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Longitudinal Assessments of fNIRS Background Functional Connectivity in Low- and Middle-Income Infants during a Social Cognition Task

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Abstract

Shortly after birth, human infants demonstrate behavioural selectivity to social stimuli. However, the neural underpinnings of this selectivity are largely unknown. Here we examine patterns of functional connectivity to determine how regions of the brain interact while processing social stimuli and how these interactions change during the first two years of life. Using functional near-infrared spectroscopy (fNIRS), we measured functional connectivity at 6 ($n = 147$) and 24 ($n = 111$) months of age in infants from Bangladesh who were exposed to varying levels of environmental adversity (i.e., low- and middle-income cohorts). We employed a background functional connectivity approach that regresses out the effects of stimulus-specific univariate responses that are believed to affect functional connectivity. At 6 months, the two cohorts had similar fNIRS patterns, with moderate connectivity estimates for regions within and between hemispheres. At 24 months, the patterns diverged for the two cohorts. Global (brain-wide) connectivity estimates increased from 6 to 24 months for the low-income cohort and decreased for the middle-income cohort. In particular, connectivity estimates among regions of interest (ROIs) within the right hemisphere decreased for the middle-income cohort, providing evidence of neural specialization by two years of age. These findings provide insights about the impact of early environmental influences on functional brain development relevant to the processing of social stimuli.

Word Count: 215

Key Words: functional near-infrared spectroscopy; social processing; infancy; functional connectivity; early adversity

Public significance statement: We investigated patterns of brain connectivity during a social cognition task for two cohorts of infants (low and middle income) from Bangladesh at 6 and 24

months of age. We found similar patterns of connectivity at 6 months, but at 24 months substantial differences were observed between the two cohorts, such that only the middle-income cohort showed evidence of specialization. Our findings reveal developmental differences in the networks engaged in social perception and the manner in which early life experiences impact these networks.

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Longitudinal Assessments of fNIRS Background Functional Connectivity in Low- and Middle-Income Infants during a Social Cognition Task

In the first months of life, infants show evidence of sensitivity to social stimuli (Farroni et al., 2002; Johnson et al., 1991). Availability of social input during the early years plays an important role in human development; deprivation of social input during infancy and toddlerhood can lead to both cognitive and social dysfunctions (e.g., Nelson et al., 2007, 2009). Research on adults shows that there is a distinct network of cortical and sub-cortical structures that are active when we process social information, including areas such as the medial prefrontal cortex (mPFC), the anterior cingulate cortex (ACC), the inferior frontal gyrus, the superior temporal sulcus (STS), the amygdala, and the anterior insula (Frith & Frith, 2007). However, how this network emerges during the dramatic changes in social processing during infancy warrants further exploration.

Neuroimaging studies are frequently used to record the magnitude of the haemodynamic response in either individual channels or regions of interest (ROIs, i.e., groups of channels) when infants are presented with social and non-social stimuli, with the goal of providing information about the localization and selectivity/sensitivity of the responses in the infant brain to this social versus non-social contrast. Beginning as early as the first day of life, newborns show selective responses of the bilateral posterior temporal cortex (relative to dynamic non-social stimuli) to facial, but not hand, motion (Farroni et al., 2013). At 5 months of age, the superior temporal cortex is active during perception of dynamic social stimuli relative to static non-social stimuli (Lloyd-Fox et al., 2009; Tsurumi et al., 2019; van der Kant et al., 2018), whereas responses of the frontotemporal cortex are dependent on the type of social motion (i.e., eye, mouth, hand movements) relative to non-social motion (Lloyd-Fox et al., 2009). Cross-sectional studies have

found that the bilateral frontotemporal cortex is active to dynamic social stimuli relative to non-social stimuli at both 6 and 36 months of age (Perdue et al., 2019). Moreover, between 4 and 24 months of age, the right posterior superior temporal cortex shows sensitivity to visual social stimuli relative to static non-social stimuli, such as images of toys (Lloyd-Fox et al., 2017). Finally, data from the sample we draw on in the present study showed that, for 6-, 24-, and 36-month-old infants, social selectivity is found in the left and right inferior frontal gyrus and posterior superior temporal sulcus/ temporoparietal junction (Pirazzoli et al., 2022).

Reliably, it has been reported that specific brain regions show consistent patterns of responses to social versus non-social stimuli, in both high-income and low-income samples, and across numerous geographic sub-populations of infants being raised in different cultures (e.g., the United Kingdom [Farroni et al., 2013; Hakuno et al., 2018, 2020; Lloyd-Fox et al., 2009], the Netherlands [Braukmann et al., 2018], the Gambia [Lloyd-Fox et al., 2014, 2017], Bangladesh [Perdue et al., 2019; Pirazzoli et al., 2022], Japan [Kobayashi et al., 2011; Nakato et al., 2011], and the United States [Fava et al., 2014]). Importantly, these studies provide evidence of focal activation within specific regions of the brain that appear to be engaged during the processing of social stimuli.

In the current study, we expand this past work to investigate the neural networks engaged during a well-established social cognition task in infancy and how these networks change longitudinally and across cohorts who have different levels of exposure to adversity. Neural connectivity patterns provide insights into the networks in the brain that are structurally and functionally interconnected (He & Evans, 2010; Sporns, 2013). In particular, functional connectivity provides information about the functional integration of, and the temporally coherent activity patterns among, brain regions (Damoiseaux et al., 2006; Fox & Raichle, 2007).

By identifying which brain regions form networks and how the strength or profile of connectivity within these networks changes across development, we can better understand the protracted developmental trajectory of long-range neural networks that support the processing of different types of environmental information. Johnson (2001) hypothesizes that these neural networks undergo developmental reorganization, and that this reorganization is related to typical and atypical developmental processes. Much of this reorganization is thought to occur early in life, with the structure (e.g., Matsuzawa et al., 2001) and function (e.g., Gao et al., 2015) of the brain developing rapidly during the first two years. Thus, investigating long-range neural networks in infancy, using functional connectivity approaches, is crucial.

Using Task-Based Functional Connectivity to Investigate Social Cognitive Development

Task-based functional connectivity analyses capture the temporally dynamic interactions between brain regions when humans are engaged in a task. Assessments of task-based functional connectivity identify changes to network connectivity and organization that map onto distinct cognitive processes (Stevens, 2016). In their review, Gonzalez-Castillo and Bandettini (2018) note that studies of task-based functional connectivity find variations in patterns of connectivity across development, with higher inter-network connectivity for younger subjects during task compared to higher inter-network connectivity for older subjects during rest. Task-based functional connectivity analyses have shown the development and maturation of cognitive control, motor control, language processing, and social information processing neural networks in adolescence (see Stevens, 2016 for review).

To date, there have been few studies investigating the development of functional networks engaged during cognitive or social tasks in infancy and early childhood (cf. Baek et al., 2022; Colomer et al., 2022; Jasińska et al., 2021). Whereas for resting-state studies infants are

often tested while asleep, for task-based studies, infants need to be awake and engaged and thus fMRI is a challenging neuroimaging methodology (though see Ellis et al., 2020; Ellis & Turk-Browne, 2018). In contrast, with its robust resistance to motion artifacts (Brigadoi et al., 2014; Nishiyori, 2016; Wilcox & Biondi, 2015), fNIRS is an effective neuroimaging method for task-based functional connectivity (Azhari et al., 2020). Moreover, data can be easily recorded in specified ROIs and subsequently standardized through spatial registration methods (Tsuzuki & Dan, 2014) so that they are anatomically co-localized across participants (Ferradal et al., 2014).

In the present study, we analyzed fNIRS data collected from a large sample of 6- and 24-month Bangladeshi infants from low- and middle-income cohorts (Pirazzoli et al., 2022). Notably, Pirazzoli et al. also include a sample of infants at 36 months of age; these infants were from a different cohort and thus were not included in the present study. Univariate analyses with samples from low- and middle-income countries mirror findings from high-income samples, with social selectivity (i.e., differences in activation between social versus non-social stimuli) present at 6 months but more localized as infants get older (see Blasi et al., 2019 for review). Moreover, data from the sample we draw on in the present study confirm that infants at 6, 24, and 36 months show specialization to social stimuli in cortical regions (Pirazzoli et al., 2022). However, how patterns of connectivity in this task change with age remains unknown. In the present study, extending previous work, we take a task-based functional connectivity approach to understand the neural networks underlying social processing in infancy and how these networks change across the first two years of life.

The Effect of Socioeconomic Status on Brain Development

There is substantial evidence for the relation between neurocognitive development and socioeconomic status (see reviews by Blair & Raver, 2016; Ursache & Noble, 2016). The impact

of low socioeconomic status and closely related factors, such as parental education and neighbourhood disadvantage, on the development of several brain networks in childhood and adolescence has been found in fMRI resting state connectivity studies (e.g., Barch et al., 2016; Brody et al., 2019; Marshall et al., 2018; Sripada et al., 2014; Su et al., 2021). In infancy, the literature is sparser, and findings are mixed. For example, using fMRI, the rate of infant brain growth, including the frontal lobe which is involved in social information processing, was found to be associated with family poverty (Hanson et al., 2013). Moreover, EEG work with toddlers shows that height-for age (Xie et al., 2019) and inflammation (Bach et al., 2022) are negatively associated with resting-state functional connectivity in the theta and beta frequency bands, suggesting a broad deleterious effect of early exposure to malnutrition and inflammation on infant brain development. In contrast, using fNIRS, at 5 months of age, no relation was found between socioeconomic status and resting-state functional connectivity in higher-order brain networks (e.g., fronto-parietal network, default mode network, network of homologous-interhemispheric connections; Chajes et al., 2022). Similarly, at 6 months, relations between baseline connectivity in sensorimotor and default mode networks and parental income and education did not survive multiple comparisons (Gao et al., 2015).

With respect to social cognitive development in infancy, few studies have investigated the link between the development of networks linked to social processing and socioeconomic status. Pirazzoli et al. (2022) note that much of the literature examining socioeconomic status and brain development has focused on networks and behavioural outcomes that are linked to later academic achievement; they argue that additional research is needed to determine which risk factors impact social cognitive development and when the impact of such factors manifest. In their fNIRS study, Pirazzoli et al. found that social selectivity was associated with psychosocial

risk factors (i.e., intimate partner violence, verbal abuse, family conflict, maternal depression) but only for toddlers (i.e., 24 and 36 months), not young infants (i.e., 6 months). With respect to connectivity, in a social versus non-social task, lower interhemispheric EEG coherence at 12 months of age was found from the frontal to the central, parietal, temporal, and occipital regions during the social condition, but only for infants from higher-income families; EEG coherence did not differ for the two conditions for infants from economically strained families (Lopera-Perez et al., 2022). In the present study, we build upon these prior studies, examining developmental patterns of task-based connectivity from early infancy (i.e., 6 months) to toddlerhood (i.e., 24 months) and whether these patterns are impacted by differing levels of exposure to adversity.

