Published in final edited form as:
Neuroimage. 2011 June 1; 56(3): 1437–1452. doi:10.1016/j.neuroimage.2011.02.073.

Functional Connectivity MRI in Infants: Exploration of the Functional Organization of the Developing Brain

Christopher D. Smyser, M.D.[a,b], **Abraham Z. Snyder, M.D., Ph.D**[a,c], and **Jeffrey J. Neil, M.D., Ph.D.**[a,b,c]

[a]Department of Neurology, Washington University, Saint Louis, Missouri

[b]Department of Pediatrics, Washington University, Saint Louis, Missouri

[c]Mallinckrodt Institute of Radiology, Washington University, Saint Louis, Missouri

Abstract

Advanced neuroimaging techniques have been increasingly applied to the study of preterm and term infants in an effort to further define the functional cerebral architecture of the developing brain. Despite improved understanding of the complex relationship between structure and function obtained through these investigations, significant questions remain regarding the nature, location, and timing of the maturational changes which occur during early development. Functional connectivity magnetic resonance imaging (fcMRI) utilizes spontaneous, low frequency (< 0.1 Hz), coherent fluctuations in blood oxygen level dependent (BOLD) signal to identify networks of functional cerebral connections. Due to the intrinsic characteristics of its image acquisition and analysis, fcMRI offers a novel neuroimaging approach well suited to investigation of infants. Recently, this methodology has been successfully applied to examine neonatal populations, defining normative patterns of large-scale neural network development in the maturing brain. The resting-state networks (RSNs) identified in these studies reflect the evolving cerebral structural architecture, presumably driven by varied genetic and environmental influences. Principal features of these investigations and their role in characterization of the tenets of neural network development during this critical developmental period are highlighted in this review. Despite these successes, optimal methods for fcMRI data acquisition and analysis for this population have not yet been defined. Further, appropriate schemes for interpretation and translation of fcMRI results remain unknown, a matter of increasing importance as functional neuroimaging findings are progressively applied in the clinical arena. Notwithstanding these concerns, fcMRI provides insight into the earliest forms of cerebral connectivity and therefore holds great promise for future neurodevelopmental investigations.

Address any correspondence to Christopher D. Smyser, M.D., Department of Neurology, Division of Pediatric Neurology, Washington University, 660 South Euclid Avenue, Campus Box 8111, Saint Louis, Missouri 63110-1093. smyserc@neuro.wustl.edu.
Christopher D. Smyser, M.D., 660 South Euclid Avenue, Campus Box 8111, Saint Louis, Missouri 63110-1093, smyserc@neuro.wustl.edu, Phone: 314.454.6120, Fax: 314.454.2523
Abraham Z. Snyder, M.D., Ph.D., 660 South Euclid Avenue, Campus Box 8225, Saint Louis, Missouri 63110-1093, avi@npg.wustl.edu
Jeffrey J. Neil, M.D., Ph.D. 660 South Euclid Avenue, Campus Box 8111, Saint Louis, Missouri 63110-1093, neil@wustl.edu

The authors declare that they have no competing financial interests or conflicts of interest.

Keywords

developmental neuroimaging; functional connectivity MRI; resting-state networks; infant; neurodevelopment

1. INTRODUCTION

Several MRI-based functional neuroimaging techniques have provided information regarding the complex interplay between cerebral structure and function that emerges during the neonatal period. Early efforts to define the functional cerebral architecture of the developing brain employed task-based functional magnetic resonance imaging (fMRI)[a] paradigms. Beginning with Born's initial description of functional activation in the occipital cortex following administration of a visual stimulus in healthy, term infants (Born, Leth et al. 1998), investigations have expanded to define anatomic localization of varied stimulus-driven cerebral functions while incorporating progressively younger populations (Born, Miranda et al. 2000; Erberich, Panigrahy et al. 2006; Heep, Scheef et al. 2009). As outlined below, results from these fMRI studies have defined functional subdivisions of the infant brain, highlighting inherent differences from older pediatric populations and demonstrating discrepancies between infants with and without cerebral injury (Born, Miranda et al. 2000; Seghier, Lazeyras et al. 2006; Seghier and Huppi 2010). While this information has afforded greater understanding of the mechanisms of normative development for specific cerebral functions, fundamental questions regarding brain development and plasticity remain unanswered. Because task-based fMRI in neonatal populations is necessarily limited, attention shifted to complementary neuroimaging techniques, including resting-state functional connectivity magnetic resonance imaging (fcMRI), using results from these inquiries as the foundation for expanded investigation.

First described in Biswal's seminal paper, fcMRI examines spontaneous, low frequency (< 0.1 Hz), coherent fluctuations in blood oxygen level dependent (BOLD) signal (Biswal, Yetkin et al. 1995; Lowe, Mock et al. 1998). These investigations reveal neural networks that collectively reflect the baseline neuronal activity of the human brain in the absence of goal-directed activity and stimulation, termed resting-state networks (RSNs) (Beckmann, DeLuca et al. 2005; Damoiseaux, Rombouts et al. 2006; Fox and Raichle 2007). Importantly, RSNs encompass regions of the brain known to be anatomically connected and co-activated by task performance (Smith, Fox et al. 2009; Zhang and Raichle 2010). fcMRI methodology has been applied in adults to characterize networks encompassing cortical and subcortical regions responsible for motor function, sensation, visual and auditory stimulus processing, memory, language, attention, and the default mode (Fox, Snyder et al. 2005; Fair, Dosenbach et al. 2007; Fox and Raichle 2007; Fair, Cohen et al. 2008; Smyser, Inder et al. 2010; Zhang and Raichle 2010) – networks previously identified through conventional neuroanatomical studies. RSNs are identifiable across varied states of consciousness and behavior (Zhang and Raichle 2010). Characterization of neural networks using fcMRI has led to greater understanding of the functional topography of the brain, aided evaluation of neuroanatomical models, and accounted for variability in task evoked fMRI responses and human behavior (Fox and Raichle 2007). Despite improved insight into functional cerebral architecture afforded by results of these investigations, the role of individual RSNs and their rational clinical application remain under investigation. Using normative findings obtained

[a]**Abbreviations:** functional magnetic resonance imaging (fMRI); functional connectivity magnetic resonance imaging (fcMRI); blood oxygen level dependent (BOLD); resting-state network (RSN); electroencephalography (EEG); diffusion tensor imaging (DTI); positron emission tomography (PET); radiofrequency (RF); seed correlation analysis (SCA); independent component analysis (ICA); post-menstrual age (PMA); diffusion weighted imaging (DWI).

from control populations as their foundation, recent inquiries have increasingly employed fcMRI to study targeted clinical populations, exploring the neurobiological mechanisms of neurological disease and neurodevelopment. Not surprisingly, fcMRI studies have also raised new research questions, particularly regarding the functional role of individual RSNs and the clinical utility of fcMRI studies.

In many regards, fcMRI is ideally suited to investigations of neonatal populations because of the intrinsic characteristics of its acquisition and analysis methods. Data can be safely acquired from subjects resting quietly or while asleep with limited need for participation. This simplifies experimental design, making consistent longitudinal studies feasible in infants (Cohen, Fair et al. 2008; Doria, Beckmann et al. 2010; Smyser, Inder et al. 2010). In addition, algorithms can be applied during analysis to minimize the impact of subject motion during acquisition, a common problem in neuroimaging investigations in this population. Further, from an acquisition lasting minutes in duration, robust information regarding global connectional properties of the developing brain can be assessed. This broadens the nature and scope of rational hypotheses, maximizing scientific potential of individual investigations. Finally, acquisition and analysis methods are readily applicable to infants with and without cerebral injury, making comparison between populations practical. This combination affords enormous potential for large-scale explorations of plasticity and recovery of the developing brain, defining the impact of genetic and environmental influences on the functional cerebral architecture (Van Dijk, Hedden et al. 2010).

To date, a limited number of investigations have applied fcMRI to the study of preterm and term infants (Fransson, Skiold et al. 2007; Lin, Zhu et al. 2008; Liu, Flax et al. 2008; Fransson, Skiold et al. 2009; Gao, Zhu et al. 2009; Damaraju, Phillips et al. 2010; Doria, Beckmann et al. 2010; Fransson, Aden et al. 2010; Smyser, Inder et al. 2010). Results of these inquiries have provided information regarding the earliest forms of cerebral connectivity in the developing brain, highlighting the intricacy of the relationship between cerebral structure and function throughout the neonatal period and providing the foundation for further application of these methods to future neurodevelopmental investigations. Despite these advances, methodological questions remain unanswered regarding appropriate mechanisms for acquisition, analysis, and interpretation of fcMRI data in this population. This review focuses upon the emerging body of literature pertinent to application of fcMRI as a neurodevelopmental tool used to query neonatal populations. It begins with an exploration of the biological basis for early RSNs identified using fcMRI. Next, we highlight technical issues pertinent to application of fcMRI in this population, including selection of RF coil and acquisition parameters, level of arousal during acquisition, atlas registration, preprocessing methods, and analysis approach. The central findings of prior investigations of infants using fcMRI and their acquisition and analysis methods are then detailed. We next review results from prior investigations, examining the relationship between anatomical and functional architecture and the role of neural plasticity and brain injury in neural network development. Finally, potential opportunities for future clinical application of fcMRI in this population are reviewed, highlighting avenues for targeted investigation.

2. BIOLOGICAL DEVELOPMENT OF NEURAL NETWORKS

The structure and function of the maturing brain change continuously throughout early development (Tau and Peterson 2010). Formed upon an evolving cerebral architecture, neural networks arise and mature through a complex set of anatomical and functional interactions, shaped by the interplay of genetics, environmental exposure, and experience. Ongoing modulation and enhancement of early neuronal connections produce functional associations with diverse configuration and performance (Penn and Shatz 1999; Le Be and

Markram 2006; Vincent, Patel et al. 2007; Damoiseaux and Greicius 2009). The result is dynamic, temporally- and topographically-specific clusters of neurons which mediate the brain's functional capacities (Tau and Peterson 2010). Application of fcMRI methods during this period enables identification and characterization of the earliest versions of these neural networks, defining the role of the emerging cerebral structure in their longitudinal development.

