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Intrinsic Functional Connectivity in Preterm Infants with Fetal Growth Restriction Evaluated at 12 Months Corrected Age

Nelly Padilla1, Peter Fransson2, Antonio Donaire3, Francesc Figueras4, Angela Arranz4, Magdalena Sanz-Cortés4, Violeta Tenorio4, Núria Bargallo5, Carme Junqué6, Hugo Lagercrantz1,7, Ulrika Ådén1,7,†, and Eduard Gratacós4,†

1Department of Women’s and Children’s Health, Karolinska Institutet, 171 76 Stockholm, Sweden, 2Department of Clinical Neuroscience, Karolinska Institutet, 171 77 Stockholm, Sweden, 3Department of Neurology, Institute of Neuroscience, Hospital Clinic, Universidad de Barcelona and Institut D’investigacions Biomèdiques August Pi i Sunyer (IDIBAPS), 08036 Barcelona, Spain, 4BCNatal—Barcelona Center for Maternal–Fetal and Neonatal Medicine (Hospital Clinic and Hospital Sant Joan de Deu), IDIBAPS, University of Barcelona, and Centre for Biomedical Research on Rare Diseases (CIBER-ER), 08028 Barcelona, Spain, 5Department of Radiology, Centre de Diagnòstic per la Imatge, CDIC, Hospital Clinic, Universitat de Barcelona, 08036 Barcelona, Spain, 6Department of Psychiatry and Clinical Psychobiology, Faculty of Medicine, Universitat de Barcelona, 08036 Barcelona, Spain, and 7Department of Neonatology, Karolinska University Hospital, 171 76 Stockholm, Sweden

Address correspondence to Nelly Padilla, MD, PhD, Neonatal Research Unit, Q2:07, Astrid Lindgren Children’s Hospital, 171 76 Stockholm, Sweden.

Email: nelly.padilla@ki.se

†E. Gratacós and U. Ådén shared last authorship

Abstract

Fetal growth restriction (FGR) affects brain development in preterm infants, but little is known about its effects on resting-state functional connectivity. We compared 20 preterm infants, born at <34 weeks of gestation with abnormal antenatal Doppler measurements and birth weights <10th percentile, with 20 appropriate for gestational age preterm infants of similar gestational age and 20 term infants. They were scanned without sedation at 12 months of age and screened for autistic traits at 26 months. Resting functional connectivity was assessed using group independent component analysis and seed-based correlation analysis. The groups showed 10 common resting-state networks involving cortical, subcortical regions, and the cerebellum. Only infants with FGR showed patterns of increased connectivity in the visual network and decreased connectivity in the auditory/language and dorsal attention networks. No significant differences between groups were found using seed-based correlation analysis. FGR infants displayed a higher frequency of early autism features, related to decreased connectivity involving the salience network, than term infants. These data suggest that FGR is an independent risk factor for disrupted intrinsic functional connectivity in preterm infants when they are 1-year old and provide more clues about the neurodevelopmental abnormalities reported in this population.

Key words: autism spectrum disorder, fetal growth restriction, neurodevelopment, prematurity, resting state functional connectivity
Introduction

Approximately 5–10% of fetuses are born growth restricted, with a minority of cases representing a severe disease associated to placental insufficiency and prematurity. The combination of prematurity and fetal growth restriction (FGR) has been shown to result in a higher rate of perinatal complications and consequently with worse neurodevelopmental outcome. (Murray et al. 2015). At 1 year of age, FGR is associated with reduce global and regional brain volume, reduced cortical gray matter, and an unusual developmental pattern of white matter (Padilla et al. 2011, 2014). Despite this array of structural global and regional level findings, it is unclear how these alterations might be reflected at a network level defined by intrinsic functional connectivity. In this regard, the functional organization of the brain in these infants remains unexplored, despite several reports that studied functional connectivity in preterm neonates (Fransson et al. 2007; Smyser et al. 2010; van den Heuvel et al. 2015; Ball et al. 2016), preterm infants (Damaraju et al. 2010; Lee et al. 2013), and term 1-year-old infants (Gao et al. 2015). Investigating functional brain connectivity in preterm infants with FGR may have important clinical implications, because deviations from normal functional brain development can indicate developmental disorders and enable clinicians to provide targeted interventions to infants at risk. We hypothesized that preterm infants with FGR, with no evidence of brain lesions on conventional MRI, would show disrupted patterns of resting-state functional connectivity at 12 ± 2 months of corrected age. We included 2 control groups for comparison purposes: firstly, a group of preterm infants appropriate for gestational age (AGA), to exclude the effect of prematurity per se, and secondly, a group of term infants as the reference group. Our primary aim was to compare whole-brain functional connectivity between the FGR infants and the AGA and term infants. Our secondary aim was to explore the frequency of early autistic features in all 3 groups and their association with functional connectivity abnormalities. We studied the spontaneous brain activity patterns by using resting-state functional MRI (rs-fMRI). Both, data-driven independent component analysis (ICA) and seed-based correlation analysis (SCA) were conducted.