The Present Study

As there are so few studies looking at the development of task-based functional networks in infancy, it is difficult to have an informed hypothesis for this study, and thus we chose a descriptive approach. However, some insights are provided by a large cross-sectional cohort of children ($n = 157$) between 6 and 20 years of age by Sanchez-Alonso et al. (2021). They assessed whole-brain functional connectivity from fMRI data obtained while participants were either in a resting-state or while viewing a movie. The connectivity patterns were reliably different in these two contexts. Moreover, across age the relative engagement of sub-networks (e.g., language versus posterior multimodal) was also different, with some networks increasing and others decreasing with age. Thus, on the one hand, in the present study, there may be increases in functional connectivity with age, mirroring the general findings from the resting-state functional connectivity literature (Bulgarelli et al., 2020). This would be evidenced by greater number of connections, greater strength of connections across the brain and/or a selective increase in the number of long-range connections, and an accompanying decrease in connectivity in number

and/or strength of short-range connectivity. On the other hand, given that the univariate functional response to social processing shows localization to smaller cortical areas, it could be that the task-based functional networks show decreases in connectivity reflecting a specialization process. These could be evidenced by a general decrease in the strength of connectivity, both for short- and long-range connections.

Furthermore, it is not clear whether we will find differences in this network across the two cohorts. On one hand, there is a high level of similarity in the localization of these responses across study populations internationally and across cohorts from similar environments (Pirazzoli et al., 2022). On the other hand, it is well-established that differential exposure to adversity early in life affects brain development in many ways (Lopera-Perez et al., 2022; Pirazzoli et al., 2022; Xie et al., 2019). Thus, we do not test specific hypotheses in this study – instead, we aim to descriptively investigate the task-based functional networks in this sample using established methods applied to other infant fNIRS datasets and compare the outcomes to the above possibilities. To this end, we examined connectivity across six ROIs spanning frontal to posterior temporal areas (i.e., inferior frontal gyrus [IFG], anterior middle temporal gyrus/superior temporal gyrus [aMTG-STG], and posterior superior temporal sulcus/temporoparietal junction [pSTS-TPJ]), as well as global connectivity (i.e., whole-brain connectivity) and connectivity between individual fNIRS channels separately in each cohort and at each age (see Supplementary Materials).

Notably, the current approach builds from established methods for using background functional connectivity with infants (see Background Connectivity for details) in which connectivity analyses are completed over several blocks using data collected with the experimental paradigm developed by Lloyd-Fox and colleagues which employed an active

baseline approach of using social stimuli compared to a baseline of non-social stimuli (Lloyd-Fox et al., 2009). Based on the constraints of this paradigm, our analytic approach provides a measure of the functional networks that are significantly modulated or engaged during the overall task, without the explicit comparison of functional networks to social versus non-social stimuli. Nonetheless, given that the stimuli in the task vary along the dimensions of social to non-social stimuli, this work makes the assumption that the functional networks engaged will be the ones that are sensitive to this contrast.

Method

Transparency and Openness

We report where the datasets were collected, criteria for data exclusion, all manipulations, and all measures in this study, and we follow the Journal Article Reporting Standards (JARS; Kazak, 2018) as recommended in the American Psychological Association (APA) guidelines. The data reported in this manuscript were collected as part of the Bangladesh Early Adversity Neuroimaging (BEAN) project (<https://www.lcn-bean.org>; see Perdue et al., 2019 for details). In brief, the BEAN project aims to investigate the relations among early risk factors and neurodevelopmental outcomes, using various methodologies including physiological measures, eye tracking, neuroimaging (MRI at 2 and 60 months; fNIRS and EEG at 6, 24, 36, and 60 months¹), and behavioural assessments. Data were analyzed using MATLAB 2021b (MATLAB, 2021) and R, Version 4.2.1 (R Core Team, 2022). The following R packages were used for data analysis and visualization: *tidyverse* (Wickham et al., 2019), *data.table* (Dowle &

¹ In the present study, longitudinal fNIRS data for both cohorts were only available at 6 and 24 months. The ages selected for the BEAN project were predetermined from another project being conducted by the International Centre for Diarrhoeal Disease Research, Bangladesh (ICDDR, B) focusing on vaccine response. The LI cohort were part of this vaccine efficacy trial. The MI cohort were recruited specifically for the BEAN project to act as a middle income control group.

Srinivasan, 2021), *lme4* (Bates et al., 2015), *ggplot2* (Wickham, 2016). This study's design and its analysis were not pre-registered.

Participants

The sample consisted of infants who were recruited for the BEAN project (see Supplementary Materials for additional demographic information). This project was approved by the Institutional Review Board of Boston's Children's Hospital and the Ethical Review Committee of International Centre for Diarrheal Disease Research, Bangladesh. Ethics approval was also received by the University of British Columbia's Behavioural Research Ethics Board. Parents provided informed consent before enrollment in the fNIRS study. The BEAN project is comprised of participants recruited in two cohorts: Low income (LI; $n = 130$) and middle income (MI; $n = 104$) from an urban district in Dhaka, Bangladesh. Notably, while the two cohorts came from the same neighbourhood, the MI cohort had higher household income ($t(231) = 14.91, p < .001$) and higher maternal education ($t(232) = 9.38, p < .001$) than the LI cohort. In the present study, for both cohorts, we use longitudinal functional near-infrared spectroscopy (fNIRS) data from infants at 6 and 24 months-of age. Inclusion criteria included no known neurological conditions, hearing impairments, or visual impairments. All infants were born full-term (> 37 weeks gestation). The final sample, used for all subsequent analyses, included 147 infants at 6 months ($n_{LI} = 83; n_{MI} = 64$) and 111 infants at 24 months ($n_{LI} = 52, n_{MI} = 59$) who provided usable fNIRS data after preprocessing and behavioural coding analyses (see fNIRS Data Collection and Preprocessing for details).

Stimuli and Task Procedure

The stimuli and experimental design used in the present study, also known as the *peek-a-boo* task, are described in detail in Lloyd-Fox et al. (2012, 2014, 2017). Visual social stimuli and

visual non-social stimuli were presented in full colour on a screen with a viewing distance of approximately 100 cm. The visual social stimuli consisted of short movie clips of women performing nursery rhyme gestures. The visual non-social stimuli consisted of photos of methods of transportation. Social sounds, non-social sounds, and silence each accompanied one third of the visual social trials.² The stimuli were adapted to be familiar to the study population, such that the women in the social videos were of South Asian descent and the modes of transportation were typical for Bangladesh (Perdue et al., 2019). Both social and non-social trials were 9-12 seconds in length. Each experimental trial was followed by a baseline trial. The maximum time for the task was approximately 10 minutes for 24-month-olds and 6 minutes for 6-month-olds.

Throughout the social cognition task, infants were seated on a parent's lap approximately 100 cm away from a video screen (1920 x 1080; 21.5 cm). Parents were instructed to only interact with their infant if they became fussy or sought their attention. The experimenter was to remain out of view from the parent and infant, standing behind them during the trials and only interacting with the infant when necessary (i.e., redirect the infant's attention toward the screen). If the infant was fussy or too distracted, the stimulus presentation was paused, and the infant was able to take a short break. The experiment ended when the maximum number of experimental trials was reached (10 trials per experimental condition at 6 months; 6 trials per experimental condition at 24 months) or when the infant became too fussy to continue. A video camera, placed above the screen, was used to record each session. These recordings were used later to code infant behaviour and potential experimenter interference.

² Notably, while the type of stimuli presented differed for social and non-social conditions (i.e., video vs. photograph; social audio vs. non-social audio vs. silence), in the present study we use a background functional connectivity approach in which the impulse response window accounted for 10 s of stimulus and 10 s of baseline. See Background Connectivity section for details.

fNIRS Data Collection and Preprocessing

Oxygenated (HbO) and deoxygenated (HbR) hemoglobin levels were measured using the Gowerlabs NTS optical topography system (Everdell et al., 2005). Two continuous wavelengths of source light at 780 and 850nm were used. At 6 months, custom-built headbands were used. These headbands consisted of 12 sources and 12 detectors arranged into 32 channels. At 24 months, either custom-built headbands or the EasyCaps were used. Both headgears had the same configuration, consisting of 14 sources and 14 detectors arranged into 38 channels. Optical locations were consistent across both headgears and 32 of the optical locations were identical across the two ages. The 6 additional channels present at 24 months were excluded from all analyses so that direct comparisons could be made between the two ages. At both ages and for both types of headgear, source-detector separation was 2 cm, with channels arranged in two bilateral arrays covering the inferior frontal, mid- to posterior temporal, and inferior parieto-occipital regions (Figure 1). To assess headgear placement quality, photographs were taken before the social cognition task from the front, left, and right sides. Infants' data were excluded from analyses if the headgear shifted more than 1 cm relative to optimal placement.

< Insert Figure 1 Here >

fNIRS data were preprocessed in MATLAB 2021b (*MATLAB*, 2021) using the HomeR2 toolbox (Huppert et al., 2009) following the pipeline described and validated in Di Lorenzo, Pirazzoli et al. (2019). Channels with very low optical density readings (mean < 0.003V) were considered not valid and were excluded; infants who had less than 2/3 valid channels were excluded from further analyses. Raw intensity data was then converted to optical density. Motion artifacts were identified and corrected using spline interpolation (Scholkmann et al., 2010) and wavelet filtering (Molavi & Dumont, 2012). A motion artifact was defined as a portion of signal

that exceeded a threshold in change of amplitude (0.40) and standard deviation (15) within a predefined time window of 1s. These portions and the data points around them (1 s) were marked. Next, spline interpolation was applied channel-by-channel to remove these artifacts ($p = .99$), followed by wavelet motion correction (IQR = 0.80). Following motion correction, any trials that still contained motion artifacts (detected using the same parameters detailed above) between -2 and 16 s relative to trial onset were discarded³.

In addition, behavioural coding was conducted to determine when an infant did not attend to at least 60% of the trial and when the experimenter or parent and child interacted (i.e., social interference) during the experimental trial or the baseline trial prior to the experimental trial. Data were rejected if infants were not looking or social interference occurred for over 50% of the task. To be included in the analyses, infants had to have at least three valid experimental trials. The average number of valid experimental trials for each group were as follows: $M_{6LI} = 20.25$, $M_{6MI} = 15.16$, $M_{24LI} = 15.29$, $M_{24MI} = 14.10$. Overall, 211 infants had fNIRS recordings at 6 months ($n_6 = 119$, $n_{24} = 92$) and 168 infants ($n_6 = 90$, $n_{24} = 78$), had fNIRS recordings at 24 months. Of those infants, 64 infants at 6 months and 57 infants at 24 months were excluded as a result of not completing the NIRS experiment ($n_6 = 4$, $n_{24} = 2$), not attending to the screen for at least 50% of the task ($n_6 = 31$, $n_{24} = 12$), poor cap placement ($n_6 = 9$, $n_{24} = 3$), less than 2/3 valid channels ($n_6 = 5$, $n_{24} = 3$), less than 3 trials ($n_6 = 5$, $n_{24} = 2$), less than 90 s of data ($n_6 = 4$, $n_{24} = 2$), or not having any behavioural coding ($n_6 = 6$, $n_{24} = 33$). The attrition rates for both ages is

³ To confirm that connectivity patterns were not driven by motion artifacts a 2(age: 6 months, 24 months) x 2 (cohort: LI, MI) between subjects ANOVA was conducted on the percentage of data excluded due to motion. There was a significant main effect of age, $F(1, 254) = 17.72$, $p < .001$, $\eta_p^2 = .07$. A greater percentage of trials were excluded at 6 months ($M = 1.42$, $SE = 0.15$) than at 24 months ($M = 0.46$, $SE = 0.17$). Because there was no significant main effect of cohort and no significant interaction, we are confident that the patterns of development in our connectivity analyses are not being driven by motion artifacts.

consistent with the average attrition rate reported in infant fNIRS research (34%; see meta-analysis by Baek et al., 2021).