2.1 Early Cerebral Connections

The structural foundation critical to neural network development is established early in gestation. Neuronal precursors emerge and proliferate within the ventricular zone beginning at 5–6 weeks gestation (Levitt 2003; Ghashghaei, Lai et al. 2007; Bystron, Blakemore et al. 2008). Differentiation and migration ensues, peaking between 12–20 weeks and finishing by 26–29 weeks gestation (Gressens 2000; Chao, Ma et al. 2009). Following migration, neurons establish connections *via* extension of axons and dendrites. These early synaptic connections are identifiable as early as 5 weeks gestation, and undergo continuous modification and refinement, influenced by surrounding synaptic and neuronal activity (Marin-Padilla 1971; Stewart and Pearlman 1987; Super, Soriano et al. 1998). Preplate neurons are the initial targets for projections from the thalamus and brainstem, forming the earliest cortical circuits (Chao, Ma et al. 2009). The subplate, an increasingly prominent transient layer within the preplate, serves as a holding area for afferent fibers to the cortex, and is among the first to exhibit regional variability (Kostovic and Rakic 1990; Allendoerfer and Shatz 1994; Bystron, Blakemore et al. 2008; Tau and Peterson 2010). Associations in this region give rise to more permanent connections within the cortical plate beginning at 20 weeks gestation and continuing through the perinatal period (Kostovic and Rakic 1990).

2.2 Cortical Development

The cortical plate begins to form as early as 7–8 weeks gestation, with migrating neurons dividing the preplate into subplate and marginal zone layers (Figure 1) (Super, Soriano et al. 1998; Bystron, Blakemore et al. 2008). Development of the cortical plate precedes the appearance of more mature cerebral connections. Transient circuitry incorporating the subplate and cortical plate form the foundation for electrophysiological phenomenon detectable during this period (Kostovic and Jovanov-Milosevic 2006). Establishment of callosal and long-range cortical connections follows, with dissolution of the subplate around 33–34 weeks gestation (Kostovic and Jovanov-Milosevic 2006). Lamination occurs at varied rates depending on layer and region (Kostovic, Judas et al. 1995). Differences in cell type, morphology, and layer correspond to the pattern of local and distant projections each cell sends and receives, with individual layers developing characteristic patterns of connections (Tau and Peterson 2010).

Synaptic density within the cortical plate increases rapidly until 26–28 weeks gestation. This is followed by thickening of the developing cortex as dendritic arborization and synaptogenesis accelerate in the third trimester. Anatomic distance plays a significant role in rate of cortical connection development, with shorter connections preceding those between more physically distant regions. Cortical gyri and sulci become increasingly identifiable during this period (Garel, Chantrel et al. 2001; Huisman, Martin et al. 2002). This expansion is offset by apoptosis of neurons and their connections, with selective elimination of weaker associations (Le Be and Markram 2006). The timing of synaptogenesis and synapse elimination differs across layers and regions, with consistent neuronal activity gaining increasing importance in molding cortical architecture and influencing survival of neural circuits (Huttenlocher 1979; Bourgeois, Jastreboff et al. 1989; Huttenlocher 1990; Chugani 1998; Le Be and Markram 2006). During this period, spontaneous neuronal firing develops complex spatial and temporal patterns independent of external input, representing a process

of ‘integration through synchronization’, shaping connection development. Additionally, regional co-activation in response to sensory-driven activity, cell-cell contact, trophic factors, and environmental stimuli affects neuronal activity, molding cortical architecture and influencing survival of neural circuits, reflecting a Hebbian process of functional connection modulation (Ehrlich and Malinow 2004; Citri and Malenka 2008; Fair, Cohen et al. 2008; Morishita and Hensch 2008; Honey, Sporns et al. 2009; Tau and Peterson 2010).

2.3 Thalamocortical Connections

Thalamic afferents emerge between 12 and 16 weeks gestation, extending to and spreading within the cortical subplate (Lagercrantz and Changeux 2009). These connections become prominent with advancing gestational age, accumulating in superficial portions of the subplate zone between 20 and 23 weeks gestation, followed by regionally-specific extension of thalamocortical axons into the cortical plate from 24 to 32 weeks gestation (Kostovic and Jovanov-Milosevic 2006). This is followed by development of more permanent connections within the cortical plate through synaptic refinement. These connections increase in number and strength through the perinatal period. During this period, there is co-existence of transient endogenous (subplate) and permanent (thalamocortical) sensory-driven circuitry within the immature cortex.

2.4 Myelination

Glial cells, including oligodendrocytes, astrocytes, and microglia, populate the developing central nervous system and perform vital neuronal support functions. Myelination begins between 20 and 28 weeks gestation, enhancing efficiency and dependability of axonal propagation of action potentials. This process proceeds rapidly during the first year of life, continuing in a hierarchical manner through childhood, demonstrating a pattern of regional variability and contributing to development and sustainability of functional connections (Huppi, Maier et al. 1998; Fair, Dosenbach et al. 2007; Cohen, Fair et al. 2008; Fair, Cohen et al. 2008; Tau and Peterson 2010).

2.5 Postnatal Cerebral Development

Progressive structural and functional development continues at a rapid pace throughout early postnatal life. Following birth, cortico-cortical fibers within the subplate remnant connect with postsynaptic cortical targets, facilitating elaboration of short cortico-cortical fibers (Kostovic and Rakic 1990; Huttenlocher and Dabholkar 1997; Kostovic and Jovanov-Milosevic 2006). Subplate neurons undergo gradual apoptosis, leaving behind endogenous circuitry and cortico-cortical connections within superficial cortical layers (Kostovic and Jovanov-Milosevic 2006). The cortex grows and develops secondary to elaboration of existing dendrites, spines, and synapses. Pruning also becomes increasingly active postnatally. Patterns of synaptic remodeling remain heavily influenced by spontaneous and evoked neuronal activity, reorganizing and modulating neural circuits (Ehrlich and Malinow 2004; Citri and Malenka 2008; Morishita and Hensch 2008; Tau and Peterson 2010). These changes provide a new anatomical substrate for evolving electrophysiological activity. Subsequently, regionally specific cerebral development continues, initially with prominence in the sensorimotor and visual cortices (Huttenlocher and Dabholkar 1997). These processes continue into childhood and early adulthood.

2.6 Identification of Early Functional Connections

Remodeling of synaptic connections in response to endogenous and sensory-driven neural input plays an important role in the early functional organization of cortical circuits (Tau and Peterson 2010). This remodeling continues throughout early development, with refinement of local connections and development of complex longer-range connections. The

dramatic, ephemeral changes in structure and function produce characteristic physiological phenomena that evolve parallel to anatomical development and are detectable *via* a variety of neurodiagnostic techniques (Kostovic and Jovanov-Milosevic 2006). These include electroencephalography (EEG), diffusion tensor imaging (DTI), task-related functional MRI (fMRI), and positron emission tomography (PET). Multiple investigations have demonstrated the evolution of electrical activity in the infant brain as measured by EEG mirrors structural development, with regional variability and increasingly synchronous activity between bilateral homologous regions (Niedermeyer 2005; Vanhatalo and Kaila 2006). DTI studies have demonstrated a process of gradual cerebral development, with variability in rate and pattern between white and gray matter and among functionally specific neuronal pathways (Neil, Miller et al. 2002; Partridge, Mukherjee et al. 2004; Huppi and Dubois 2006). Varied stimulus types used in task-related fMRI elicit regionally-specific evoked cerebral activation in term and preterm infants (Born, Miranda et al. 2000; Erberich, Panigrahy et al. 2006; Heep, Scheef et al. 2009). Finally, a limited number of PET studies have demonstrated regionally variable patterns of metabolic activity in the developing brain (though potential for continued investigation using this modality is uncertain secondary to risks associated with exposure to ionizing radiation) (Chugani, Phelps et al. 1987; Chugani 1998). It is likely that connections unique to the developing brain – including transient connections to the subplate and those whose firing is endogenously driven – are detectable by fcMRI. Thus, application of fcMRI enables *in vivo* assessment of the earliest forms of functional connectivity, complementing prior efforts and providing a more complete understanding of the interrelationship between structure and function.

3. TECHNICAL ASPECTS

Sensitivity of functional MRI is inherently limited by low signal-to-noise ratio and compromised by sources of spurious variance, both subject and equipment dependent. Application of functional MRI techniques to neonatal populations provides unique technical challenges, confounded by the evolving milieu of infant neurophysiology (Erberich, Friedlich et al. 2003). One of these challenges is ensuring patient safety. The safety of performing MRI investigations in infants has been increasingly established, including at higher field strengths (Schenck 2005; Dagia and Ditchfield 2008; Merchant, Groves et al. 2009; Benavente-Fernandez, Lubian-Lopez et al. 2010). Emerging from these inquiries, new technology and modified acquisition practices have been applied in an effort to obtain high quality images while ensuring patient safety (Erberich, Friedlich et al. 2003; Bluml, Friedlich et al. 2004; Barkovich 2006; Mathur, Neil et al. 2008; Merchant, Groves et al. 2009; van Wezel-Meijler, Leijser et al. 2009). Other challenges are related to data analysis. Advanced techniques to improve anatomic registration, reduce signal change present due to non-neuronal causes, and identify RSNs have been developed. These measures have significantly improved the quality of results obtained in functional investigations of infants. Despite these advances, questions remain regarding optimal methods for fcMRI image acquisition and analysis. These factors must be considered when planning future studies or evaluating results obtained from investigations of functional connectivity in neonates.

3.1 Image Acquisition

3.1.1 Radiofrequency Coil—As with all MR studies, shape and size of radiofrequency (RF) coils utilized for data acquisition impacts image quality. Early functional MRI studies of neonates utilized adult head, surface, or knee RF coils (Yamada, Sadato et al. 1997; Born, Leth et al. 1998). This yielded relatively low signal-to-noise ratio and, consequently, limited spatial resolution (Souweidane, Kim et al. 1999). Since that time, RF coils dedicated to neonatal brain imaging have become commercially available. These age-specific coils improve signal-to-noise by as much as 2–3 times (Erberich, Friedlich et al. 2003; Bluml,

Friedlich et al. 2004). Further, these coils can be integrated into MR-compatible incubator systems (Erberich, Friedlich et al. 2003; Barkovich 2006). These systems accommodate life-sustaining and monitoring equipment, enabling study of progressively younger populations, including very preterm infants (born less than 30 weeks gestation), while minimizing clinical risk and signal artifact. They provide a stable thermal environment, minimizing physiological responses which alter cerebral blood flow. Duration of scanning is also reduced due to improved subject tolerance of acquisition (*i.e.*, reduced need for repeat acquisition secondary to movement artifact), limiting procedural risk (Barkovich 2006; Seghier, Lazeyras et al. 2006). Utilization of age-specific coils with integrated systems has become standard in this population and is necessary for optimal fcMRI image quality.