Methods

Population

The infants of this study were part of a larger prospective research program on FGR involving fetal assessment and short- and long-term postnatal follow up at the Hospital Clinic (Barcelona, Spain). The study cohort comprised of 20 singleton premature infants with FGR diagnosed before 34 weeks of gestation, 20 preterm AGA infants matched with the FGR group by gestational age at birth (±1 week), and 20 AGA term infants. FGR was defined as 1) fetal weight below the 10th percentile for gestational age confirmed at birth and 2) umbilical artery Doppler pulsatility index ≥ 95th percentile in at least 2 consecutive examinations 24 h apart. The exclusion criteria for all groups were: 1) congenital malformations, including chromosomal abnormalities and infections; 2) clinical criteria for choioamnionitis; and 3) intraventricular hemorrhage grade III–IV and/or any grade of periventricular leukomalacia on the neonatal ultrasound. Although we excluded preterm infants with evidence of structural brain injury on conventional MRI, we are fully aware that microscopic white matter lesions not readily seen by conventional neuroimaging might be present in the preterm groups (Haynes et al. 2009). Written informed consent was obtained from the parents of the participating infants and the Institutional Ethics Committee approved the study protocol, recruitment, and scanning procedures.

Screening for Early Autistic Features

In line with previous recommendations for early screening of autism spectrum disorder in infants, we used the Modified Checklist of Autism in Toddlers (M-CHAT) (Zwaigenbaum et al. 2015).

The M-CHAT is a 23-item parent report checklist to screen children for autistic features between 20 and 48 months that simply requires yes or no answers. The checklist assesses developmental domains, such as sensory responsiveness, early language and communication, social relations and early joint attention. A subset of 6 items is considered critical and a child has a positive screening result if he or she fails at least 2 of the 6 critical items or fails any 3 of the total items (Kuban et al. 2009).

MRI Data Acquisition

The final sample that underwent functional connectivity analyses comprised 13 FGR infants, 19 AGA infants, and 19 term infants. Nine datasets—7 from the FGR and one each from the other 2 groups—were excluded because of excessive motion artifacts that required more than one-third of the volumes to be removed, as explained in the image analysis section. All subjects were scanned at 12 ± 2 months of corrected age while they were in a natural sleep state. Data were acquired with a Magneto Tim Trio 3-T system, using a 32-channel head coil (Siemens, Erlangen, Germany). A set of high-resolution T1-weighted was acquired using a previously described protocol (Padilla et al. 2014). Resting state MRI data were collected by using a gradient echo, echo-planar image sequence sensitized to T2* BOLD contrast (TR 2000 ms; TE echo time 20 ms; voxel size 2.0 × 2.0 × 2.0 mm3; flip angle 90°). Each fMRI run included 240 volumes acquired in 8.14 min. The acquisition time of the entire protocol was 30 min. T1- and T2-weighted images were reviewed by a board-certified neuroradiologist (N.B.) to verify that there were no apparent abnormalities.