Finally, a bandpass filter from 0.03 Hz to 1 Hz was applied to reduce slow drifts and high-frequency noise. Data was converted from optical density to HbO and HbR haemoglobin concentrations using the modified Beer Lambert Law (6 months $\text{ppf} = 5.1$; 24 months $\text{ppf} = 6.0$).

Anatomical Localization

For each infant, an anatomical localization of the optode locations was performed to account for variation in head size and array positions. Age and head measurements (i.e., circumference, ear-to-ear over forehead, and over top of head), were used to match each infant to an individual MRI scan from the Neurodevelopmental database (Richards et al., 2016; Sanchez et al., 2012). A virtual array was placed on the MRI head model, based on the algorithm developed by Lloyd-Fox et al. (2014). Array placement was checked for each infant and any inaccuracies were adjusted to align with the photos taken during the experiment. Diffuse optical tomography (DOT) sensitivity functions for each source-detector pair comprising a channel were estimated using photon propagation modeling (Fang, 2010; Fu & Richards, 2021a, 2021b). For this study, we generated an atlas for three ROIs, for both the left and right hemisphere: IFG, aMTG-STG, and pSTS-TPJ. The peek-a-boo task has been shown to elicit activations over these regions (Lloyd-Fox et al., 2009, 2013, 2017; Pirazzoli et al., 2022). The sensitivity of each channel to each ROI was estimated using DOT data: channels with greater than 10% sensitivity to an ROI relative to the brain were selected. If a channel exceeded the 10% threshold for more than one ROI, the ROI with the highest value was selected so that each channel could only be assigned to one ROI. See Figure 1 for the average location of the channels for each ROI at 6 and 24 months.

Background Connectivity

A background connectivity approach was used to assess task-based functional connectivity (i.e., connectivity during the social cognition task) at 6 and 24 months (see Figure S1). This analytic approach reveals functional connectivity without the potential confounding effect of the temporal structure of the stimulus presentation. Failure to control for task-evoked responses may lead to over-inflated correlation estimates because hemodynamic responses in different neural areas are correlated not only because of the connectivity between the areas but also due to these synchronized task-evoked responses (Cole et al., 2019). Using the background connectivity approach, stimulus-evoked responses were modelled and linearly regressed from the data prior to measuring correlations in residual spontaneous fluctuations (Al-Aidroos et al., 2012; Baek et al., 2022; Jaffe-Dax et al., under review).

Specifically, using custom MATLAB (*MATLAB*, 2021) scripts, task-evoked activations were modeled by fitting a generalized linear model (GLM) using a finite impulse responses (FIR) approach to approximate the hemodynamic response (Santosa et al., 2018). This approach was chosen because the hemodynamic responses in infants is not definitively known and using an FIR model gives the most flexibility in modeling the response. In the present study, the impulse response window was 20 s starting at stimulus onset, thereby accounting for 10 s of stimulus and 10 s of baseline. Twenty FIR regressors (one per second) were used to model the neural response. The shape of the hemodynamic response, which we subsequently used as a measure of task-evoked cortical activity, was then estimated using the weighted sums of those impulses. After the model was fit, the residuals from the model were used to measure functional connectivity. For each subject, Pearson correlations over the residuals were used to determine the measures of connectivity. Three estimates of background connectivity were calculated: global

background connectivity, connectivity patterns across neuro-anatomically defined ROIs, and connectivity patterns across channels. To obtain a single global measure of functional connectivity, we averaged the estimates of connectivity between each pair of channels for each infant. For connectivity patterns across ROIs, we averaged the fNIRS data into six ROIs (left and right inferior frontal, mid- to posterior-temporal, and inferior parieto-occipital) before estimating background connectivity. Averaging within ROIs has been shown to increase the quality of the fNIRS signals (Jaffe-Dax et al., under review). For channel-wise connectivity, we estimated the pairwise connectivity between all channels (i.e., 496 estimates of connectivity at 6 and 24 months). In all calculations, the Pearson's r values were Fisher transformed to z -values to aggregate across subjects, and then Fisher transformed back to Pearson's r values. To better understand the patterns of connectivity at both ages and for both cohorts, for all three measures we analyzed both HbO and HbR (e.g., Lloyd-Fox et al., 2011).

Results

Global Background Functional Connectivity

Figure 2 presents spaghetti plots of the change in global connectivity for each infant from 6 months to 24 months. The patterns of global connectivity were quantified in a linear mixed effects model with age (6 months, 24 months), cohort (LI, MI), and chromophore (HbO, HbR) as fixed effects and subject as a random effect. All post hoc comparisons were Bonferroni corrected.

There was a significant main effect of cohort, $F(1, 72) = 5.48, p = .02, \eta_p^2 = .07$. Infants in the LI cohort had greater global connectivity ($M = .14, SE = .008$) than infants in the MI cohort ($M = .11, SE = .007$). There was also a significant main effect of chromophore, $F(1, 216) = 121.88, p < .001, \eta_p^2 = .36$. Consistent with the infant literature in which effects are often

found for HbO, but not HbR (e.g., Cristia et al., 2014) and in part due to surface vascular noise in HbO data (Noah et al., 2021), there was greater connectivity for HbO ($M = .16$, $SE = .006$) than HbR ($M = .09$, $SE = .006$) concentrations. There was no significant main effect of age.

There was a significant age by cohort interaction, $F(1, 216) = 143.69$, $p < .001$, $\eta_p^2 = .40$. Global connectivity increased from 6 to 24 months for the LI cohort, whereas it decreased from 6 to 24 months for the MI cohort (see Figure 2). Moreover, compared to the MI cohort, at 6 months the LI cohort had lower estimates of connectivity whereas at 24 months the LI cohort had greater estimates of connectivity. There were no other significant interactions. Importantly, the patterns of connectivity were similar for the two chromophores with the LI cohort showing an increase and the MI cohort showing a decrease in both HbO and HbR from 6 to 24 months. Thus, for subsequent analyses we only present HbO results in text and include HbR results in Supplementary Materials.

< Insert Figure 2 Here >

Region of Interest Functional Connectivity

We considered 3 ROIs on each of the two hemispheres (6 ROIs total): IFG, aMTG-STG, and pSTS-TPJ. Functional connectivity was estimated for each pair of ROIs (15 total; 6 within hemisphere; 9 between hemisphere) at 6 and 24 months for the LI and MI cohorts. Figure 3 shows the patterns of connectivity for 6 and 24 months-of-age for each cohort. *T*-tests were conducted for each pair of regions to determine if there was a significant change in functional connectivity between regions from 6 to 24-months-of-age (see Table 1), and if there were significant differences in functional connectivity within a region between the LI and MI cohorts (see Table 2). For all analyses, we corrected for multiple comparisons using the Benjamini-Hochberg false discovery rate procedure (FDR; Benjamini & Hochberg, 1995).

For the LI cohort, there was a significant increase in connectivity for 7 pairs of ROIs (2 within-hemisphere, 5 between hemispheres) from 6 to 24 months. Much of the increase in connectivity was observed between hemispheres, suggesting that over time, infants in the LI cohort were forming stronger long-range neural networks that were recruited for the social cognition task. For the MI cohort, there was a significant decrease in connectivity for 3 pairs of ROIs within the right hemisphere and a significant increase in connectivity for 1 pair of ROIs within the left hemisphere and 1 pair of ROIs between hemispheres.

Next, we compared connectivity between cohorts at each age. There were no significant differences between the two cohorts at 6 months of age. Both cohorts had similar patterns, showing moderate estimates of connectivity between ROIs. At 24 months, the differences between the cohorts are much more pronounced. The MI cohort had significantly lower connectivity between 8 pairs of ROIs (3 within the right hemisphere, 5 between hemispheres) than the LI cohort. Although there were differences in the strength of estimates of connectivity for ROIs between hemispheres for the LI and MI cohorts, these differences were less pronounced than the within-hemisphere differences (Figure 3).

< Insert Figure 3 Here >

In summary, the ROI analyses revealed that at 6 months of age connectivity among ROI pairs was also similar for both cohorts, with moderate estimates of connectivity between the IFG, aMTG-STG, and pSTS-TPJ ($r_s = .19$ to $.48$). Just as for global connectivity, the two cohorts diverged at 24 months. In the LI cohort, all ROI pairs both between and within hemispheres had estimates of connectivity that either increased or remained stable ($r_s = .29$ to $.52$), while in the MI cohort all ROI pairs within the right hemisphere had estimates of connectivity that decreased

($r_s = .08$ to $.10$). Again, significant differences in ROI connectivity between the two cohorts were observed only at 24 months.

Table 1

Connectivity between Regions of Interest: Comparisons at 6 and 24 Months within Cohorts

	LI 6 vs. 24	MI 6 vs. 24
Within Hemisphere		
R-IFG, R-aMTG-STG	$t(32) = -3.14, p = .004^*$	$t(40) = 4.92, p < .001^*$
R-IFG, R-pSTS-TPJ	$t(33) = 0.07, p = .945$	$t(41) = 7.03, p < .001^*$
R-aMTG-STG, R-pSTS-TPJ	$t(32) = -1.38, p = .177$	$t(48) = 6.69, p < .001^*$
L-IFG, L-aMTG-STG	$t(33) = -2.24, p = .032$	$t(45) = -3.15, p = .003^*$
L-IFG, L-pSTS-TPJ	$t(33) = -1.43, p = .163$	$t(45) = -0.86, p = .395$
L-aMTG-STG, L-pSTS-TPJ	$t(33) = -4.15, p < .001^*$	$t(49) = -1.52, p = .135$
Between Hemisphere		
R-IFG, L-IFG	$t(33) = -0.57, p = .571$	$t(39) = 0.51, p = .61$
R-IFG, L-aMTG-STG	$t(33) = -3.36, p = .002^*$	$t(41) = -2.56, p = .014^*$
R-IFG, L-pSTS-TPJ	$t(33) = -3.58, p = .001^*$	$t(41) = 0.58, p = .568$
R-aMTG-STG, L-IFG	$t(32) = -2.64, p = .013^*$	$t(44) = 0.41, p = .685$
R-aMTG-STG, L-aMTG-STG	$t(32) = -1.34, p = .190$	$t(48) = -0.42, p = .674$
R-aMTG-STG, L-pSTS-TPJ	$t(32) = -5.33, p < .001^*$	$t(48) = -0.52, p = .602$
R-pSTS-TPJ, L-IFG	$t(33) = 1.00, p = .324$	$t(45) = 0.09, p = .926$
R-pSTS-TPJ, L-aMTG-STG	$t(33) = -1.57, p = .126$	$t(49) = -0.18, p = .861$
R-pSTS-TPJ, L-pSTS-TPJ	$t(33) = -3.12, p = .004^*$	$t(49) = 1.63, p = .109$

Note. * significant at the $p < .05$ level after Benjamini-Hochberg corrections for false detection rates

Table 2

Connectivity between Regions of Interest: Comparisons between Low- and Middle-Income Cohorts at 6 and 24 Months