3.1.2 Acquisition Parameters—MRI acquisition parameters also significantly affect image quality. Recently published investigations of functional connectivity have included noteworthy differences in pulse sequence timing, slice thickness and in-plane resolution (Beckmann, DeLuca et al. 2005; Fox, Snyder et al. 2005; Fransson, Skiold et al. 2007; Liu, Flax et al. 2008; Long, Zuo et al. 2008; Thomason, Chang et al. 2008). In addition, methods by which appropriate acquisition parameters were identified have varied (Fransson, Skiold et al. 2007; Liu, Flax et al. 2008; Van Dijk, Hedden et al. 2010). Nevertheless, recent studies demonstrated consistent identification of RSNs in adult subjects studied with varied run structure, temporal resolution, and spatial resolution, suggesting the basic phenomenology is robust (Lowe, Mock et al. 1998; Van Dijk, Hedden et al. 2010). It has been suggested that, in adults, resting-state acquisitions as short as 4–5 minutes may enable identification of RSNs, with limited benefit from extension beyond 6 minutes. Concatenation of multiple brief runs produced results comparable to those obtained with a single continuous run of identical duration (Van Dijk, Hedden et al. 2010). However, it is increasingly apparent that RSNs exhibit non-stationarity (*i.e.*, dynamic changes within the time scales of seconds to minutes) (Chang and Glover 2010). Hence, optimal or even minimal acquisition time in adults remains uncertain (Anderson, Druzgal et al. 2010).

It is unclear whether these results can be extrapolated to younger populations. Optimal parameters for structural image acquisition are known to be a function of subject age (Jones, Palasis et al. 2004; Rivkin, Wolraich et al. 2004). However, there has been no systematic study of optimal functional image acquisition parameters in neonates. This is of critical importance because significant changes in cerebral volume and structure occur during early development. In particular, the relationship between brain size, which increases rapidly during development, and image spatial resolution has not been examined. The impact of acquisition duration, including age-specific effects of non-stationarity, also remains unexplored. In the only evaluation of acquisition parameters in infants currently available, Lin demonstrated comparable functional activation in the sensorimotor cortex with varied TR times (Lin, Zhu et al. 2008) (Figure 2). Further investigation of the impact of these variables on fcMRI studies of preterm and term infants is required.

3.1.3 Level of Arousal—Recent investigations established the feasibility of performing useful functional neuroimaging in subjects at varied levels of arousal, a factor which imparts inherent benefits to the study of infants. RSNs incorporating higher order functional systems and primary sensory systems have been identified in pediatric and adult subjects during different levels of sleep, under anesthesia, and in coma, indicating correlations are not obliterated by these conditions (Kiviniemi, Jauhiainen et al. 2000; Kiviniemi, Haanpaa et al. 2005; Fukunaga, Horovitz et al. 2006; Redcay, Kennedy et al. 2007; Boly, Phillips et al. 2008; Greicius, Kiviniemi et al. 2008; Horovitz, Fukunaga et al. 2008; Horovitz, Braun et al. 2009; Larson-Prior, Zempel et al. 2009; Martuzzi, Ramani et al. 2010; Mhuircheartaigh, Rosenorn-Lanng et al. 2010). However, effects of level of arousal on neural network detection remains incompletely understood, with correlation strength modulated in a

network specific manner by state (Greicius, Kiviniemi et al. 2008; Horovitz, Braun et al. 2009; Mhuircheartaigh, Rosenorn-Lanng et al. 2010). Prior studies of functional connectivity in infants have included results obtained from subjects studied both with (Fransson, Skiold et al. 2007; Damaraju, Phillips et al. 2010; Doria, Beckmann et al. 2010) and without (Lin, Zhu et al. 2008; Liu, Flax et al. 2008; Fransson, Skiold et al. 2009; Gao, Zhu et al. 2009; Damaraju, Phillips et al. 2010; Doria, Beckmann et al. 2010; Fransson, Aden et al. 2010; Smyser, Inder et al. 2010) light sedation. In those performed without sedation, subjects are assumed to be studied during natural sleep or resting quietly. In some studies, comparable results were obtained for subjects with and without sedation (Fransson, Skiold et al. 2009; Damaraju, Phillips et al. 2010; Doria, Beckmann et al. 2010), suggesting findings from older subjects may translate to this population.

The ability to study infants who are awake is typically precluded by patient motion during acquisition, with reports of neonates studied while awake particularly limited (Dehaene-Lambertz, Dehaene et al. 2002). Studies of infants who awaken during acquisition are frequently corrupted by motion artifact (see Section 3.2.2). Additionally, the ability of investigators to dictate subject state is difficult in this population, a factor which may be of increasing value in resting-state investigations (Van Dijk, Hedden et al. 2010). These factors underscore the importance of appropriate monitoring of subject state throughout acquisition in neonates (*e.g.*, clinical observation, EEG monitoring, etc.). They also emphasize the need for further study of the effects of natural or induced changes in level of arousal, including the impact of sleep stage, on detection of RSNs in this age group.

3.2 Analysis

3.2.1 Image Registration—Spatial normalization to standardized stereotactic space is necessary to provide a common framework for statistical comparison of inter- and intra-individual functional neuroimaging results due to variability in individual cerebral morphology (Wilke, Schmithorst et al. 2002; Seghier, Lazeyras et al. 2006; Kazemi, Moghaddam et al. 2007; Altaye, Holland et al. 2008). While comparability of functional data obtained from pediatric subjects following transformation to common stereotactic space has been validated (Burgund, Kang et al. 2002; Kang, Burgund et al. 2003), utilization of adult templates for this procedure in pediatric populations introduces increased variability and error due to age-specific differences in cerebral volume and structure (Muzik, Chugani et al. 2000; Burgund, Kang et al. 2002; Hoeksma, Kenemans et al. 2005; Altaye, Holland et al. 2008). This led to creation of age-specific atlases. Use of these targets consistently demonstrated better results for registration of gray matter, white matter, and cerebrospinal fluid (Wilke, Schmithorst et al. 2002; Kazemi, Moghaddam et al. 2007; Altaye, Holland et al. 2008). For infants, this procedure is further complicated by variability in morphology associated with normative development. Rapid, regional specific changes in cerebral volume, cortical density and thickness, and gyration can all appreciably affect image registration and normalization, interfering with identification of functional correlations (Gaillard, Grandin et al. 2001; Seghier, Lazeyras et al. 2006; Kazemi, Moghaddam et al. 2007). These differences have necessitated development of targets specific not only to infants, but to relatively narrow periods of gestation (*i.e.*, every 2–4 weeks post-menstrual age [PMA]) (Figure 3). Utilization of focused targets improves image normalization and registration (Kazemi, Moghaddam et al. 2007; Damaraju, Phillips et al. 2010; Doria, Beckmann et al. 2010; Smyser, Inder et al. 2010), making their use fundamental to accurate analysis of functional neuroimaging data for preterm and term infants. Increasing numbers of atlases are now readily accessible (Dehaene-Lambertz, Dehaene et al. 2002; Prastawa, Gilmore et al. 2005; Kazemi, Moghaddam et al. 2007; Altaye, Holland et al. 2008), however, many targets have been developed using limited numbers of subjects. Further development of freely available, standardized gestational age-specific targets from greater

numbers of subjects is necessary to ensure accuracy of results and comparability across studies.

3.2.2 Movement—Despite institution of methods designed to limit head movement during acquisition, motion artifact remains a pervasive problem in functional investigations of neonates (Fransson, Skiold et al. 2007; Lin, Zhu et al. 2008; Liu, Flax et al. 2008; Gao, Zhu et al. 2009; Smyser, Inder et al. 2010). Patient motion during acquisition results in translation and rotation of the head in the field of view. This can be corrected by spatial resampling (*i.e.*, image realignment). However, patient motion also induces major artifactual changes in reconstructed image intensity ("spin history" effects) that cannot be remedied by spatial resampling. Prospective motion correction does not rectify "spin history" artifact (Thesen, Heid et al. 2000). Thus, exclusion of epochs corrupted by extreme head motion is currently the only effective method to address this problem. Consistent with what has been documented in older pediatric populations, head motion during individual studies is typically self-limited and intermittent, but is often extreme (O'Shaughnessy, Berl et al. 2008). As reported previously, this occurs most prominently in awake subjects.

Methods to detect and/or account for signal variation induced by head motion differ. Preprocessing measures typically include movement analysis using rigid body realignment to correct for head motion within and across runs (see Section 3.2.4). This strategy effectively deals with slow drifts in head position. Utilizing the results derived from head motion correction, some investigations use the presence of prominent motion as the basis for exclusion of the entire data set from analysis. An alternative is based upon results demonstrating that concatenation of non-contiguous epochs produces fcMRI results comparable to those from continuous collection (Fair, Schlaggar et al. 2007; Van Dijk, Hedden et al. 2010). Adapting this concept, volumes containing extreme motion are excluded from fcMRI analysis, with the remaining data treated as continuous (Fransson, Skiold et al. 2007; Smyser, Inder et al. 2010) (Figure 4). In this approach, a minimum amount of utilizable data is designated as the criterion for inclusion in subsequent analyses, with studies below this threshold excluded. This can permit inclusion of a higher proportion of data sets. Though each measure may limit performance of within subject analyses, they are critical to minimizing the influence of motion artifact. The impact of motion can be further reduced *via* inclusion of parameters computed during rigid body head motion correction as nuisance regressors during fcMRI preprocessing (see Section 3.2.4), however, such regression does not completely eliminate "spin history" artifact. Because sedation as a means of limiting movement is not an option at many institutions, further refinement of acquisition techniques would be helpful. In addition, improved analysis methods are necessary to minimize the effects of motion.