Image Analysis and Statistics

Functional data were preprocessed using FMRIB Software Libraries version 5.0.1 (FMRIB Laboratory, University of Oxford, England, UK) (Smith et al. 2004). Briefly, it included discarding the first ten volumes of each data set to allow stabilization of the fMRI data signal, slice timing correction, brain extraction, motion correction, spatial realignment, spatial smoothing (5mm full width), and a high-pass filtering cut-off of 100 s (Gaussian-weighted least-squares straight line fitting, with sigma = 50.0 s). The fMRI data were registered to the infant’s T1-weighted structural image, which leads to improve reliability in automatic brain MRI segmentation in this population. Images were then normalized to a 1-year pediatric template (Shi et al. 2011).

Independent Component Analysis

First, an ICA was performed for each of the 3 groups separately using MELODIC v 3.1 software (FSL, Oxford, UK) (Beckmann and Smith 2005). The dimensionality estimation was calculated using the Laplace approximation, implemented in FSL's...
MELODIC. Resting state networks were classified based on their spatial similarity to functional networks described in healthy infants at the same age (Gao et al. 2015) by visually inspecting the aggregate spatial maps. The characteristics of the independent component time courses and their associated Fourier frequency spectrums were also evaluated. Components that apparently represented motion, artifacts, or vessels were identified and excluded.

During the second step, the whole group of preprocessed data, consisting of 51 infants, were concatenated and entered into a ICA group to identify large-scale patterns of functional connectivity common to the whole population. The set of spatial maps from the group average analysis was used to generate subject-specific versions of the spatial maps, and associated time-series, using dual regression (Beckmann et al. 2009). We then tested for differences in connectivity among the infants using SPM8 software (http://www.fil.ion.ucl.ac.uk/spm) running in MATLAB version 7.5 (MathWorks, Natick, MA, USA). First, a one-way analysis of covariance was performed to determine the main effect of groups on functional connectivity, followed by 2-sample t-tests analyses. Frame displacement, intracranial volume, gestational age, and weight at 12 months were included in the models as covariates. We tested statistical thresholds of $P < 0.05$ family-wise error (FWE) corrected for multiple comparisons and $P < 0.001$ uncorrected.

**Seed-Based Functional Connectivity**

To remove the effect of images corrupted by motion we applied a scrubbing procedure, using the global measure of signal change and the frame-wise displacement method at a 0.5 threshold (Power et al. 2012). An average of 17.42 (range 0–67), 6.61 (range 0–31), and 14.3 volumes (range 0–59) were censored in the FGR, AGA, and term groups, respectively. The groups did not differ significantly when it came to the number of data points removed. (FGR vs. AGA, $P = 0.09$; FGR vs. term, $P = 0.66$; AGA vs. term $P = 0.15$). A minimum of 5 min of rs-fMRI data, excluding censored frames, was required for subjects to be included in this study. The mean frame-wise displacement calculated for the image volumes that were actually included in the analysis did not significantly differ across groups (0.08 [0.40], FGR group; 0.07 [0.23], AGA group, 0.09 [0.04], term group; $F = 0.74, P = 0.39$), suggesting that the groups were well matched for residual motion.

A temporal band-pass filtering (0.01-0.1Hz) was applied. The mean signal from the white matter, cerebrospinal fluid, whole brain, and 6-motion parameters were removed using linear regression. T1-weighted structural images underwent brain extraction, co-registration to the functional images and segmentation into gray matter, white matter and cerebrospinal fluid components using a pediatric template from 1-year-old infants (Shi et al. 2011).

This analysis produced individual participant-level correlation maps identifying regions that were functionally connected to the region of interest. The seeds were selected based on previous rs-fMRI investigations in healthy 1-year-old infants (Gao et al. 2015). Seeds were defined as 6mm (radius) spheres in the left hemisphere and placed in the occipital cortex (medial and pole regions), primary motor cortex (leg and hand and face regions), auditory/language cortex (Heschl’s gyrus), dorsal parietal cortex, posterior cingulate cortex, thalamus, anterior insular cortex, and cerebellar cortex. The peak coordinates are shown in Supplementary Table S1. All seeds were initially centered on the anatomical atlas from 1-year-old infants at coordinates corresponding to the chosen regions. The average time series were then extracted from each seed region for each individual subject and correlated with all the voxels in the whole brain to define the functional connectivity map of each network by using the FEAT v6.0. Intracranial volume, gestational age, and weight at 12 months of age were also included as additional regressors in the model. After that, a group level analysis was carried out using FMRIB’s Local Analysis of Mixed Effects (FLAME) (Woolrich et al. 2004). The general linear model was applied to test for group averages (F test) and differences among the groups (t-test) in terms of decreased and increased connectivity. Z statistic images were thresholded using clusters determined by $Z > 2.3$ and a cluster significance threshold of corrected $P < 0.05$ and uncorrected $P < 0.001$.