	6 months LI vs. MI	24 months LI vs. MI
Within Hemisphere		
R-IFG, R-aMTG-STG	$t(147) = -0.66, p = .511$	$t(104) = 7.67, p < .001^*$
R-IFG, R-pSTS-TPJ	$t(147) = 0.08, p = .938$	$t(106) = 4.40, p < .001^*$
R-aMTG-STG, R-pSTS-TPJ	$t(157) = -0.98, p = .331$	$t(104) = 8.60, p < .001^*$
L-IFG, L-aMTG-STG	$t(151) = -2.41, p = .017$	$t(107) = -2.03, p = .045$
L-IFG, L-pSTS-TPJ	$t(152) = -1.54, p = .125$	$t(107) = -0.53, p = .598$
L-aMTG-STG, L-pSTS-TPJ	$t(156) = -2.29, p = .023$	$t(108) = 0.89, p = .373$
Between Hemisphere		
R-IFG, L-IFG	$t(144) = -0.18, p = .855$	$t(106) = -0.17, p = .865$
R-IFG, L-aMTG-STG	$t(146) = -1.04, p = .302$	$t(107) = 0.02, p = .985$
R-IFG, L-pSTS-TPJ	$t(147) = -0.64, p = .521$	$t(107) = 3.23, p = .002^*$
R-aMTG-STG, L-IFG	$t(152) = -1.88, p = .062$	$t(104) = 3.24, p = .002^*$
R-aMTG-STG, L-aMTG-STG	$t(156) = -1.99, p = .049$	$t(105) = -0.24, p = .810$
R-aMTG-STG, L-pSTS-TPJ	$t(157) = -1.24, p = .217$	$t(105) = 3.57, p = .001^*$
R-pSTS-TPJ, L-IFG	$t(152) = 0.26, p = .795$	$t(106) = -0.16, p = .869$
R-pSTS-TPJ, L-aMTG-STG	$t(156) = 0.03, p = .978$	$t(107) = 2.96, p = .004^*$
R-pSTS-TPJ, L-pSTS-TPJ	$t(157) = -0.69, p = .493$	$t(107) = 4.40, p < .001^*$

Note. *significant at the $p < .05$ level after Benjamini-Hochberg corrections for false detection rates

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Channel-Wise Functional Connectivity

All reported significant results with channel-wise functional connectivity were significant after correcting for multiple comparisons with FDR. There were 496 estimates of connectivity among the 32 channels (256 between hemispheres, 120 within the left hemisphere, 120 within the right hemisphere; see Figure 4). To compare 6 versus 24 months, *t*-tests were conducted for each pair of channels to determine if there was a significant change in functional connectivity over time. For the LI cohort, 214 channel pairs were significantly different, with 195 of those pairs showing greater connectivity at 24 months. For the MI cohort, 249 channel pairs were significantly different, with 201 of those pairs showing less connectivity at 24 months. Overall, estimates of connectivity at the channel level changed from 6 to 24 months such that 43% and 50% of channel pairs for the LI and MI cohort, respectively, were significantly different. Of the channel pairs that significantly differed at 6 and 24 months, 91% showed greater connectivity at 24 months for the LI cohort whereas 81% showed less connectivity at 24 months for the MI cohort.

<Insert Figure 4 Here >

Finally, we compared channel-wise connectivity between the two cohorts at 6 and 24 months. At 6 months, there were 42 pairs of channels that significantly differed, with the MI cohort showing greater connectivity for 41 of those pairs than the LI cohort. By contrast, at 24 months, there were 318 pairs of channels that significantly differed, with the MI cohort showing less connectivity for 262 of those pairs than the LI cohort. Overall, estimates of connectivity at the channel level were similar for both groups at 6 months of age. In contrast, by 24 months, 64% of estimates of connectivity differed at the channel level, with over 80% of those differences revealing less connectivity for the MI cohort compared to the LI cohort.

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In summary, the channel-wise analyses revealed that, consistent with both global and ROI analyses, we observed similar patterns of connectivity in both cohorts at 6 months and a diverging picture at 24 months (see Fig 4A). While the LI cohort predominantly showed increases in connectivity from 6 to 24 months, the MI cohort showed decreases in connectivity.

Discussion

In recent years, there has been growing interest in investigating the neural regions supporting social perception during the first two postnatal years using fNIRS. Participants in these studies were recruited from both high-income and low- and middle-income countries or populations (Braukmann et al., 2018; Farroni et al., 2013; Lloyd-Fox et al., 2009, 2011, 2014, 2017; Perdue et al., 2019; Pirazzoli et al., 2022). Consistently, these studies find an early cortical specialization for social perception in the superior temporal cortex, which develops over the first several years. The present study used background functional connectivity analyses (at the global, ROI, and channel level) to investigate the task-based functional connectivity networks supporting social perception and their development in a large sample of infants from Dhaka, Bangladesh. This work extends previous findings to consider not only individual fNIRS channels and brain regions, but the communication between channels and regions to identify the networks responsible for social perception and the development of these networks longitudinally in infancy. Overall, all analytic approaches used for assessing functional connectivity show that the development of neural networks engaged during social processing (the observed developmental/age-related increases and decreases) differ for low-income (LI) versus middle-income (MI) cohorts, with only the latter showing evidence of specialization.

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Social Perception Development and Differential Exposure to Adversity

We found that the two cohorts – low-income and middle-income – showed divergent developmental patterns in functional connectivity during a social perception task. At 6 months of age, for both cohorts, short- and long-range connections, which we classify as within- and between-hemisphere connections, respectively, were engaged during the social versus non-social task. At this age, Baek et al. (2022) similarly found evidence of long-range functional connectivity in the frontoparietal network when infants were engaged in a temporal predictability task and speculated that the functional engagement of the network may act as a catalyst for the anatomical development of the more sophisticated network we would expect to form during development consistent with the Interactive Specialization Hypothesis (Johnson, 2011). Moreover, estimates of connectivity were similar for the two SES-cohorts regardless of the scale at which task-based connectivity was evaluated. Thus, while there may be differences in functional connectivity at this earlier age, it was not possible to see them with our analyses. These findings are consistent with several studies that have investigated the impact of adversity on neural development, finding small or non-significant effects during the first few months of life (Pirazzoli et al., 2022; Walker et al., 2011; Xie et al., 2019).

At 24 months of age the two cohorts had different patterns of functional connectivity. In particular, for the LI cohort, increased estimates of connectivity were evident both within- and between-hemispheres. These results are only partially consistent with the resting state literature. In particular, while there was evidence for the expected increase in long-range connectivity, there was no expected decrease in short-range connectivity (Damaraju et al., 2014). Xie et al. (2019) found that higher functional connectivity was negatively associated with child growth. One possibility is that, consistent with this finding, the LI cohort has delayed synaptic

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pruning potentially reflecting malnutrition. Differences in family income may have meant that fewer resources (e.g., food) were available to the LI cohort and thus under/delayed synaptic pruning may be reflective of malnutrition. As a result, the connections within the networks may be less organized and unessential pathways may fail to be eliminated with development (Hulshoff Pol et al., 2000; Zhan et al., 2014).

Distinct from the LI cohort and the resting-state literature, the MI cohort exhibited a sharp decrease in connectivity in the right hemisphere between 6 and 24 months. Previous research has suggested that the right hemisphere is selectively involved in social and emotional processing later in development and in adulthood (Schore, 1998). Moreover, studies using the peek-a-boo task report increases in specialization with age (Lloyd-Fox et al., 2017). Thus, the decreased connectivity of the right hemisphere network involved in social processing may reflect increasing cortical specialization for social cognition (Changeux & Dehaene, 1989) or a type of pruning of this network (Homae et al., 2010).

The relation between connectivity and socioeconomic status has been found previously in the resting state fMRI literature. Poverty in school-aged children and adolescents has been linked to reduced connectivity in the default mode network, between the hippocampus and amygdala and other regions (i.e., superior frontal cortex, lingual gyrus, posterior cingulate, and putamen), and between networks that support cognition and emotion regulation (Barch et al., 2016; Brody et al., 2019; Sripada et al., 2014). Moreover, factors closely related to socioeconomic status, such as parent education and neighbourhood disadvantage, are also associated with connectivity. Language-related resting state connectivity in adolescence was positively related to parental education and reading skills in childhood (Su et al., 2021). In childhood and adolescence, community-level socioeconomic disadvantage was associated with reduced connectivity in

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systems that modulate affective processing, goal-directed behaviour and reward evaluation (Marshall et al., 2018). With respect to the development of social networks in infancy, similar to the findings from the present study, Lopera-Perez et al. (2022) found connectivity differences related to family income. Specifically, lower interhemispheric coherence was found during social compared to non-social conditions from the frontal to the central, parietal, temporal, and occipital regions for infants from higher-income, but not lower-income, families (Lopera-Perez et al., 2022).

Implications for the Development of Social Cognition

While there is strong prior evidence (see Introduction) that infants have early preferences for social stimuli, our findings show dramatic developmental changes in the neural systems that are supporting social processing throughout infancy. Thus, while these early preferences are important in showing the emergence of social cognition in infancy, the neural processes that support social cognition also show a substantial amount of developmental change. Furthermore, our findings also suggest that these developmental changes are sensitive to an infant's early life experiences.

While the analyses we conducted did not reveal notable differences between LI and MI cohorts at 6 months, the two groups exhibit virtually opposite patterns of development by 24 months of age with regards to functional connectivity. This is striking for a number of reasons including that these two groups were recruited from the same urban neighbourhood in Bangladesh. Recent work looking at the impacts of socioeconomic status on development in the U.S. have shown persistent effects of neighbourhood over even individual family socioeconomic status (Aikens & Barbarin, 2008; Chen & Paterson, 2006). Moreover, studies of the magnitude of responses in individual brain regions showed some sensitivity to early life experiences (Pirazzoli

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et al., 2022) but it was more modest than the findings reported here. Thus, a major implication of our findings is that a metric of functional connections supporting social cognition may provide a more sensitive measure of experiential and developmental changes than the magnitude of responses within individual brain regions.

Constraints on Generality

Our findings provide insights into the neural development of infants from a developing nation. Our large sample, well-established infant paradigm (i.e., peek-a-boo task) that has been used in both developed and developing nations (e.g., Lloyd-Fox et al., 2009, 2017), and use of established infant neuroimaging methodologies and pipelines (Di Lorenzo, Pirazzoli et al., 2019) lead us to expect that the results would generalize to other infants from similar populations. However, we do not have evidence that our findings will generalize to high income populations, nor do we have evidence that differences in connectivity patterns would be evident for other experimental paradigms. Similarly, when studies are conducted with high income populations, it is not clear if they will generalize to other populations. Additional research (see Future Research) is necessary to further comment on the generality of our findings. Overall, we have no reason to believe that our results depend on other characteristics of the participants, materials, or context.

In addition, given the constraints of the experimental paradigm, our analytic approach relied on the assumption that the functional networks detected are the networks that are modulated by the contrast of social and non-social stimuli and thus are reflective of social processing. However, future work can be designed to further test the specificity of these networks to social stimuli. In particular, a background connectivity approach can be applied to several blocks of social stimuli compared to baseline as well as several blocks of non-social stimuli compared to the same baseline. Moreover, future methods development will be beneficial

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to be able to investigate networks engaged in specific trials or conditions (e.g., with psychophysiological interaction [PPI] techniques; Jasińska et al., 2021) but to date, these techniques have not been successfully applied to infant fNIRS and were not attempted on the current dataset.