3.2.3 Frequency Characteristics—Beginning with Biswal's landmark observation of correlated functional activation at frequencies less than 0.08 Hz (Biswal, Yetkin et al. 1995), exploration of RSNs has centered on low frequency bands. While initial investigations demonstrated low-frequency BOLD fluctuations resulted from neuronal activity rather than physiological artifact (Lowe, Mock et al. 1998; Cordes, Haughton et al. 2001; Kiviniemi, Ruohonen et al. 2005; De Luca, Beckmann et al. 2006; Wu, Gu et al. 2008), subsequent studies illustrated functional connections exhibit increasing power at lower frequencies and further defined spectral features of specific RSNs (Cordes, Haughton et al. 2001; Achard, Salvador et al. 2006; Fox and Raichle 2007; Salvador, Martinez et al. 2008; Wu, Gu et al. 2008). Connection distance was found to play an important role in frequency specificity, with long distance connections appearing more frequency specific (peaking at lower frequencies) than shorter connections. This effect was most prominent for cortically based neural networks of interest in neonates (Wu, Gu et al. 2008). However, it remains incompletely understood whether fast frequency spectral characteristics represent true

physiology or the effect of movement artifact, which artificially inflates spurious short-range correlations through corruption of BOLD signal.

While these investigations included only adult subjects, their results have been generalized to pediatric populations. All previous investigations of infants included low pass filters to isolate signal components relevant to fcMRI, conventionally frequencies below ~0.1 Hz. However, selection of appropriate frequency limits must account for population-specific physiologic noise. Baseline cardiac and respiratory rates are typically faster in infants than in older populations, and there may be greater variability in tidal volume and heart rate. This suggests that resting-state investigations in neonates may be especially susceptible to incomplete modeling of non-neuronal signal. While the contribution of these variables in older subjects was previously determined to be minor (Cordes, Haughton et al. 2001), recent results suggest modeling of these physiological confounds can reduce their impact in measured results (Birn, Diamond et al. 2006; Birn, Smith et al. 2008; Chang, Cunningham et al. 2009). Although model-based strategies for reducing noise introduced by cardiac and respiratory pulsations are used routinely in some laboratories (*e.g.*, RETROICOR) (Glover, Li et al. 2000), their efficacy is not universally appreciated. Application of these methods in neonatal populations has not yet been attempted. Additionally, compensation for respiratory rate induced changes in pCO_2, which is known to induce profound effects on the BOLD signal in adults (Kastrup, Kruger et al. 1999; Chang and Glover 2009), also remains uninvestigated. Physiological data, including cardiac and respiratory signals, have not previously been routinely acquired during fcMRI in infants. Identification of age-specific artifact reduction strategies in this population will require systemized collection of physiologic data across all age groups. While currently employed retrospective preprocessing measures potentially reduce non-neuronal noise (see Section 3.2.4), refinement of frequency selection in infants and modeling of age-specific sources of physiologic noise would be helpful, particularly as investigations progress to examine subcortical regions.

3.2.4 Data Preprocessing—Fluctuations in fcMRI signal due to non-neuronal causes originate from multiple sources, including scanner artifact, physiological noise, and subject motion (Biswal, Yetkin et al. 1995; Lowe, Mock et al. 1998; Uddin, Supekar et al. 2010; Van Dijk, Hedden et al. 2010). Their impact must be recognized and corrected, as any global effect can artificially inflate or mask apparent correlations between regions. These variables are presumed independent of subject age, and measures applied to account for them in investigations of infants are consistent with those in older populations. Standard preprocessing measures account for signal variation intrinsic to acquisition and correct for head motion within and across runs. Some laboratories also correct for asynchronous slice timing, but the necessity of doing so has not been established. Spatial smoothing increases the signal to noise ratio (Uddin, Supekar et al. 2010; Van Dijk, Hedden et al. 2010). Additional measures minimize sources of physiological artifact not eliminated *via* temporal filtering, including inclusion of regressors for white matter, cerebrospinal fluid, whole brain signal, and rigid body parameters derived from head motion correction in linear regression modeling. White matter and cerebrospinal fluid regions are presumed to include high proportions of noise related to cardiac and respiratory signals, and addition of these nuisance regressors minimizes their effects (Dagli, Ingeholm et al. 1999; Lund, Madsen et al. 2006; de Munck, Goncalves et al. 2008; Van Dijk, Hedden et al. 2010). Use of whole brain signal – the time course of the average signal intensity within the brain – improves correlation specificity and reduces noise (Fox, Zhang et al. 2009; Weissenbacher, Kasess et al. 2009). Further, some studies suggest global signal correlates with the effects of pCO_2 variation, so its inclusion potentially minimizes the effects of this central variable (Wise, Ide et al. 2004; Chang and Glover 2009). Importantly, this step improves identification of negative correlations, but should be applied with understanding that subsequent seed-based

correlation analyses represent partial correlations accounting for widely shared variance (Chang and Glover 2009; Fox, Zhang et al. 2009; Murphy, Birn et al. 2009; Weissenbacher, Kasess et al. 2009; Van Dijk, Hedden et al. 2010). Questions remain regarding appropriate applicability and interpretation of these measures across all populations.

3.2.5 Analysis Technique—Multiple statistical analysis techniques have been applied to identify spatial patterns of spontaneous BOLD activity in infants, most commonly seed correlation analysis (SCA) and independent component analysis (ICA) (Fox and Raichle 2007). SCA has been validated in pediatric subjects (Fox, Snyder et al. 2005; Fair, Dosenbach et al. 2007; Fair, Cohen et al. 2008; Lin, Zhu et al. 2008; Smyser, Inder et al. 2010). It is based upon calculation of temporal correlation between a single region and all other regions of the brain. It requires *a priori* assumption of regions of interest, and hence is model-based. Seed selection can profoundly impact results, with small differences in location altering network identification (Cohen, Fair et al. 2008). While it is straightforward, sensitive, and easily interpretable, its use is dedicated to investigation of pre-determined regions (Boly, Phillips et al. 2008). ICA has also been used to identify functional connections, including in neonates (Beckmann, DeLuca et al. 2005; Damoiseaux, Rombouts et al. 2006; Fransson, Skiold et al. 2007; Fransson, Skiold et al. 2009; Damaraju, Phillips et al. 2010; Doria, Beckmann et al. 2010). ICA utilizes statistical calculation to decompose data sets into maximally independent components based upon the signal intensity time course of spatial maps, and hence is data-driven rather than model-based. It is dependent upon the number of components the analysis produces and requires *a priori* assumption for system selection (Beckmann, DeLuca et al. 2005; Fox and Raichle 2007; Boly, Phillips et al. 2008). Its principle advantage is direct extraction of spatial maps with their associated time course, allowing separation of spontaneous brain activity from noise components without specifying an explicit temporal model (Beckmann, DeLuca et al. 2005; Boly, Phillips et al. 2008). Recent studies demonstrated similar results are generated using both techniques, suggesting each method extracts common signals (Beckmann, DeLuca et al. 2005; Long, Zuo et al. 2008; Thomason, Chang et al. 2008; Van Dijk, Hedden et al. 2010). Limited investigation of this finding has been performed in neonates (Damaraju, Phillips et al. 2010; Doria, Beckmann et al. 2010). Additionally, while prior investigation demonstrated networks in infants and children differ from those in adults (Fair, Dosenbach et al. 2007; Fair, Cohen et al. 2008; Thomason, Chang et al. 2008; Fransson, Skiold et al. 2009; Kelly, Di Martino et al. 2009), *a priori* assumptions previously applied when using each technique to investigate neonates are based upon results obtained in older populations. Additional systematic investigation is warranted.

Identification of individual RSNs provides the foundation for subsequent application of modern network analysis techniques to detail temporal and spatial organizational properties of large-scale functional networks and their role in information processing (He, Wang et al. 2009). These investigations focused heavily on application of graph theory, an approach for quantitative characterization and mapping of complex systems using mathematical models to investigate pairwise relationships between topographically distinct regions (Achard, Salvador et al. 2006; Achard and Bullmore 2007; Buckner, Sepulcre et al. 2009; Bullmore and Sporns 2009; Gao, Zhu et al. 2009; van den Heuvel and Hulshoff Pol 2010). Investigations of adult subjects applied these concepts to fcMRI data to enumerate global and local cerebral architectural properties, illuminating ‘small world’ characteristics of functional connections (Bassett and Bullmore 2009; Bullmore and Sporns 2009; Dosenbach, Nardos et al. 2010). The results provide insight into cerebral architecture not afforded through investigations of individual RSNs (Van Dijk, Hedden et al. 2010). The feasibility of applying this methodology in neurodevelopmental investigations has been recently established (Fair, Cohen et al. 2009; Gao, Zhu et al. 2009; Fransson, Aden et al. 2010), though these inquiries have been limited in size and scope. Application of these methods in

infant populations requires further refinement. However, early results suggest these techniques allow quantification of large-scale changes in RSNs throughout the earliest periods of development, complementing results obtained through prior RSN investigations in neonates.

In summary, while fcMRI has been used to establish the earliest forms of RSNs in infants, fundamental questions remain regarding optimal acquisition and analysis techniques. Subtle differences in methodology at numerous analysis steps can profoundly affect results. Direct investigations have been limited and incomplete, with a subset of studies performing targeted investigation on a fraction of key issues. Further, adaptation of methods used in older populations to the study of neonates raises questions regarding both interpretation of results and correlation with prior investigations of neurodevelopment performed using other modalities (Van Dijk, Hedden et al. 2010). This methodological chasm merits further systematic investigation with clarification and optimization of age-specific fcMRI techniques. Having identified these caveats, we now turn to a summary of the current literature on fcMRI in infants.

4. INVESTIGATIONS OF RESTING-STATE NETWORK DEVELOPMENT IN INFANTS USING fcMRI

Results from the limited number of fcMRI studies investigating preterm and term infants reflect the complex interplay of evolving structural and functional architecture during early development (Table 1). These studies have typically examined RSN development from term age through the first two years of life, though a subset of studies have also included preterm infants. The majority of these studies included healthy subjects born at term, seeking to define normal neural network development in this population. Five studies have also explored longitudinal development. RSNs involving cortical and subcortical structures have been identified as early as 26 weeks PMA. Recognized networks demonstrate a pattern of gradual maturation and dynamic configuration which correlates with subject age. While demonstrating the applicability of the technique in this population, these studies utilized varied acquisition and analysis techniques, at times exhibiting noted variability in results. As expanding numbers of fcMRI investigations of infants further define patterns of normative neural network development, findings can be increasingly correlated with those obtained from complementary modalities, providing greater understanding of the mechanisms of functional cerebral development.