**Early Autistic Features and Functional Connectivity**

As a complementary analysis we compared the functional connectivity maps obtained from ICA and SCA of the salience network and the default mode network (DMN) (Bos et al. 2014) from infants with positive screen and those with negative screen. Comparisons between groups were performed applying a 2-sample t-tests analyses on the corresponding components obtained from the ICA and FLAME on the correlation maps obtained from the SCA as described above.

**RESULTS**

**Population**

The perinatal, demographic, and anthropometric characteristics of the infants are detailed in Table 1. There were no significant differences between the FGR infants with low- and high-quality fMRI scans (Supplementary Table S2). The preterm groups were similar in term of perinatal status, as shown by the revised Score for Neonatal Acute Physiology Perinatal Extension (Richardson et al. 1993). Infants with FGR had significantly lower length and head circumference measurements at 12 months in relation to the other groups.

**Group Independent Component Analysis**

The ICA produced 38, 42, and 36 independent components in the FGR, AGA, and term infants, respectively. Of these, 16 FGR, 20 AGA, and 20 term components were judged to be noise-related artifacts, representing cerebral spinal fluid, ventricles, head motion, and/or large blood vessels. We identified 11 FGR, 11 AGA, and 4 term AGA components as unknown or not matching any network. There were no significant differences in motion parameters across datasets (translation, $F = 1.51, P = 0.23$; rotation, $F = 1.39, P = 0.26$).

The rest of the components were grouped into 10 cortical, subcortical, and cerebellar functional networks previously described in 1-year-old infants (Fig. 1). Minor differences were identified with regard to the cerebellar and salience networks. In the FGR group the cerebellar network was incomplete, covering the left hemisphere and only part of the right hemisphere (Fig. 1E). In the AGA and term groups, the cerebellar network was seen as separate right and left maps significantly correlated (AGA group, $r = 0.31, P < 0.001$; term group, $r = 0.48, P < 0.001$). The salience network showed a different topology on each of the 3 groups (Fig. 1I). Of note, the presence of fragmented networks has been previously reported in pediatric populations (Liu et al. 2008; Damaraju et al. 2010) and is considered to
be related with a high ICA model order estimation (Abou-Elseoud et al. 2010).

**Dual regression analysis**

We found significant differences in the visual (occipital pole) $F = 10.5, P < 0.001$; dorsal attention, $F = 11.0, P < 0.001$; and auditory/language networks, $F = 2.3, P < 0.001$ (Fig. 2A). The differences in the visual network survived FWE correction. In the visual network the FGR group showed a pattern of increased connectivity compared with the AGA group (calcarine fissure, voxels = 15, peak t = 6.1, FWE $P = 0.02$) (Fig. 2A). In the dorsal attention network, the FGR group showed hypoconnectivity compared with the term group (frontal pole, voxels = 15, peak t = 5.17, $P < 0.001$) (Fig. 2B). No differences were detected between the AGA and term groups. In the auditory/language network, the AGA group showed increased connectivity compared with the FGR group (middle frontal gyrus, voxels = 19, peak t = 4.42, $P < 0.001$) and term groups (middle frontal gyrus, voxels = 19, peak t = 4.79, $P < 0.001$) (Fig. 2C). The FGR group did not display differences with the term group.

**Seed-Based Correlation Analysis**

The correlation maps for the infants with FGR were generally similar to those observed in the AGA and term groups (Fig. 3). No significant differences were observed across the groups.