Future Research

Going forward, it would be useful to longitudinally investigate the development of social perception networks in a high-income sample to determine whether there is a similar longitudinal reduction in connectivity as seen in the MI cohort. Moreover, a more in-depth look at the differences between cohorts would provide insights into the driving forces behind the different developmental patterns found in this study. For example, it is possible that the cohort effect observed is mediated (or moderated) by other variables that coincide with SES (e.g., malnourishment, maternal education, inflammation).

Second, we found differences in estimates of connectivity for LI and MI cohorts at 24 months. However, with only two timepoints, spanning a large developmental period (18 months) we cannot determine if the heightened functional connectivity among infants in the LI cohort is evidence of a delay in the reduction of functional connectivity seen in the MI cohort (i.e., they would start to show similar patterns to the MI cohort at an older age) or if there are enduring differences between the cohorts. Moreover, because we did not have data between 6 and 24 months (e.g., at 12 or 18 months) we cannot conclude when the networks first began to diverge for the two cohorts.

Third, we cannot say for certain what the developmental implications are for the changes in connectivity and corresponding cognitive states. For example, it is possible that more connectivity is equated with more engagement in a task and thus increases in connectivity would

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be evidence for development of the networks. However, it is also possible that more connectivity is indicative of a less-efficient network or less specialization and thus decreases in connectivity would be evidence for the development of the networks. Thus, future work that ties changes in task-based functional connectivity to key developmental changes and milestones in social cognition (or simply behavioural measures in a social cognition task) will provide more context to both the cohort differences and the developmental changes that we report here. Moreover, future research will be beneficial in determining whether differences in connectivity between the two cohorts is predictive of non-neural outcomes, whether these differences in connectivity are specific to social processing or if they are reflective of general differences in neural network development (and thus would be seen in other networks such as attention, motor, and sensorimotor networks), and whether these differences are meaningfully related to, or predictive of, developmental outcomes.

Limitations

This study demonstrates the utility of background functional connectivity analyses in investigating the development of long-range neural networks during infancy. However, the study is not without limitations. First, both cohorts were living in impoverished environments. Thus, the effects we report, which are based on income-to-needs, may not truly capture the driving force behind the differences found (e.g., malnutrition, maternal education, inflammation). For example, other studies with infants from the BEAN project did not split the two cohorts (e.g., Bach et al., 2022; Pirazzoli et al., 2022; Xie et al., 2019). Instead, they show that continuous variables such as exposure to psychosocial adversity, malnutrition, and inflammation are associated with neural responses. Notably, however, the large differences across relatively similar groups of infants observed in the present study suggests that task-based functional

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connectivity is a sensitive measure, revealing distinct patterns of neural responses and developmental change from univariate analyses.

Second, we only measure activity in the frontotemporal regions of the brain. These are regions that have previously been found to be selectively responsive to social processing, as measured in this task. Thus, our estimates of global connectivity do not take the whole brain into account, nor can we compare the networks of regions that are selective for social perception to those that are not.

Third, the co-registration was imperfect because our atlas consisted of MRI images from U.S. infants who, on average, have larger head and brain sizes than Bangladeshi infants. At present, there is no useable set of MRI images from Bangladeshi infants.

Fourth, our data were only obtained from a social cognition task. As a result, we cannot determine whether our findings are specific to social cognitive development or whether the development of other networks may also be affected by different levels of exposure to adversity. Given that associations between socioeconomic status and resting state connectivity have been found in several networks (e.g., somatosensory, motor, frontal cognition) and that exposure to adversity, more generally, has been linked to widespread alterations in neurodevelopment (Rakesh, Kelly, et al., 2021; Rakesh, Zalesky, et al., 2021), we speculate that our findings are not specific to social cognitive development, but additional task-based connectivity research with infants from different levels of exposure to adversity is needed to draw any definitive conclusions.

Conclusion

The current study is the first to investigate longitudinal changes in task-based functional connectivity during the first two years of life. We find that at 6 months of age infants already

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have substantial long-range connections engaged during social processing, with virtually no differences between the cohorts (low- and middle- income). Our findings indicate that substantial changes occur from 6- to 24-months in the networks responsible for social perception, and we find substantial differences in connectivity between low- and middle-income populations at 24 months. More specifically, for infants in our middle-income cohort, right-hemisphere connectivity decreased over time, suggesting a process of specialization or synaptic pruning for social processing during the first two years of life. In contrast, for infants in our low-income cohort, there were increases in interhemispheric connectivity, which could signal delays in the reductions of connectivity seen in the middle-income cohort or possibly enduring differences in the networks responsible for social perception. We conclude that task-based functional connectivity is a sensitive measure that reveals developmental differences in the neural networks engaged in social perception, as well as the manner in which these networks are impacted by early life experiences.

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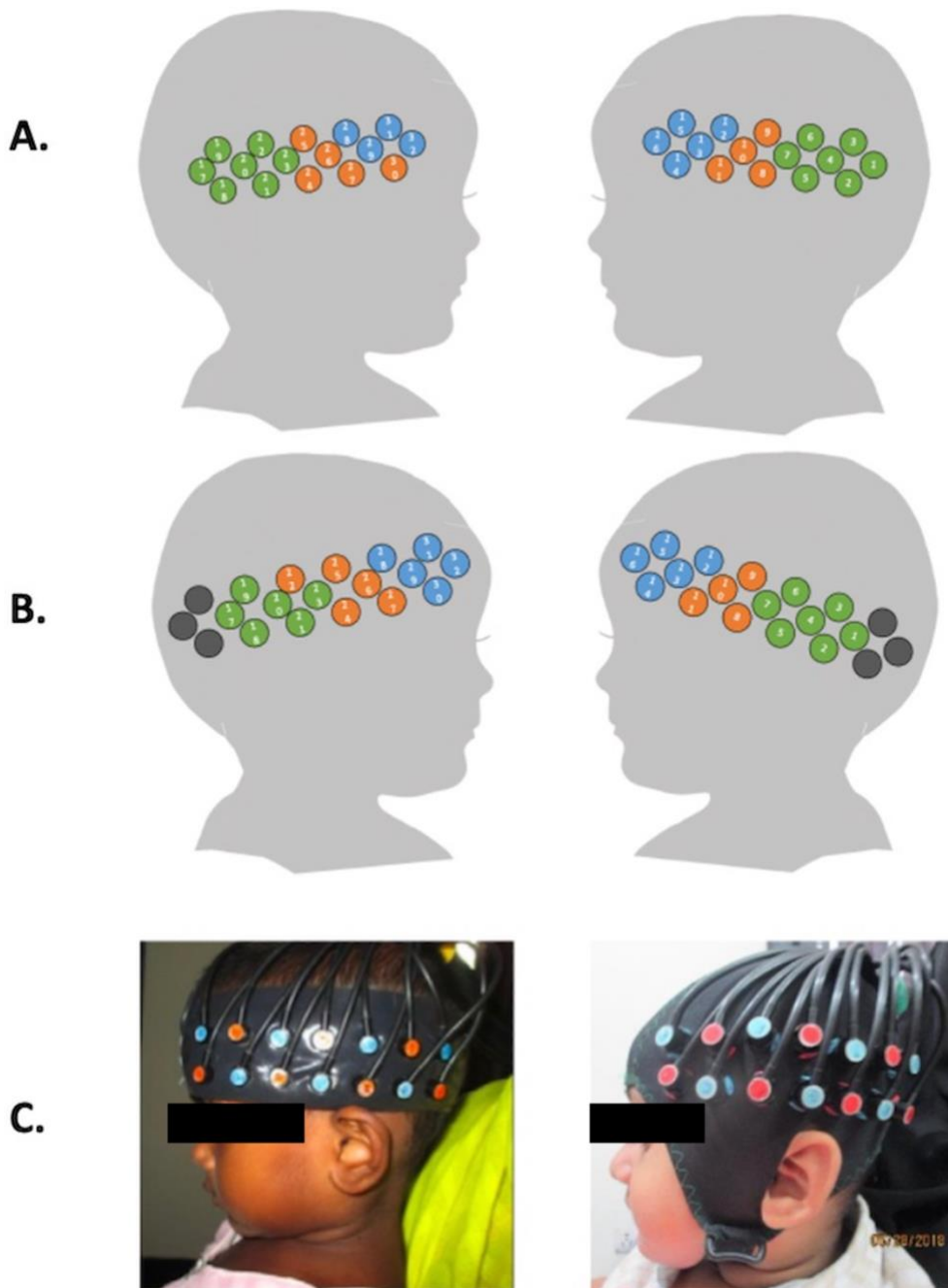
Zhan, Y., Paolicelli, R. C., Sforzini, F., Weinhard, L., Bolasco, G., Pagani, F., Vyssotski, A.

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Figure 1*Schematic View of Channel Locations at 6 and 24 months*

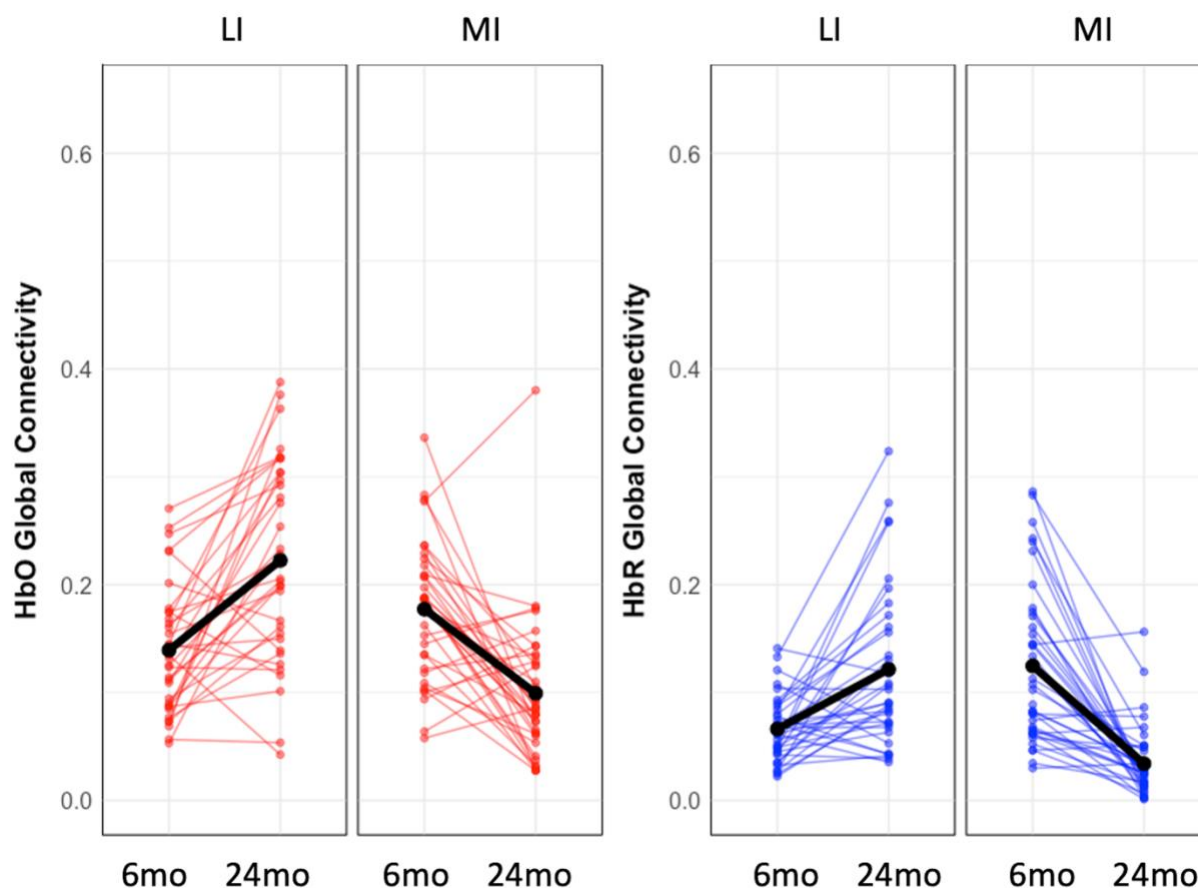
Note. Panel A) shows schematic view at 6 months: 12 sources and 12 detectors arranged into 32 channels. Average ROIs for each channel are shown in blue (IFG), orange (aMTG-STG), and green (pSTS-TPJ). Panel B) shows schematic view at 24 months: 14 sources and 14 detectors arranged into 38 channels. Six channels, shown in grey, were excluded from all analyses to

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match the configuration at 6 months. Average ROIs for each channel are shown in blue (IFG), orange (aMTG-STG), and green (pSTS-TPJ). Panel C) shows headband (left) and EasyCap (right) headgears.