Fransson et al. performed the first reported investigation of neural network development in neonates, studying 12 former preterm infants (5 female, 7 male) at term-equivalent PMA (Fransson, Skiold et al. 2007). Infants were scored for the presence of brain injury, and only infants without parenchymal hemorrhagic infarction or white matter injury were included. Varied perinatal conditions, including bronchopulmonary dysplasia, patent ductus arteriosus ligation, and necrotizing enterocolitis were reported in a subset of infants. Subjects were sedated with chloral hydrate. Images were acquired on a 1.5-T scanner using an 8-channel receive only head coil. Parameters were verified *via* reproduction of known RSNs in a single adult subject. Data containing significant motion were removed from the analysis during postprocessing. Spontaneous brain activity was detected using ICA. The majority of the signal variance occurred in the 0.01–0.05 Hz range. A total of 5 RSNs were reported. These were located in the occipital, somatomotor, temporal, parietal, and anterior prefrontal cortices. Networks were bilateral, with strong correlation across hemispheres. Limited intrahemispheric connectivity between disparate regions was found. The authors also reported a ‘proto-default-mode network’ which included the posterior cingulate and lateral parietal cortices. There was no identified network involving anterior and posterior midline regions. The authors concluded that the infant brain hosts resting-state activity,

demonstrating commonalities and disparities with patterns previously noted in neural networks identified in adults.

Fransson et al. subsequently sought to identify RSNs in healthy term infants (Fransson, Skiold et al. 2009). Nineteen healthy, term-born infants (11 female, 8 male) were studied. Studies were typically performed within the first 2 weeks of life. Infants were scanned during natural sleep. fcMRI acquisition parameters and analysis techniques were comparable to those described above. Six RSNs were reported, located in the occipital, sensorimotor, temporal, parietal, and prefrontal cortices in addition to the basal ganglia. Networks demonstrated varied strength based upon location. Results were similar to those reported in their initial study of preterm infants. A comparable 'proto-default-mode network' was reported. The authors concluded that multiple resting networks were identifiable in the term-born infant brain, with neither preterm birth nor light sedation interfering with the ability to detect resting-state patterns in the infant brain.

Recently, Fransson et al. applied network analysis using graph-theoretical measures to fcMRI data collected from infants in an effort to identify 'cortical hubs' of RSNs in this population (Fransson, Aden et al. 2010). Eighteen healthy, term-born infants (11 female, 7 male) were studied using similar methodology to prior investigations. Graph theory was applied to correlation matrices containing the pairwise correlation coefficients for target brain voxels with respect to all other voxels. Cortical hubs (*i. e*., brain regions demonstrating a disproportionately high level of anatomical connectivity and which presumably play a critical role in information flow) were identified primarily in regions within or adjacent to the primary sensorimotor cortices, with higher association cortices, including those previously identified to compose the mature default mode network and frontoparietal attention network, found to be less substantive candidates. In addition, functional connections associated with identified cortical hubs largely involved primary sensory systems, including the sensorimotor, auditory, and visual systems. Each of these patterns stood in direct contrast to results obtained from adult subjects studied with comparable techniques, for which cortical hubs were identified in higher association cortices such as the insula, precuneus, and ventromedial prefrontal cortex. The networks identified in both infants and older subjects demonstrated properties of a small world network organization, with short path lengths combined with local clustering enabling efficient data processing and information flow at local and global levels. Finally, presumed precursors of the default mode network, predominantly centered on the posterior cingulate cortex, were again reported. The authors concluded their findings supported the concept that neural networks undergo gradual maturation with advancing age, with functional connections necessary for completing tasks related to basal perception and action behavior maturing much earlier than those associated with higher order cognitive functions.

Smyser et al. performed longitudinal resting-state network analysis in a cohort of preterm infants aged 26 weeks PMA through term equivalent age at PMA-specific time points (Smyser, Inder et al. 2010). A total of 90 data sets were collected from 53 preterm infants. Longitudinal data were available for 28 infants. Infants were excluded if found to have high grade intraventricular hemorrhage, multicystic periventricular leukomalacia, and/or large cerebellar hemorrhage. Ten term control infants were also studied. Subjects were scanned on a 3T scanner using an infant head coil during natural sleep or while resting quietly. Data containing significant motion were removed during processing. SCA was used. Multiple RSNs were identified as early as 26 weeks PMA involving varied cortical regions, in addition to the thalamus and cerebellum (Figure 5). Identified networks demonstrated characteristic patterns of development based upon location, with distance playing a critical role in rate of RSN development. Early neural networks predominantly consisted of localized connections centered on cortical regions of interest, with nearby voxels

demonstrating high levels of correlation that decreased with distance (*i.e.*, 'local bloom'). These networks gradually became more focused (*i.e.*, decreasing size of localized activation) with advancing age, likely reflecting the maturing structural and functional localized organization of the developing brain. Further, functional connections between physically more distant regions did not appear until later in development. Significant differences were identified in infants born at term versus those born prematurely. Putative precursors of the default mode network were identified in term control infants. These connections were not detected in preterm infants, including those at term equivalent age. The identified patterns of network development demonstrate the earliest forms of cerebral functional connectivity, and were consistent with prior investigations of functional neurodevelopment in this population.

Doria et al. subsequently explored early development of RSNs through longitudinal investigation of prematurely-born infants aged 29 weeks PMA through term-equivalent (Doria, Beckmann et al. 2010). Sixty-two preterm data sets (28 female, 34 male) were included in the analysis. Diffuse high signal intensity was identified in 35 infants, with no additional overt anatomic lesions reported. Eight term-born infants were also investigated. Studies were performed on a 3T scanner using an 8-channel phased-array head coil. Investigations at term-equivalent were performed under sedation with chloral hydrate, with a subset of infants studied at earlier PMA scanned during natural sleep. Data sets containing pervasive motion were excluded. Both ICA and SCA were employed. Visual, auditory, somatosensory, motor, default mode, frontoparietal, and executive control RSNs comparable to those identified in adult subjects were identified in infants as early as 30 weeks PMA. A subset of networks was not identifiable in infants in the earliest age categories, but all networks were recognizable by term equivalent. Identified RSNs demonstrated network specific rates of development, exhibiting increasingly coherent interhemispheric activity with advancing PMA. Comparable results were obtained using each analysis approach, with increasing thalamic connectivity noted using SCA. No significant discrepancies were identified between infants scanned with and without light sedation. Recognized differences between prematurely-born and term infants were limited by correction for multiple comparisons. These findings further established the importance of this developmental period in the emergence of RSNs, while initiating investigation of important methodological concerns in this population.

Liu et al. performed the initial fcMRI study of older infants in natural sleep by investigating RSNs in the sensorimotor area in a cohort of one-year-old subjects (Liu, Flax et al. 2008). Eleven healthy, term-born infants (6 female, 5 male) were studied. Studies were performed on a 1.5-T scanner. A standard (adult) RF head coil was utilized to permit use of sound attenuation equipment. Image acquisition parameters were selected in part to maintain lower sound levels for sleeping subjects. Data containing significant motion were removed during processing. ICA was utilized to identify RSNs. Two networks were reported in the sensorimotor cortex, one located in the hand region and the other in the foot/leg region. Peak frequency for signal variance was 0.02–0.04 Hz. Unilateral correlation was identified for 9 individual subjects and in the group analysis, with bilateral correlation between homotopic counterparts identified in the remaining two infants. The authors postulated the asymmetry in networks was in part secondary to differing levels of sleep, hypothesizing that deep sleep reduces *inter*hemispheric connectivity, but not *intra*hemispheric connectivity. They also suggested identified networks represented more mature forms of those reported in Fransson et al.'s investigation of former preterm infants. However, this predominance of intrahemispheric connectivity in the sensorimotor cortex has not been confirmed in subsequent investigations in this age group, with identified neural networks in this region demonstrating bilateral associations between homotopic counterparts, including during sleep (see below) (Lin, Zhu et al. 2008).

Lin et al. performed the first reported investigation of longitudinal development within selected neural networks during infancy and early childhood (Lin, Zhu et al. 2008). A total of 35 fcMRI data sets were analyzed: 16 collected from late preterm and term infants 2–4 weeks of age, 12 from one-year-old infants and 7 from two-year-old infants. Subjects were healthy infants born between 35 and 42 weeks gestation with normal anatomic images. Subjects were scanned during natural sleep. Studies were performed on a 3T scanner. In a subset of infants and one-year-old subjects, TR time was decreased with constant total acquisition time. Data containing significant motion were removed during processing. Three regions were investigated utilizing SCA, including the primary motor, sensory, and visual cortices. Neural networks were identified in each region for subjects within each age category. For each region, functional correlations between homotopic counterparts demonstrated increasing size and strength with advancing age, with variability in rate of development between the sensorimotor and occipital cortices. Similar results were obtained for images collected with both TR times.

The same group performed a similar investigation of temporal and spatial development within the default mode network. Results from a cohort including 20 infants (11 female, 9 male), 24 one-year-olds (8 female, 16 male), and 27 two-year-olds (17 female, 10 male) were reported (Gao, Zhu et al. 2009). Inclusion criteria were similar to those described above. Subjects were scanned without sedation. Studies were again performed on a 3T scanner with identical acquisition parameters. In this study, ICA was utilized to identify neural networks, and graph theory was applied to group correlation matrices, with a spring embedding algorithm used to depict connection patterns. The authors report an incomplete, primitive default mode network in infants which increases markedly in number of regions connected and strength by one year of age. This process continues through two years of age, with the network increasingly similar to that identified in older pediatric populations and adults. Functional associations between the medial prefrontal cortex and posterior cingulate cortex were observed in each age group, and connections to the medial prefrontal cortex emerged more strongly after one year of age. Application of graph theory demonstrated regionally-specific patterns of evolving connections between regions composing the mature default mode network. The medial prefrontal cortex and posterior cingulate cortex were most strongly connected with other regions (*i.e.*, ‘hubs’), while areas located at greater distances away from the midline demonstrated lower connection strength.