**Screening for Early Autistic Features and Functional Connectivity**

Six parents (FGR group = 1; term group = 5) did not answer the questionnaire. The FGR group had a higher frequency of positive results (6/19 [31.6%]) than the control groups (Table 1). The FGR group with positive screen (N = 6) showed hypoconnectivity between the seed based in the anterior salience network and the angular gyrus compared with the term group with negative screen (N = 15) (Fig. 4). No differences were detected between the FGR with positive screen and the AGA groups with negative screen. Analysis of FGR infants with and without positive screen found no significant differences. We did not have sufficient data to perform comparisons between the preterm infants.

### Table 1 Characteristics of the groups

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>IUGR (N = 20)</th>
<th>Preterm (N = 20)</th>
<th>Term (N = 20)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gestational age at delivery, (mean ± SD)</td>
<td>30.7 ± 2.37*</td>
<td>30.0 ± 2.45*</td>
<td>38.5 (1.17)</td>
</tr>
<tr>
<td>range</td>
<td>26.5–34.0</td>
<td>26.0–34.00</td>
<td>37.0–41.5</td>
</tr>
<tr>
<td>Apgar 5 min, median (range) gender (M/F), N (%)</td>
<td>8 (2–10)</td>
<td>9 (2–10)</td>
<td>10 (9–10)</td>
</tr>
<tr>
<td>Birth weight (g), (mean ± SD) range</td>
<td>1094.2 (320.3)**</td>
<td>1390.0 (523.7)*</td>
<td>3559 (496.5)</td>
</tr>
<tr>
<td>Length at birth (cm), (mean ± SD) range</td>
<td>37.9 (3.76)*</td>
<td>40.5 (4.40)*</td>
<td>49.0 (2.25)</td>
</tr>
<tr>
<td>Head circumference at birth (cm), (mean ± SD) range</td>
<td>62.70 (6.6)*</td>
<td>67.0 (7.1)</td>
<td>78.5 (6.9)</td>
</tr>
<tr>
<td>Days in NICU, (mean ± SD) range</td>
<td>12 (10–14)</td>
<td>12 (10–14)</td>
<td>13 (12–14)</td>
</tr>
<tr>
<td>Neonatal morbidity</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Prenatal corticosteroids, N (%)</td>
<td>14/20 (70)</td>
<td>18/20 (90)</td>
<td>–</td>
</tr>
<tr>
<td>SNAP-PE, mean ± SD</td>
<td>18 (0–67)</td>
<td>16 (0–67)</td>
<td>–</td>
</tr>
<tr>
<td>Mechanical ventilation, N (%)</td>
<td>3/20 (15)</td>
<td>4/20 (20)</td>
<td>–</td>
</tr>
<tr>
<td>Postnatal corticosteroids, N (%)</td>
<td>3/20 (15)</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>Late-onset neonatal sepsis, N (%)</td>
<td>4/20 (20)</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>Hypoglycemia, N (%)</td>
<td>7/20 (35)</td>
<td>4/20 (20)</td>
<td>–</td>
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<tr>
<td>Hyperglycemia, N (%)</td>
<td>5/20 (25)</td>
<td>7/20 (35)</td>
<td>–</td>
</tr>
<tr>
<td>Patent ductus arteriosus, N (%)</td>
<td>3/20 (15)</td>
<td>4/20 (20)</td>
<td>–</td>
</tr>
<tr>
<td>Intraventricular hemorrhage grade I-II</td>
<td>4/20 (20)</td>
<td>5/20 (25)</td>
<td>–</td>
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<tr>
<td>Demographic</td>
<td></td>
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<tr>
<td>Maternal age (years)</td>
<td>33.0 (3.89)</td>
<td>32.5 (4.63)</td>
<td>31.0 ± 4.12</td>
</tr>
<tr>
<td>Breast milk, N (%)</td>
<td>13/18 (72.2)</td>
<td>13/17 (76.5)</td>
<td>13/15 (86.7)</td>
</tr>
<tr>
<td>Smoking, N (%)</td>
<td>4/16 (25)</td>
<td>5/17 (29.4)</td>
<td>5/15 (33.3)</td>
</tr>
<tr>
<td>Use of early intervention services</td>
<td>4/20 (20)</td>
<td>4/20 (20)</td>
<td>4/20 (20)</td>
</tr>
<tr>
<td>Head circumference at 12 months (cm), median /range</td>
<td>42.0–46.5</td>
<td>42.0–49.0</td>
<td>43.0–47.5</td>
</tr>
<tr>
<td>Autism symptoms screening (infant)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean age at assessment (months)</td>
<td>26.57 (2.37)</td>
<td>26.92 (2.06)</td>
<td>26.50 ±2.51</td>
</tr>
<tr>
<td>Positive screening, N (%)</td>
<td>6/19 (31.6)**</td>
<td>2/20 (10)</td>
<td>1/15 (6.6)</td>
</tr>
</tbody>
</table>