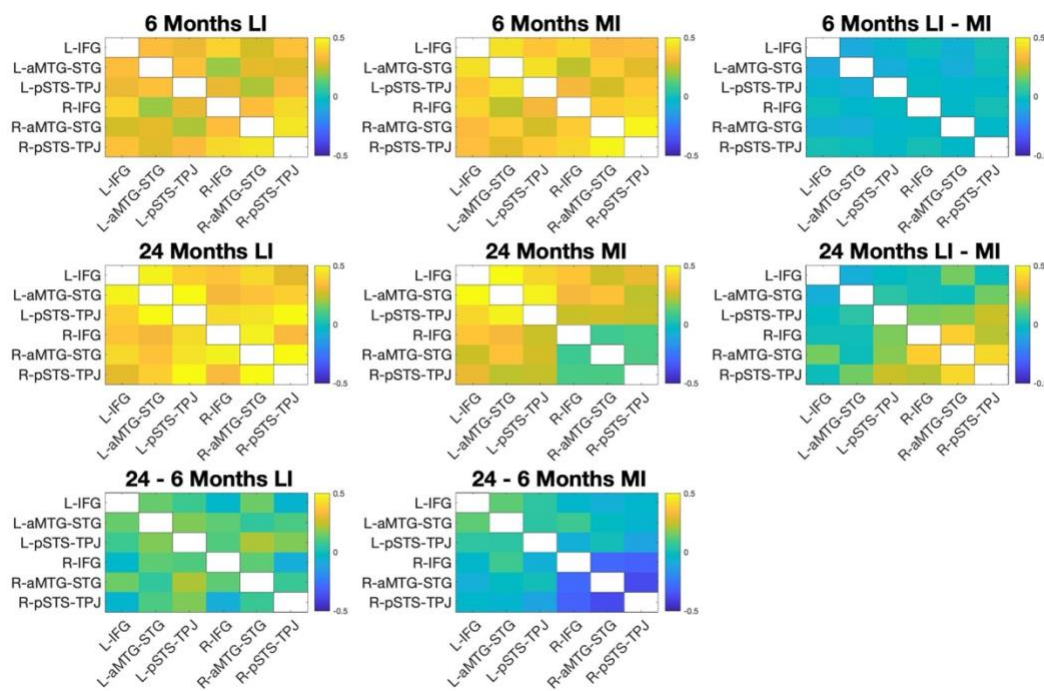
Figure 2

Estimates of Global Connectivity



Note. Spaghetti plots of functional connectivity (HbO and HbR) separated by age and cohort for infants with data at both 6 and 24 months ($n = 74$). Black dots represent means and black lines connect means at 6 months to means at 24 months. These visualizations show that, on average, for both HbO and HbR the low income (LI) and middle income (MI) cohorts had opposite patterns of global connectivity development, with the LI cohort experiencing an increase and the MI cohort experiencing a decrease in global connectivity from 6 to 24 months.

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Figure 3*Functional Connectivity for Regions of Interest*

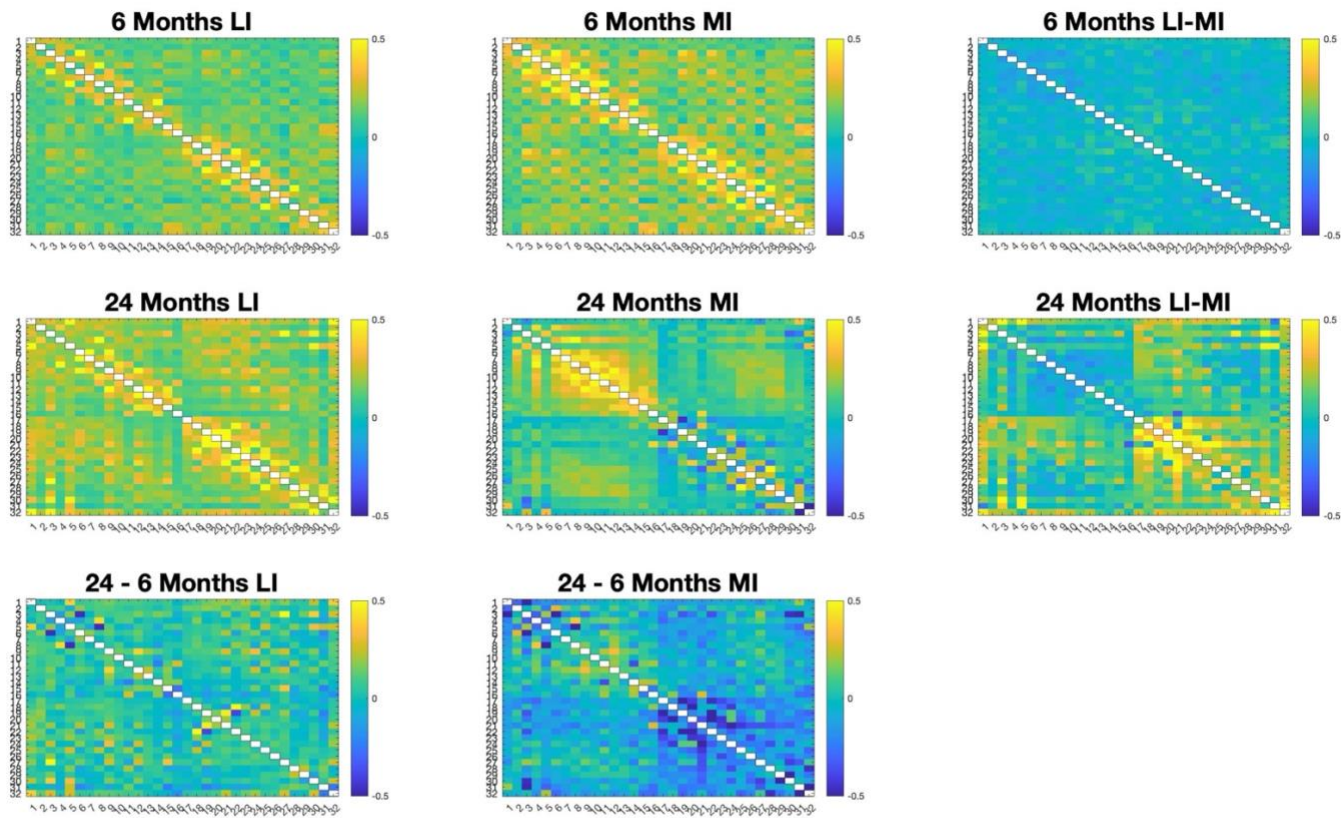
Note. Functional connectivity (HbO) between six ROIs – left and right IFG, aMTG-STG, and pSTS-TPJ. The top row shows functional connectivity estimates at 6 months of age for each cohort. The middle row shows functional connectivity estimates at 24 months of age for each cohort. Coloured bars represent correlation coefficients (ranging from -.50 to .50). For illustrative purposes, the rightmost column shows the differences in connectivity at each age between the two cohorts (i.e., LI connectivity estimates – MI connectivity estimates) and the bottom row shows the differences in connectivity from 6 to 24 months for each cohort. This figure shows that at 6 months, on average the MI cohort had slightly higher estimates of connectivity than the LI cohort both within hemispheres and between hemispheres. At 24 months, the LI cohort had slightly higher estimates of connectivity between hemispheres, whereas the MI cohort had slightly higher estimates of connectivity in the left hemisphere but much lower estimates of connectivity in the right hemisphere. These patterns suggest only the MI cohort is showing evidence of decreased connectivity at 24-months-of-age.

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Figure 4

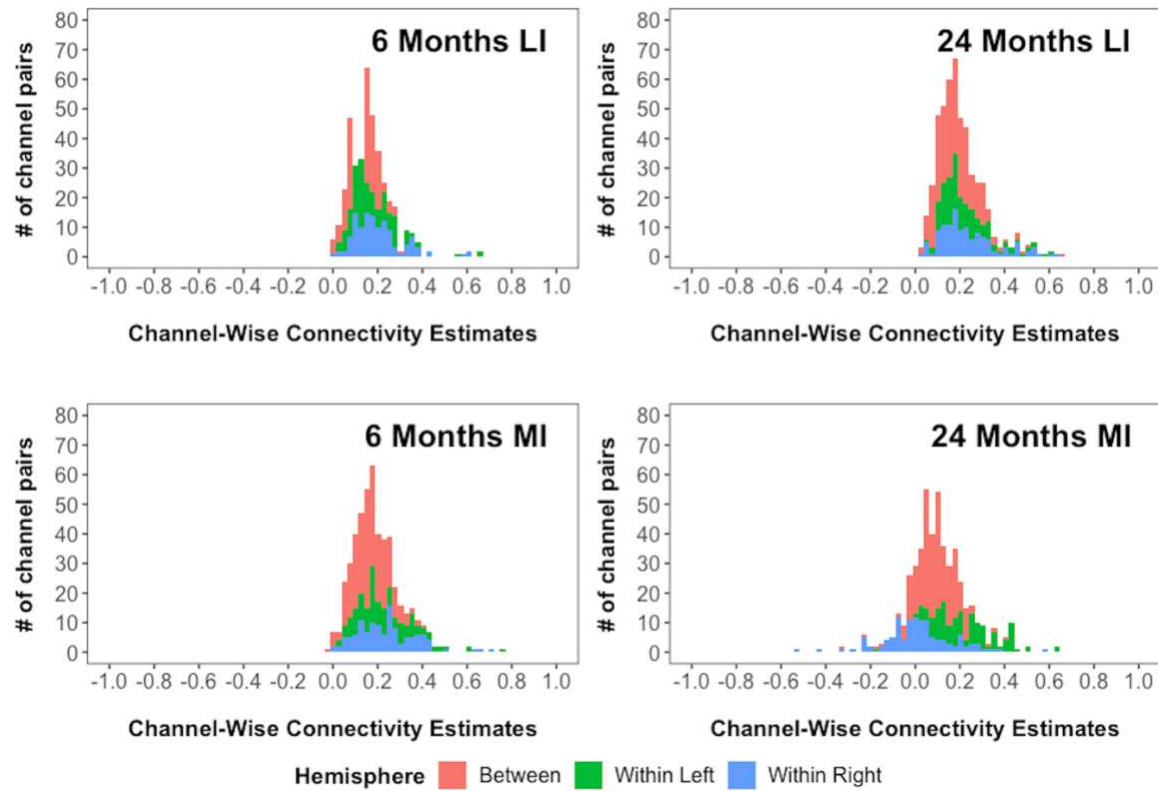
Channel-Wise Functional Connectivity

A.