Damaraju et al. investigated the effects of prematurity on the spatial and temporal properties of RSN development during early childhood (Damaraju, Phillips et al. 2010). fcMRI data were analyzed from 47 infants, including 16 preterm and 9 term infants at 18 months of age and 13 preterm and 9 term infants at 36 months of age. All subjects had normal development and structural MRI scans. Studies were performed on a 3T scanner. Subsets of preterm infants were sedated with chloral hydrate, while term infants were studied without sedation. Both ICA and SCA were employed, with ICA results used to define regions of interest for subsequent application in SCA. Time course spectra were determined using multi-taper spectral estimation. RSNs including visual, temporal, motor, basal ganglia, and default mode networks were identified in each group, with similar group mean patterns obtained using each analysis approach. Spatial properties between preterm and term-born infants were generally comparable in each age category. However, discrepancies in power spectrum and connection strength were identified between RSNs of infants categorized by gestational age at birth studied at 36 months of age. In infants born prematurely, neural networks including the basal ganglia demonstrated increased spectral energy at low frequencies. Additionally, RSNs in term infants were stronger than those in their preterm counterparts, with differences evident in a network specific pattern. The influence of sedation was limited throughout the analyses, with the significance of increased connection strength in non-sedated subjects restricted by small subject numbers. The authors concluded anatomical locations of RSNs

are established early across all groups, with differences between neural networks in preterm infants and those born at term more prominent with increasing age.

5. FUTURE DIRECTIONS

5.1 Investigation of Relationship between Brain Structure and Function

Anatomical and functional connectivity are interrelated, but not identical (Vincent, Patel et al. 2007; Damoiseaux and Greicius 2009). The intricate nature of this relationship is only beginning to be understood, with initial investigations demonstrating functional connection strength, persistence, and spatial characteristics are formed upon the large-scale anatomical structure of the developing cerebral cortex (Shatz 1996; Koch, Norris et al. 2002; Hagmann, Jonasson et al. 2006; Honey, Sporns et al. 2009). Currently, *in vivo* assessment of cerebral structural architecture is most commonly performed using diffusion weighted imaging (DWI) (including DTI and diffusion spectrum imaging [DSI]), which defines and quantifies cerebral microstructure through measurement of anisotropic water molecule diffusion (Neil, Miller et al. 2002). Investigations combining structural and functional measures provide increased breadth to assessments of cerebral connectivity, allowing characterization of heterogeneous development and maturation of the functional cerebral architecture (Supekar, Uddin et al. 2010). In pursuit of this aim, a growing number of studies have examined the interrelationship of results obtained using these modalities, predominantly through the investigation of adult subjects (Koch, Norris et al. 2002; Hagmann, Cammoun et al. 2008; Skudlarski, Jagannathan et al. 2008; van den Heuvel, Mandl et al. 2008; Zhang, Snyder et al. 2008; Damoiseaux and Greicius 2009; Greicius, Supekar et al. 2009; Honey, Sporns et al. 2009; Zhang and Raichle 2010).

Beginning with Koch's initial investigation collecting fMRI and DTI in a single axial slice in 7 adult subjects (Koch, Norris et al. 2002), these inquiries have demonstrated that identified functional connections correlate with defined anatomical connections at both a local and aggregate level (Hagmann, Cammoun et al. 2008; Honey, Sporns et al. 2009). Targeted investigation of individual networks provides the ability to identify structural connections of varied strength between functionally connected regions, supplying measures to quantify the relationship between structural and functional connectivity (van den Heuvel, Mandl et al. 2008; Greicius, Supekar et al. 2009). Global surveys have demonstrated a robust relationship between structural and functional connectivity, with strength of structural connections predictive of that of functional connections (Hagmann, Cammoun et al. 2008; Skudlarski, Jagannathan et al. 2008; Honey, Sporns et al. 2009). Importantly, these studies also established that strong functional connections exist between regions without direct structural connections (Honey, Sporns et al. 2009). One possible explanation for this finding is related to differences between DTI and fcMRI. Diffusion-based tractography presumably identifies only monosynaptic connections, since once a fiber tract enters gray matter, the tissue anisotropy is no longer high enough to continue tracking. fcMRI, in theory, can identify polysynaptic connections if the starting and downstream areas share similar spontaneous activity patterns. It is likely that some degree of neural processing takes place at each successive synapse, and this would alter overall firing patterns so similarities to that of the starting point are reduced. Thus, in a simplified sense, fcMRI would be expected to be most sensitive to monosynaptic connections, while additionally providing the ability to detect polysynaptic connections.

Other groups have combined diffusion tractography and fcMRI to investigate potential differences in populations categorized by age (including subjects as studied as early as 7 years of age) (Andrews-Hanna, Snyder et al. 2007; Damoiseaux, Beckmann et al. 2008; Supekar, Uddin et al. 2010) and neuropathology (Lowe, Beall et al. 2008). Case reports have detailed aberrant RSN development in both children and adults with congenital or induced

disruptions in corpus callosum integrity (Quigley, Cordes et al. 2003; Johnston, Vaishnavi et al. 2008; Uddin, Mooshagian et al. 2008). While a single study combined DTI and task-based fMRI to demonstrate presumed structural connections underlying functional activation in response to motor stimulation in preterm and term infants (Arichi, Moraux et al. 2010), similar investigation of RSNs in this population has not yet been performed. With the methods for successful implementation of these neuroimaging techniques in infants increasingly established, further studies applying methods which combine these modalities are likely. The result will be improved comprehension of the evolving interactions between microstructural architecture and emerging functional connections in the developing brain and characterization of the age-specific factors which define this relationship.

5.2 Neural Plasticity /Effects of Injury

The maturing brain contains an endogenous pool of multipotent progenitor cells which can proliferate and differentiate into new neurons and glial cells throughout development (Scafidi, Fagel et al. 2009). This plasticity endows a remarkable ability for structural change and external stimulation to shape neural network development through reorganization of existing circuitry. It has been shown that neural activity – both endogenous and sensory driven – is critical for refining and shaping the circuitry of the developing cerebral cortex (Penn and Shatz 1999). Further, both deprivation and abundance of sensory input at temporally specific stages of development induce physiological and structural changes that modify neural circuitry and subsequent neurodevelopmental outcomes (Penn and Shatz 1999; Tau and Peterson 2010). This inherent elasticity empowers inconsistent and unregulated environmental variables, such as those intrinsic to Neonatal Intensive Care Unit settings, to alter longitudinal neural network development, an important consideration in investigations of select populations of preterm and term infants.

Additionally, a growing body of literature has established that cerebral lesions powerfully influence neural network development, shaping their contribution to subsequent neurodevelopmental disability (Fox and Greicius 2010; Uddin, Supekar et al. 2010; Zhang and Raichle 2010). Detailed patterns of neural network abnormality have been correlated with varied forms of neuropathology, differing in magnitude based upon the extent and spatial pattern of the lesion (Penn and Shatz 1999; Honey and Sporns 2008; Alstott, Breakspear et al. 2009; Wang, Yu et al. 2010). These studies demonstrated the consequences of cerebral injury depend on effects local to the defect site, but also affect larger-scale pathways and interregional interactions with evolution over time (Honey and Sporns 2008; Wang, Yu et al. 2010). These studies suggest this effect is regionally specific, with abnormalities in the cortical midline (both anteriorly and posteriorly) and temporoparietal junction producing large, widely distributed changes in functional connectivity within and across hemispheres, whereas the effects of lesions affecting the primary sensorimotor and visual cortices remain more localized (Honey and Sporns 2008; Alstott, Breakspear et al. 2009; He, Wang et al. 2009). Though these investigations have primarily been performed in older populations, their results shape interpretation of fcMRI measures acquired from neonates and provide the foundation for further study. Preterm and term infants are known to be vulnerable to specific forms of brain injury. In addition, it is presumed the Neonatal Intensive Care Unit environment exposes infants to overwhelming and potentially injurious experiences. fcMRI in neonatal populations has potential to define the role of various risk factors in determining neurodevelopmental outcome and assist efforts targeting neuroprotection.

5.3 Clinical Applications

Early clinical investigations using fcMRI in older patients focused upon comparisons between control populations and patients with neurological and psychiatric disorders,

providing a framework for more advanced inquiries (Fox and Greicius 2010; Uddin, Supekar et al. 2010; Zhang and Raichle 2010). Focused interpretation of fcMRI data has expanded to include disease diagnosis and classification, prognosis, treatment efficacy, and longitudinal monitoring. These inquiries have shown decreased strength across multiple neural networks in individuals with varied disease states as well as significant correlations between connectivity strength and symptom severity in select patient populations (Zhang and Raichle 2010). This pattern was consistent across clinical conditions that afflict both older (*e.g.*, Alzheimer's disease, multiple sclerosis) and younger (*e.g.*, Tourette's syndrome, autism) populations (Uddin, Supekar et al. 2010; Zhang and Raichle 2010). Additionally, changes in functional connectivity have been detected in typically aging populations (Andrews-Hanna, Snyder et al. 2007; Damoiseaux, Beckmann et al. 2008). Clinical application of the methodology in specific circumstances (*i.e.*, pre-surgical planning for patients with brain tumors or epilepsy) has also been reported (Kokkonen, Nikkinen et al. 2009; Liu, Buckner et al. 2009; Shimony, Zhang et al. 2009; Zhang and Raichle 2010). Importantly, early results demonstrate fcMRI possesses inherent benefits in comparison to task-based fMRI paradigms owing to factors including improved signal to noise ratio, minimization of experimental confounds, and expanded patient populations (Shimony, Zhang et al. 2009; Zhang, Johnston et al. 2009; Fox and Greicius 2010). Investigations of clinical populations suggest the technique possesses high sensitivity, but its specificity remains undetermined (Van Dijk, Hedden et al. 2010).

The clinical utility of fcMRI in neonates remains unexplored. Early translation of fcMRI to infant pediatric practice will likely focus on two central issues - correlation of fcMRI results with neurodevelopmental outcomes and longitudinal investigation of the impact of specific forms of neuropathology on neural network development. Preliminary results have demonstrated significant differences in RSN development in preterm infants with common forms of neuropathology, including high grade intraventricular hemorrhage and periventricular leukomalacia (Smyser, Lambeth et al. 2010). Continued application of these methods in targeted clinical investigations of normal and aberrant neurodevelopment may ultimately serve to establish fcMRI as a technique for identifying neurodevelopmental biomarkers.