SD, standard deviation; SNAP-PE, Score for Neonatal Acute Physiology-Perinatal Extension; NICU, Neonatal Intensive care unit.

*P < 0.05 compared with term infants; **P < 0.05 compared with preterm infants. Data are mean ± SD.
The major finding in this study was that preterm infants with FGR had altered intrinsic functional connectivity at 1 year of age independently of prematurity. Although these differences were modest in magnitude, infants with FGR were characterized by patterns of decreased and increased functional connectivity, with a predominant pattern of hypoconnectivity involving the visual, auditory/language and attention networks. Infants with FGR were more likely to display early autism features and those infants who screened positive for autism showed decreased connectivity in the salience network. The current results complement our previous studies and support the notion that FGR is an independent risk factor for altered intrinsic functional connectivity in preterm infants at 12 months of corrected age.

The preterm FGR group demonstrated increased connectivity in the visual network compared with the AGA group. There were no major differences between the AGA group and the term group, suggesting that FGR was a major factor that affected the development of this network in our cohort of preterm infants. Increased regional connectivity in the visual cortex is consistent with a previous study showing an accelerated maturation of visual evoked potentials as a result of hemodynamic adaptation to chronic placental insufficiency (Scherjon et al. 1996). This is in accordance with our previous work demonstrating white matter increases in the occipital cortex in FGR infants (Padilla et al. 2011). Correlation of our findings with neurodevelopmental outcomes will be valuable, and is being undertaken in this cohort.

Unexpectedly, the AGA group showed increased connectivity in the auditory/language network compared with the other 2 groups. The lack of differences between the FGR and term groups suggests that in this analysis the term group constitutes an intermediate between the AGA and FGR groups, with the
latter showing the lowest connectivity. These findings may suggest a compensatory strategy in the AGA group (Gozzo et al. 2009) and an impaired cerebral plasticity in the FGR group (Alexandre-Gouabau et al. 2012). Although these differences were found at uncorrected level of significance, reduced connectivity in the auditory/language network is consistent with previously reported language problems in FGR children (Murray et al. 2015). The finding in FGR infants would be considered as a potential biomarker for language problems that may in turn facilitate early interventions.

The FGR group showed decreased connectivity in the dorsal attention network compared with the term infants. This finding was expected, since previous studies have demonstrated attention problems in FGR infants from the neonatal period (Figueras et al. 2011) to adolescence and adulthood (Sucksdorff et al. 2015). Despite the lack of comparable studies, the results are consistent with previous investigations that have related decreased functional activation in parietal regions during a working-memory selective attention task with reduced attention capacity in children born preterm (Griffiths et al. 2013). Our study adds new data concerning the possible functional substrates related to attention difficulties in preterm infants with FGR.

The mechanisms underlying the observed altered functional connectivity in the FGR group would be related with the fetal brain response to chronic hypoxia. In FGR fetuses the cerebral blood flow shows regional changes as the fetal condition worsens (Hernandez-Andrade et al. 2008). After an early increased blood flow in the frontal area a decreased frontal perfusion in favor of the basal ganglia and cerebellum is established. Thus, the vulnerability or protection of specific brain regions will depend on the stage of fetal hemodynamic deterioration. In addition, given that the structural networks shape the topology of functional networks in an interdependent relationship (van den Heuvel and Sporns 2013), it is likely that microstructural white matter alterations and regional gray matter volume

Figure 2. Within network connectivity: independent component analysis. Functional networks with altered functional connectivity. (A) Visual network (FGR > AGA, voxels = 14). (B) Dorsal attention network (FGR < term, voxels =13). (C) auditory/language network (FGR < AGA, voxels = 17; AGA > term, voxels=17).
changes described in FGR infants (Padilla et al. 2014) may contribute to the altered pattern of functional connectivity found here.