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B.



Note. Panel A. Average functional connectivity between each pair of channels separated by age and cohort. Channels 1-16 are in the left hemisphere and 17-32 are in the right hemisphere. Coloured bars represent correlation coefficients (ranging from -.50 to .50).

Panel B. The distribution of connectivity estimates (correlation coefficients, r) for channel pairs with connections that were between hemisphere (red), within the left hemisphere (green), and within the right hemisphere (blue).

Supplementary Materials

Demographic Characteristics

The two cohorts were enrolled in the Bangladesh Early Adversity Neuroimaging (BEAN) project. Demographic information for these two cohorts can be found in Table S1. Manuscripts and conference presentations that have reported the fNIRS data collected through the BEAN project can be found in Appendix A.

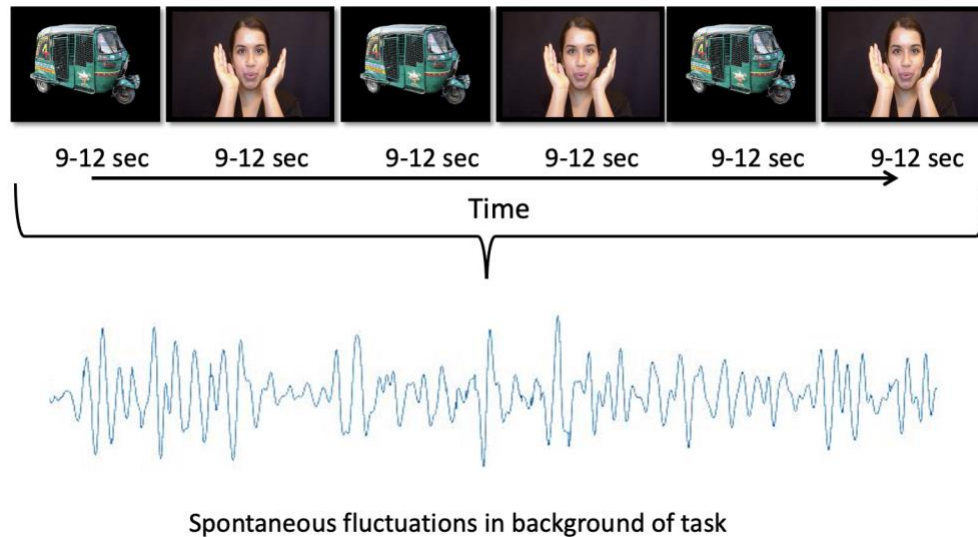
Table S1

Demographic Information for the Low- and Middle-Income Cohorts

	LI Cohort ($n = 130$, 56 males)				MI Cohort ($n = 104$, 47 males)			
	Mean	SD	Min.	Max.	Mean	SD	Min.	Max
Family monthly income (takas)	15457	10076	3000	60000	53374	19462	35000	130000
Maternal education (years)	5.59	3.59	1	16	9.74	3.05	1	16
Paternal education (years)	5.57	3.99	1	16	10.75	3.13	1	16
Exact age, 6 months	5.98	0.15	5	6	6.00	< 0.01	6	6
Exact age, 24 months	26.35	2.22	24	30	24.22	0.71	23	28

Background Functional Connectivity

A background connectivity approach was used to assess task-based functional connectivity at 6 and 24 months (see Figure S1). Three estimates of background connectivity were calculated: global background connectivity, connectivity patterns across neuro-anatomically defined regions of interest (ROIs), and connectivity patterns across channels. Below, we present the results for the deoxygenated hemoglobin (HbR) analyses.

Figure S1*Experimental Design and Background Functional Connectivity Approach*

Note. Experimental Design. Stimulus alternates between social and non-social stimuli. Auditory stimulus is either silence (VS), social sounds (AS), or non-social sounds (AN). To avoid the potential confounding effect of the temporal structure of the stimulus presentation (i.e., irrespective of content), we factored out the stimulus evoked response and kept only the residual variance around that response. By correlating this residual between cortical regions, we revealed the background connectivity that is not directly driven by the onset of the clips.

Region of Interest Functional Connectivity: HbR

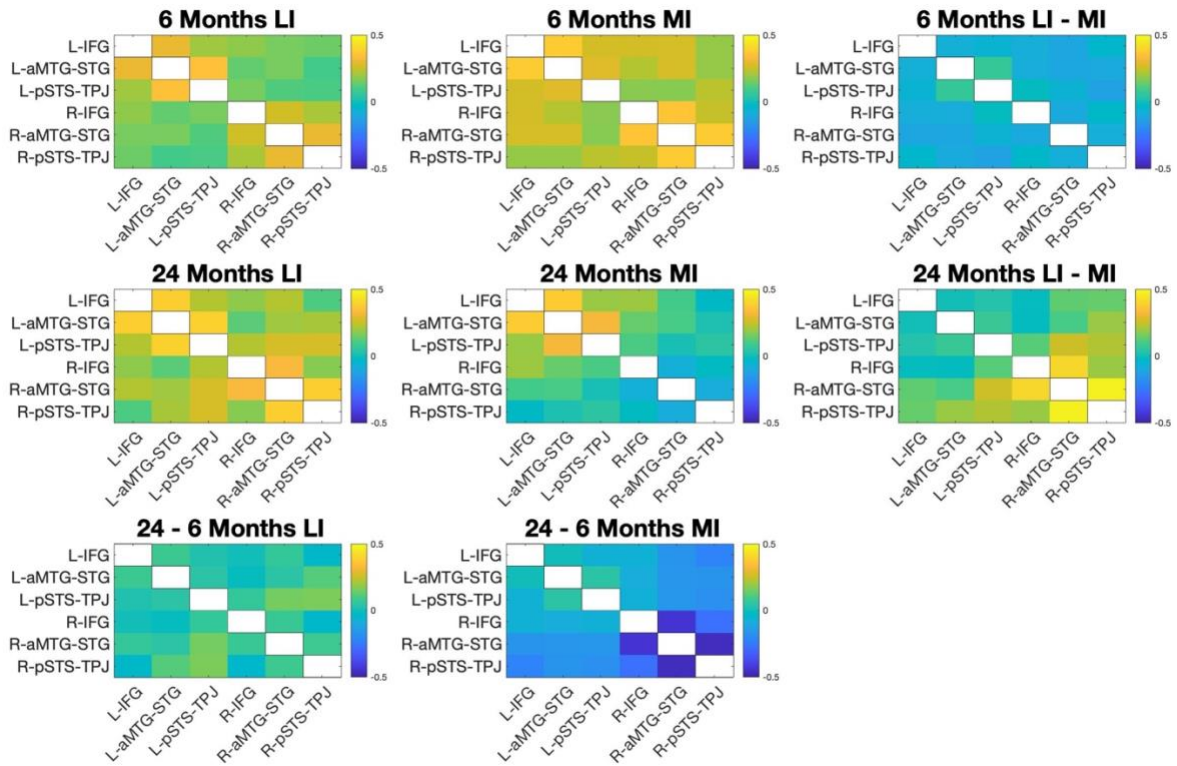
We considered 3 ROIs on each of the two hemispheres (6 ROIs total): the inferior frontal gyrus (IFG), anterior middle temporal gyrus/superior temporal gyrus (aMTG-STG), and posterior superior temporal sulcus/temporoparietal junction (pSTS-TPJ). Functional connectivity was estimated for each pair of ROIs (15 total; 6 within hemisphere; 9 between hemisphere) at 6 and 24 months for the LI and MI cohorts. Figure S2 shows the patterns of connectivity at 6 and 24 months-of-age for each cohort. *T*-tests were conducted for each pair of regions to determine if there was a significant change in functional connectivity between regions from 6 to 24-months-

of-age (see Table S2), and if there were significant differences in functional connectivity within a pair of regions between the LI and MI cohorts (see Table S3). For all analyses, we corrected for multiple comparisons using the Benjamini-Hochberg false discovery rate procedure (FDR; Benjamini & Hochberg, 1995).

For the LI cohort, of the 15 pairs of regions, there was a significant increase in connectivity for 2 pairs of ROIs (between hemispheres). For the MI cohort, there was a significant decrease in connectivity for 10 pairs of ROIs (3 within the right hemisphere, 1 within the left hemisphere, 6 between hemispheres). Comparing connectivity between cohorts at 6 months, there was significantly higher connectivity between 5 pairs of ROIs (1 within-hemisphere, 4 between hemispheres) for the MI cohort compared to the LI cohort. At 24 months, the MI cohort had significantly lower connectivity between 10 pairs of ROIs (3 within the right hemisphere, 7 between hemispheres).

Figure S2

Functional Connectivity for Regions of Interest



Note. Functional connectivity (HbR) between six ROIs – left and right inferior frontal gyrus (IFG), anterior middle temporal gyrus/superior temporal gyrus (aMTG-STG), and posterior superior temporal sulcus/temporoparietal junction (pSTS-TPJ). Coloured bars represent correlation coefficients (ranging from -.50 to .50). The top row shows functional connectivity estimates at 6 months of age for each cohort. The middle row shows functional connectivity estimates at 24 months of age for each cohort. For illustrative purposes, the rightmost column shows the differences in connectivity at each age between the two cohorts (i.e., LI connectivity estimates – MI connectivity estimates) and the bottom row shows the differences in connectivity from 6 to 24 months for each cohort.