5.4 Confounds of fcMRI Interpretation in Infants

Despite numerous advances in application of fcMRI to infants detailed above, confounds in appropriate interpretation of these early results persist. Persistent unresolved questions focus upon appropriate interpretation of measured BOLD signals, which provide an indirect measure of neuronal activity (Logothetis, Pauls et al. 2001; Logothetis 2003). As a result, uncertainty regarding the dynamics of neurovascular coupling may complicate extrapolation of results obtained in adults to neonatal populations. Further, the normal limits of various measures for BOLD signal have not yet been firmly established in infants. Due to this lack of standardized values for significance, it is unclear how the magnitude of measured correlations should be construed, often leading to arbitrary selection of thresholds in analysis (Fox and Raichle 2007; Ment, Hirtz et al. 2009). Additionally, RSNs detected using fcMRI are not specific to monosynaptic connections and are influenced by task state (Fransson 2006), limiting inferences which can be made to the large-scale connectional architecture of the brain (Van Dijk, Hedden et al. 2010). Critical to investigations of neonates, these uncertainties are compounded by incomplete understanding of the impact of brain growth and development on the BOLD signal central to fcMRI investigations.

Potential imprecision introduced by "standard" processing measures must also be considered in interpretation of fcMRI results. The acquired BOLD signal is contaminated by spurious non-neuronal artifact due to a multitude of factors, some of which are typically more problematic in infants (*i.e.*, subject motion). This necessitates implementation of a complex

series of preprocessing measures designed to eliminate sources of spurious correlation due to non-neuronal causes, each of which raises potential interpretive issues (Van Dijk, Hedden et al. 2010). Additionally, many measures intended to improve signal to noise ratio – adjustments in voxel size, spatial smoothing and normalization, multisubject averaging – serve to limit spatial resolution (Bandettini 2009; Van Dijk, Hedden et al. 2010). Appropriate interpretation of negative correlations remains a subject of debate, particularly when the global signal has been removed by regression during preprocessing (Chang and Glover 2009; Fox, Zhang et al. 2009; Uddin, Supekar et al. 2010; Van Dijk, Hedden et al. 2010; Zhang and Raichle 2010).

Methodological concerns specific to investigations of infants also exist. As with other pediatric populations, test-retest reliability of fcMRI results (pertinent to establishment of normal limits) have not yet been firmly established (Uddin, Supekar et al. 2010). In addition, the impact of variables such as sex and ethnicity on functional connectivity development for this population remain to be explored, as these factors meaningfully affect results obtained in other neurodevelopmental investigations (Ment and Constable 2007; Scafidi, Fagel et al. 2009). Further, as fcMRI investigations expand to include infants with brain injury, the impact of these insults on cerebral vasculature, and subsequently on BOLD signal, remains incompletely understood (Seghier and Huppi 2010). Finally, in investigations of preterm infants, no “true” control population exists, as fetal MRI is not routinely performed on a research basis, and attempts at functional imaging in this population have produced mixed results due to factors such as fetal motion (Fulford, Vadeyar et al. 2004). Advances in imaging sequences and analysis techniques and a growing track record for subject safety may eventually afford this opportunity (Gowland and Fulford 2004; Rutherford, Jiang et al. 2008). Substantive inquiry into each of these areas is ongoing and will continue, as researchers seek to maximize the potential for interpretation and application of fcMRI results in investigations seeking to refine the human connectome.

6. CONCLUSIONS

fcMRI offers great promise as an investigational tool of neurodevelopment, providing unique insight into maturation of the functional cerebral architecture of the developing brain. Its use in targeted investigations of infants has recently expanded, in parallel with the explosion in application of the technique in adults. While the field remains in its early stages, much has already been learned about the principles that guide early neural network development. Additional efforts are necessary to optimize methods for image acquisition, analysis, and interpretation of results. However, the groundwork has been laid for expanded investigations designed to further define normative findings within this population and identify and characterize factors which induce aberrant neural network development. The result will be a novel neuroimaging approach which ultimately affords greater understanding of the processes of typical functional cerebral development and enable identification of infants at increased risk for disability, possibly allowing early neuroprotection with the objective of improving neurodevelopmental outcomes.

Acknowledgments

The authors would like to thank Joshua S. Shimony for valuable discussion and manuscript editing.

FUNDING

This work was supported by the National Institutes of Health [grant number R01 HD05709801 to J.J.N]. The funder had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

REFERENCES

Achard S, Bullmore E. Efficiency and cost of economical brain functional networks. PLoS Comput Biol. 2007; 3(2):e17. [PubMed: 17274684]

Achard S, Salvador R, et al. A resilient, low-frequency, small-world human brain functional network with highly connected association cortical hubs. J Neurosci. 2006; 26(1):63–72. [PubMed: 16399673]

Allendoerfer KL, Shatz CJ. The subplate, a transient neocortical structure: its role in the development of connections between thalamus and cortex. Annu Rev Neurosci. 1994; 17:185–218. [PubMed: 8210173]

Alstott J, Breakspear M, et al. Modeling the impact of lesions in the human brain. PLoS Comput Biol. 2009; 5(6):e1000408. [PubMed: 19521503]

Altaye M, Holland SK, et al. Infant brain probability templates for MRI segmentation and normalization. Neuroimage. 2008; 43(4):721–730. [PubMed: 18761410]

Anderson JS, Druzgal TJ, et al. Network anticorrelations, global regression, and phase-shifted soft tissue correction. Hum Brain Mapp. 2010

Andrews-Hanna JR, Snyder AZ, et al. Disruption of large-scale brain systems in advanced aging. Neuron. 2007; 56(5):924–935. [PubMed: 18054866]

Arichi T, Moraux A, et al. Somatosensory cortical activation identified by functional MRI in preterm and term infants. Neuroimage. 2010; 49(3):2063–2071. [PubMed: 19854281]

Bandettini PA. What's new in neuroimaging methods? Ann N Y Acad Sci. 2009; 1156:260–293. [PubMed: 19338512]

Barkovich AJ. MR imaging of the neonatal brain. Neuroimaging Clin N Am. 2006; 16(1):117–135. viii–ix. [PubMed: 16543088]

Bassett DS, Bullmore ET. Human brain networks in health and disease. Curr Opin Neurol. 2009; 22(4):340–347. [PubMed: 19494774]

Beckmann CF, DeLuca M, et al. Investigations into resting-state connectivity using independent component analysis. Philos Trans R Soc Lond B Biol Sci. 2005; 360(1457):1001–1013. [PubMed: 16087444]

Benavente-Fernandez I, Lubian-Lopez PS, et al. Safety of magnetic resonance imaging in preterm infants. Acta Paediatr. 2010; 99(6):850–853. [PubMed: 20178510]

Birn RM, Diamond JB, et al. Separating respiratory-variation-related fluctuations from neuronal-activity- related fluctuations in fMRI. Neuroimage. 2006; 31(4):1536–1548. [PubMed: 16632379]

Birn RM, Smith MA, et al. The respiration response function: the temporal dynamics of fMRI signal fluctuations related to changes in respiration. Neuroimage. 2008; 40(2):644–654. [PubMed: 18234517]

Biswal B, Yetkin FZ, et al. Functional connectivity in the motor cortex of resting human brain using echo-planar MRI. Magn Reson Med. 1995; 34(4):537–541. [PubMed: 8524021]

Bluml S, Friedlich P, et al. MR imaging of newborns by using an MR-compatible incubator with integrated radiofrequency coils: initial experience. Radiology. 2004; 231(2):594–601. [PubMed: 15128999]

Boly M, Phillips C, et al. Intrinsic brain activity in altered states of consciousness: how conscious is the default mode of brain function? Ann N Y Acad Sci. 2008; 1129:119–129. [PubMed: 18591474]

Born AP, Miranda MJ, et al. Functional magnetic resonance imaging of the normal and abnormal visual system in early life. Neuropediatrics. 2000; 31(1):24–32. [PubMed: 10774992]

Born P, Leth H, et al. Visual activation in infants and young children studied by functional magnetic resonance imaging. Pediatr Res. 1998; 44(4):578–583. [PubMed: 9773849]

Bourgeois JP, Jastreboff PJ, et al. Synaptogenesis in visual cortex of normal and preterm monkeys: evidence for intrinsic regulation of synaptic overproduction. Proc Natl Acad Sci U S A. 1989; 86(11):4297–4301. [PubMed: 2726773]

Buckner RL, Head D, et al. A unified approach for morphometric and functional data analysis in young, old, and demented adults using automated atlas-based head size normalization: reliability

and validation against manual measurement of total intracranial volume. Neuroimage. 2004; 23(2): 724–738. [PubMed: 15488422]

Buckner RL, Sepulcre J, et al. Cortical hubs revealed by intrinsic functional connectivity: mapping, assessment of stability, and relation to Alzheimer's disease. J Neurosci. 2009; 29(6):1860–1873. [PubMed: 19211893]

Bullmore E, Sporns O. Complex brain networks: graph theoretical analysis of structural and functional systems. Nat Rev Neurosci. 2009; 10(3):186–198. [PubMed: 19190637]

Burgund ED, Kang HC, et al. The feasibility of a common stereotactic space for children and adults in fMRI studies of development. Neuroimage. 2002; 17(1):184–200. [PubMed: 12482076]

Bystron I, Blakemore C, et al. Development of the human cerebral cortex: Boulder Committee revisited. Nat Rev Neurosci. 2008; 9(2):110–122. [PubMed: 18209730]

Chang C, Cunningham JP, et al. Influence of heart rate on the BOLD signal: the cardiac response function. Neuroimage. 2009; 44(3):857–869. [PubMed: 18951982]

Chang C, Glover GH. Effects of model-based physiological noise correction on default mode network anti-correlations and correlations. Neuroimage. 2009; 47(4):1448–1459. [PubMed: 19446646]

Chang C, Glover GH. Time-frequency dynamics of resting-state brain connectivity measured with fMRI. Neuroimage. 2010; 50(1):81–98. [PubMed: 20006716]

Chao DL, Ma L, et al. Transient cell-cell interactions in neural circuit formation. Nat Rev Neurosci. 2009; 10(4):262–271. [PubMed: 19300445]