In line with previous studies, we found an increased frequency of early autistic features in the FGR group (Gardener et al. 2011). However, in contrast with previous evidence (Padilla et al. 2015) we did not find any increase of autistic traits in the preterm AGA group. The reasons for this unexpected finding are not clear. Further diagnostic assessment of the preterm groups is needed to determine the true rates of autism in these infants. Compared with the term group with negative screen, the FGR group with positive screen showed reduced functional connectivity between the anterior insula and the angular gyrus, identified by SCA. Even that we could not find specific differences in the DMN, impaired connectivity in the angular gyrus indirectly reflects involvement of the DMN since this structure is identified as a key parietal node of the DMN (Seghier 2013). Notably, the hypoconnectivity pattern was not reflected by ICA. In this regard, hypoconnectivity in autism is generally found in studies using SCA compared with studies using ICA, suggesting that hypoconnectivity may be contingent on specific methodological choices (Müller et al. 2011). There were no differences between the FGR group with positive screen and the AGA group with negative screen. This could be due to that in terms of brain differences the AGA infants constitute an intermediate group between the other groups with more pronounced differences in the FGR infants as compared with the term infants (Padilla et al. 2011). Thus, differences between preterm groups may not be evident and may require a larger sample size.

Despite the lack of comparable studies, it should be noted that reduced connectivity in one-year old infants at risk for autism has also been previously reported (Righi et al. 2014). Using electroencephalography, the authors in this study found that toddlers at risk of developing autism had reduced linear coherence between frontal and temporo-parietal areas while the infants were listening to speech sounds. Our results complement this previous evidence showing a first assessment of the possible link between prematurity-FGR and impaired functional connectivity. Because of the very small sample size included in this analysis, our results should be interpreted as exploratory and preliminary. Future studies, including larger sample sizes at the same age, are clearly needed to explore this issue.

Preterm FGR infants continued to have significantly lower weight, length, and head circumference measurements at 1 year of age. Given that both groups of preterm infants have
similar levels of neonatal morbidity, the poor growth demonstrated in the FGR group could have been the consequence of nutritional and environmental factors during the first year of life. In order to minimize the detrimental effects of this confounding factor in our results we included the weight at 12 months as a covariate in our statistical models with mainly unchanged results. Thus, the findings here could be attributed in a major extend to FGR.

In this study we used 2 distinct methodological approaches for studying functional connectivity, ICA, and SCA. ICA performs a completely data-driven analysis using spatial independence to decompose data into spatio-temporal components and SCA identifies correlations between the time courses of a group of voxels previously defined and the voxels of the rest of the brain (Kelly et al. 2010). Thus, both methodologies would yield similar results, but differences are expected given that the 2 methodologies process the time-series information in a very different way, which could explain why we could not find significant differences between groups using the maps obtained from the SCA.

The strengths of this study include a well-defined cohort, that was characterized prenatally and followed prospectively, and that the definition of FGR included antenatal Doppler findings. A possible limitation of this study was the number of participants who could not be included in the fMRI analyses. In this regard, functional brain scans of preterm infants with FGR were more often affected by motion artifacts (35%) than AGA scans (5%) likely due to a poorer quality of sleep in the FGR group (Leitner et al. 2002). We did not use any sedation in order to not influence the acquisition of the fMRI (Liu et al. 2008). In order to further reduce the effect of motion on functional connectivity measures, we employed the motion censoring approach (scrubbing), which appears to improve data quality more than many motion regression approaches.

Conclusion

Infants with FGR were characterized by patterns of increased and decreased functional connectivity, with a predominant pattern of hypoconnectivity. The vulnerable networks were the visual, auditory/language, and attention networks. The FGR group faced the highest risk of early autistic features and those who screened positive on the M-CHAT presented decreased functional connectivity, with a predominant pattern of hypoconnectivity. The vulnerable networks were the visual, auditory/language, and attention networks.

Supplementary Material

Supplementary material can be found at: http://www.cercor.oxfordjournals.org/.

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Notes

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