Table S2*Connectivity (HbR) between Regions of Interest: Comparisons at 6 and 24 Months within**Cohorts*

	LI 6 vs. 24	MI 6 vs. 24
Within Hemisphere		
R-IFG, R-aMTG-STG	$t(32) = -2.01, p = .053$	$t(40) = 6.14, p < .001^*$
R-IFG, R-pSTS-TPJ	$t(33) = -0.02, p = .985$	$t(41) = 5.02, p < .001^*$
R-aMTG-STG, R-pSTS-TPJ	$t(32) = -2.31, p = .027$	$t(48) = 7.27, p < .001^*$
L-IFG, L-aMTG-STG	$t(33) = -1.08, p = .287$	$t(45) = 0.10, p = .924$
L-IFG, L-pSTS-TPJ	$t(33) = -0.24, p = .814$	$t(45) = 2.35, p = .023^*$
L-aMTG-STG, L-pSTS-TPJ	$t(33) = -1.48, p = .149$	$t(49) = -1.27, p = .211$
Between Hemisphere		
R-IFG, L-IFG	$t(33) = -0.43, p = .673$	$t(39) = 0.47, p = .642$
R-IFG, L-aMTG-STG	$t(33) = -0.80, p = .428$	$t(41) = 0.96, p = .342$
R-IFG, L-pSTS-TPJ	$t(33) = -2.58, p = .015$	$t(41) = 1.14, p = .262$
R-aMTG-STG, L-IFG	$t(32) = -2.08, p = .045$	$t(44) = 2.46, p = .018^*$
R-aMTG-STG, L-aMTG-STG	$t(32) = -1.65, p = .108$	$t(48) = 2.54, p = .014^*$
R-aMTG-STG, L-pSTS-TPJ	$t(32) = -3.43, p = .002^*$	$t(48) = 3.99, p < .001^*$
R-pSTS-TPJ, L-IFG	$t(33) = -0.65, p = .519$	$t(45) = 3.25, p = .002^*$
R-pSTS-TPJ, L-aMTG-STG	$t(33) = -2.39, p = .023$	$t(49) = 3.13, p = .003^*$
R-pSTS-TPJ, L-pSTS-TPJ	$t(33) = -2.94, p = .006^*$	$t(49) = 3.68, p < .001^*$

Note. *significant at the $p < .05$ level after Benjamini-Hochberg corrections for false detection rates

Table S3*Connectivity (HbR) between Regions of Interest: Comparisons between Low- and Middle-Income**Cohorts at 6 and 24 Months*

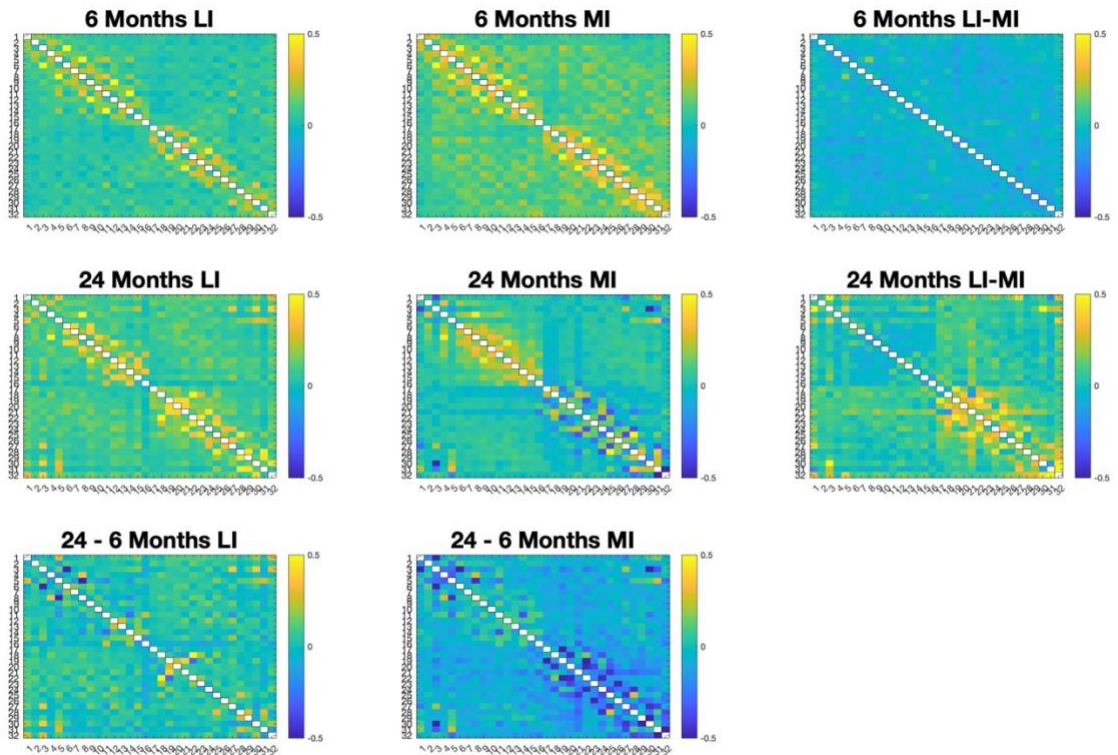
	6 Months LI vs. MI	24 Months LI vs. MI
Within Hemisphere		
R-IFG, R-aMTG-STG	$t(147) = -2.55, p = .012^*$	$t(104) = 6.55, p < .001^*$
R-IFG, R-pSTS-TPJ	$t(147) = -1.09, p = .277$	$t(106) = 3.8, p < .001^*$
R-aMTG-STG, R-pSTS-TPJ	$t(157) = -2.09, p = .038$	$t(104) = 8.92, p < .001^*$
L-IFG, L-aMTG-STG	$t(151) = -1.6, p = .111$	$t(107) = -0.16, p = .877$
L-IFG, L-pSTS-TPJ	$t(152) = -1.76, p = .081$	$t(107) = 0.39, p = .697$
L-aMTG-STG, L-pSTS-TPJ	$t(156) = 1.55, p = .123$	$t(108) = 1.24, p = .217$
Between Hemisphere		
R-IFG, L-IFG	$t(144) = -1.68, p = .095$	$t(106) = -0.51, p = .610$
R-IFG, L-aMTG-STG	$t(146) = -2.29, p = .024$	$t(107) = -0.59, p = .557$
R-IFG, L-pSTS-TPJ	$t(147) = -0.45, p = .651$	$t(107) = 2.22, p = .029^*$
R-aMTG-STG, L-IFG	$t(152) = -2.73, p = .007^*$	$t(104) = 2.49, p = .014^*$
R-aMTG-STG, L-aMTG-STG	$t(156) = -2.76, p = .006^*$	$t(105) = 2.27, p = .026^*$
R-aMTG-STG, L-pSTS-TPJ	$t(157) = -1.66, p = .100$	$t(105) = 5.96, p < .001^*$
R-pSTS-TPJ, L-IFG	$t(152) = -1.04, p = .298$	$t(106) = 3.15, p = .002^*$
R-pSTS-TPJ, L-aMTG-STG	$t(156) = -2.67, p = .008^*$	$t(107) = 4.41, p < .001^*$
R-pSTS-TPJ, L-pSTS-TPJ	$t(157) = -3.27, p = .001^*$	$T(107) = 3.82, p < .001^*$

Note. *significant at the $p < .05$ level after Benjamini-Hochberg corrections for false detection rates

Channel-Wise Functional Connectivity: HbR

All reported significant results with channel-wise functional connectivity were significant after correcting for multiple comparisons with FDR. There were 496 estimates of connectivity among the 32 channels (256 between hemispheres, 120 within the left hemisphere, 120 within the right hemisphere; see Figure S3). To compare 6 versus 24 months, *t*-tests were conducted for each pair of channels to determine if there was a significant change in functional connectivity over time. For the LI cohort, 125 channel pairs were significantly different, with 111 of those pairs showing greater connectivity at 24 months. For the MI cohort, 196 channel pairs were significantly different, with 181 of those pairs showing less connectivity at 24 months.

Finally, we compared channel-wise connectivity between the two cohorts at 6 and 24 months. At 6 months, there were 114 pairs of channels that significantly differed for HbR, with the MI cohort showing greater connectivity than the LI cohort for 113 of those pairs. At 24 months, there were 215 pairs of channels that significantly differed for HbR, with the MI cohort showing less connectivity than the LI cohort for 201 of those pairs.

Figure S3*Channel-Wise Functional Connectivity (HbR)*

Note. Average functional connectivity between each pair of channels separated by age and cohort. Coloured bars represent correlation coefficients (ranging from -.50 to .50). Channels 1-16 are in the left hemisphere and 17-32 are in the right hemisphere.

Appendix A: Manuscripts and Presentations Using the fNIRS Data from the BEAN Project***Manuscripts***

Perdue, K. L., Jensen, S. K. G., Kumar, S., Richards, J. E., Kakon, S. H., Haque, R., Petri, W. A.,

Lloyd-Fox, S., Elwell, C., & Nelson, C. A. (2019). Using functional near-infrared spectroscopy to assess social information processing in poor urban Bangladeshi infants and toddlers. *Developmental Science*, 22(5). <https://doi.org/10.1111/desc.12839>

Pirazzoli, L., Sullivan, E., Xie, W., Richards, J. E., Bulgarelli, C., Lloyd-Fox, S., Shama, T.,

Kakon, S. H., Haque, R., Petri, W. A. Jr., & Nelson, C. A. (2022). Association of psychosocial adversity and social information processing in children raised in a low-resource setting: An fNIRS study. *Developmental Cognitive Neuroscience*, 56, 101125. <https://doi.org/10.1016/j.dcn.2022.101125>

Zinszer, B. D., Pirazzoli, L., Nelson, C. A., Emberson, L., Aslin, R., & Bejjanki, V. R. (2023).

Multivariate fNIRS response patterns to social information are increasingly discriminable from six to sixty months of age [Preprint]. PsyArXiv. <https://doi.org/10.31234/osf.io/zmrkn>

Conference Presentations

Di Lonardo Burr, S. M., Mangardich, H., Pirazzoli, L., Magee, A., Hebolzheimer, A., Sanchez-

Alonso, S., Zinszer, B., Eggebrecht, A., Aslin, R., Nelson, C. A., Bejjanki, V. R., &

Emberson, L. L. (2021, October). *Increased task-based connectivity across development in a social cognition task: Links to developmental outcomes?* Presentation at the fNIRS 2021

Biennial Meeting of the Society for functional Near Infrared Spectroscopy.

- Di Lonardo Burr, S. M., Pirazzoli, L., Dopierala, A. W., Sanchez-Alonso, S., Aslin, R., Nelson, C. A., Richards, J., Bejjanki, V. R., & Emberson, L. L. (2022, October). *The development of large-scale neural networks: A longitudinal investigation of resting-state and background task-based connectivity in infancy*. Presentation at the fNIRS 2022 Biennial Meeting of the Society for functional Near Infrared Spectroscopy, Boston, Massachusetts, USA.
- Di Lonardo Burr, S. M., Pirazzoli, L., Mangardich, A., Dopierala, A. W., Sanchez-Alonso, S., Zinszer, B., Bejjanki, V. R., Nelson, C. A., & Emberson, L. L. (2022, July). *Exploring the relations between resting-state and task-based connectivity during the first two years of postnatal life*. Poster presentation at the International Conference for Infant Studies, Ottawa, Ontario, Canada.
- Pirazzoli, L., Bulgarelli, C., Sullivan, E.F., Richards, J.E., Haque, R., Petri, W.A., Nelson, C.A. (2022, October). *Impact of adversity on resting state functional connectivity in fNIRS in Bangladeshi children*. Poster presentation at the fNIRS 2022 Biennial Meeting of the Society for functional Near Infrared Spectroscopy, Boston, Massachusetts, USA.
- Pirazzoli, L., Sullivan, E.F., Xie, W., Richards, J.E., Haque, R., Petri, W.A., Nelson, C.A. (2021, October). *Association of psychosocial adversity and social information processing in urban Bangladeshi children: a fNIRS study*. Presentation at the fNIRS 2021 Biennial Meeting of the Society for functional Near Infrared Spectroscopy.
- Sullivan, E.F., Pirazzoli, L., Richards, J.E., Haque, R., Petri, W.A., Nelson, C.A. (2021, October). *The use of fNIRS in the study of statistical learning in two- and five-year-old Bangladeshi children: Associations with early adversity and language outcomes*. Poster presentation at the fNIRS 2021 Biennial Meeting of the Society for functional Near Infrared Spectroscopy.

Sullivan, E.F., Pirazzoli, L., Richards, J.E., Haque, R., Petri, W.A., Nelson, C.A. (2023, September). *Auditory statistical learning in two-year-old Bangladeshi children: An fNIRS study*. Presentation at the Fetal, Infant, and Toddler Neuroimaging Group Conference, Santa Rosa, CA.