cortices and to bilateral lateral temporal cortices.

Abbreviations: AMC: absolute mean curvature, FT: full-term, FS-IQ: full-scale IQ, LTC: lateral temporal cortex, PoCC: postcentral cortex, VP/VLBW: very preterm and/or very low birth weight.

Figure 4. Brain gyrification development mediates the association between prematurity and adult full-scale IQ.

Three-way path diagrams are shown to illustrate results of mediation analyses:

A) 'Sequential' mediation analysis comparing the mediating effects of AMC in the postcentral cortices (PoCC) and the lateral temporal cortices (LTC) between prematurity (= causal variable) and FS-IQ (= outcome variable). Indirect mediating effect of LTC-AMC alone is significant (1.41±0.71; 95%-CI: 0.23-3.00). Sequential mediation effect of PoCC-AMC on FS-IQ via LTC-AMC is significant (1.22±0.56; 95%-CI: 0.23-2.39)

B) 'Sequential' mediation analysis comparing the mediating effects of AMC in the PoCC and the LTC between gestational age (= causal variable) and FS-IQ (= outcome variable). Indirect

mediating effect of LTC-AMC alone is significant (0.63±0.27; 95%-CI: 0.15-1.20). Sequential mediation effect of PoCC-AMC on FS-IQ via LTC-AMC is significant (0.32±0.17; 95%-CI: 0.05-0.70)

Abbreviations: AMC: absolute mean curvature, FS-IQ: full-scale IQ, PoCC: postcentral cortices, LTC: lateral temporal cortices, CI: confidence interval.