Chugani HT. A critical period of brain development: studies of cerebral glucose utilization with PET. Prev Med. 1998; 27(2):184–188. [PubMed: 9578992]

Chugani HT, Phelps ME, et al. Positron emission tomography study of human brain functional development. Ann Neurol. 1987; 22(4):487–497. [PubMed: 3501693]

Citri A, Malenka RC. Synaptic plasticity: multiple forms, functions, and mechanisms. Neuropsychopharmacology. 2008; 33(1):18–41. [PubMed: 17728696]

Cohen AL, Fair DA, et al. Defining functional areas in individual human brains using resting functional connectivity MRI. Neuroimage. 2008; 41(1):45–57. [PubMed: 18367410]

Cordes D, Haughton VM, et al. Frequencies contributing to functional connectivity in the cerebral cortex in "resting-state" data. AJNR Am J Neuroradiol. 2001; 22(7):1326–1333. [PubMed: 11498421]

Dagia C, Ditchfield M. 3T MRI in paediatrics: challenges and clinical applications. Eur J Radiol. 2008; 68(2):309–319. [PubMed: 18768276]

Dagli MS, Ingeholm JE, et al. Localization of cardiac-induced signal change in fMRI. Neuroimage. 1999; 9(4):407–415. [PubMed: 10191169]

Damaraju E, Phillips JR, et al. Resting-state functional connectivity differences in premature children. Front Syst Neurosci. 2010; 4

Damoiseaux JS, Beckmann CF, et al. Reduced resting-state brain activity in the "default network" in normal aging. Cereb Cortex. 2008; 18(8):1856–1864. [PubMed: 18063564]

Damoiseaux JS, Greicius MD. Greater than the sum of its parts: a review of studies combining structural connectivity and resting-state functional connectivity. Brain Struct Funct. 2009; 213(6): 525–533. [PubMed: 19565262]

Damoiseaux JS, Rombouts SA, et al. Consistent resting-state networks across healthy subjects. Proc Natl Acad Sci U S A. 2006; 103(37):13848–13853. [PubMed: 16945915]

De Luca M, Beckmann CF, et al. fMRI resting state networks define distinct modes of long-distance interactions in the human brain. Neuroimage. 2006; 29(4):1359–1367. [PubMed: 16260155]

de Munck JC, Goncalves SI, et al. A study of the brain's resting state based on alpha band power, heart rate and fMRI. Neuroimage. 2008; 42(1):112–121. [PubMed: 18539049]

Dehaene-Lambertz G, Dehaene S, et al. Functional neuroimaging of speech perception in infants. Science. 2002; 298(5600):2013–2015. [PubMed: 12471265]

Doria V, Beckmann CF, et al. Emergence of resting state networks in the preterm human brain. Proc Natl Acad Sci U S A. 2010; 107(46):20015–20020. [PubMed: 21041625]

Dosenbach NU, Nardos B, et al. Prediction of individual brain maturity using fMRI. Science. 2010; 329(5997):1358–1361. [PubMed: 20829489]

Ehrlich I, Malinow R. Postsynaptic density 95 controls AMPA receptor incorporation during long-term potentiation and experience-driven synaptic plasticity. J Neurosci. 2004; 24(4):916–927. [PubMed: 14749436]

Erberich SG, Friedlich P, et al. Functional MRI in neonates using neonatal head coil and MR compatible incubator. Neuroimage. 2003; 20(2):683–692. [PubMed: 14568444]

Erberich SG, Panigrahy A, et al. Somatosensory lateralization in the newborn brain. Neuroimage. 2006; 29(1):155–161. [PubMed: 16112875]

Fair DA, Cohen AL, et al. The maturing architecture of the brain's default network. Proc Natl Acad Sci U S A. 2008; 105(10):4028–4032. [PubMed: 18322013]

Fair DA, Cohen AL, et al. Functional brain networks develop from a "local to distributed" organization. PLoS Comput Biol. 2009; 5(5):e1000381. [PubMed: 19412534]

Fair DA, Dosenbach NU, et al. Development of distinct control networks through segregation and integration. Proc Natl Acad Sci U S A. 2007; 104(33):13507–13512. [PubMed: 17679691]

Fair DA, Schlaggar BL, et al. A method for using blocked and event-related fMRI data to study "resting state" functional connectivity. Neuroimage. 2007; 35(1):396–405. [PubMed: 17239622]

Fox MD, Greicius M. Clinical applications of resting state functional connectivity. Front Syst Neurosci. 2010; 4:19. [PubMed: 20592951]

Fox MD, Raichle ME. Spontaneous fluctuations in brain activity observed with functional magnetic resonance imaging. Nat Rev Neurosci. 2007; 8(9):700–711. [PubMed: 17704812]

Fox MD, Snyder AZ, et al. The human brain is intrinsically organized into dynamic, anticorrelated functional networks. Proc Natl Acad Sci U S A. 2005; 102(27):9673–9678. [PubMed: 15976020]

Fox MD, Zhang D, et al. The global signal and observed anticorrelated resting state brain networks. J Neurophysiol. 2009; 101(6):3270–3283. [PubMed: 19339462]

Fransson P. How default is the default mode of brain function? Further evidence from intrinsic BOLD signal fluctuations. Neuropsychologia. 2006; 44(14):2836–2845. [PubMed: 16879844]

Fransson P, Aden U, et al. The Functional Architecture of the Infant Brain as Revealed by Resting-State fMRI. Cereb Cortex. 2010

Fransson P, Skiold B, et al. Spontaneous brain activity in the newborn brain during natural sleep--an fMRI study in infants born at full term. Pediatr Res. 2009; 66(3):301–305. [PubMed: 19531974]

Fransson P, Skiold B, et al. Resting-state networks in the infant brain. Proc Natl Acad Sci U S A. 2007; 104(39):15531–15536. [PubMed: 17878310]

Fukunaga M, Horovitz SG, et al. Large-amplitude, spatially correlated fluctuations in BOLD fMRI signals during extended rest and early sleep stages. Magn Reson Imaging. 2006; 24(8):979–992. [PubMed: 16997067]

Fulford J, Vadeyar SH, et al. Fetal brain activity and hemodynamic response to a vibroacoustic stimulus. Hum Brain Mapp. 2004; 22(2):116–121. [PubMed: 15108299]

Gaillard WD, Grandin CB, et al. Developmental aspects of pediatric fMRI: considerations for image acquisition, analysis, and interpretation. Neuroimage. 2001; 13(2):239–249. [PubMed: 11162265]

Gao W, Zhu H, et al. Evidence on the emergence of the brain's default network from 2-week-old to 2-year-old healthy pediatric subjects. Proc Natl Acad Sci U S A. 2009; 106(16):6790–6795. [PubMed: 19351894]

Garel C, Chantrel E, et al. Fetal cerebral cortex: normal gestational landmarks identified using prenatal MR imaging. AJNR Am J Neuroradiol. 2001; 22(1):184–189. [PubMed: 11158907]

Ghashghaei HT, Lai C, et al. Neuronal migration in the adult brain: are we there yet? Nat Rev Neurosci. 2007; 8(2):141–151. [PubMed: 17237805]

Glover GH, Li TQ, et al. Image-based method for retrospective correction of physiological motion effects in fMRI: RETROICOR. Magn Reson Med. 2000; 44(1):162–167. [PubMed: 10893535]

Gowland P, Fulford J. Initial experiences of performing fetal fMRI. Exp Neurol. 2004; 190 Suppl 1:S22–S27. [PubMed: 15498538]

Greicius MD, Kiviniemi V, et al. Persistent default-mode network connectivity during light sedation. Hum Brain Mapp. 2008; 29(7):839–847. [PubMed: 18219620]

Greicius MD, Supekar K, et al. Resting-state functional connectivity reflects structural connectivity in the default mode network. Cereb Cortex. 2009; 19(1):72–78. [PubMed: 18403396]

Gressens P. Mechanisms and disturbances of neuronal migration. Pediatr Res. 2000; 48(6):725–730. [PubMed: 11102537]

Hagmann P, Cammoun L, et al. Mapping the structural core of human cerebral cortex. PLoS Biol. 2008; 6(7):e159. [PubMed: 18597554]

Hagmann P, Jonasson L, et al. Understanding diffusion MR imaging techniques: from scalar diffusion-weighted imaging to diffusion tensor imaging and beyond. Radiographics. 2006; 26 Suppl 1:S205–S223. [PubMed: 17050517]

He Y, Wang J, et al. Uncovering intrinsic modular organization of spontaneous brain activity in humans. PLoS One. 2009; 4(4):e5226. [PubMed: 19381298]

Heep A, Scheef L, et al. Functional magnetic resonance imaging of the sensorimotor system in preterm infants. Pediatrics. 2009; 123(1):294–300. [PubMed: 19117895]

Hoeksma MR, Kenemans JL, et al. Variability in spatial normalization of pediatric and adult brain images. Clin Neurophysiol. 2005; 116(5):1188–1194. [PubMed: 15826861]

Honey CJ, Sporns O. Dynamical consequences of lesions in cortical networks. Hum Brain Mapp. 2008; 29(7):802–809. [PubMed: 18438885]

Honey CJ, Sporns O, et al. Predicting human resting-state functional connectivity from structural connectivity. Proc Natl Acad Sci U S A. 2009; 106(6):2035–2040. [PubMed: 19188601]

Horovitz SG, Braun AR, et al. Decoupling of the brain's default mode network during deep sleep. Proc Natl Acad Sci U S A. 2009; 106(27):11376–11381. [PubMed: 19549821]

Horovitz SG, Fukunaga M, et al. Low frequency BOLD fluctuations during resting wakefulness and light sleep: a simultaneous EEG-fMRI study. Hum Brain Mapp. 2008; 29(6):671–682. [PubMed: 17598166]

Huisman TA, Martin E, et al. Fetal magnetic resonance imaging of the brain: technical considerations and normal brain development. Eur Radiol. 2002; 12(8):1941–1951. [PubMed: 12136312]

Huppi PS, Dubois J. Diffusion tensor imaging of brain development. Semin Fetal Neonatal Med. 2006; 11(6):489–497. [PubMed: 16962837]

Huppi PS, Maier SE, et al. Microstructural development of human newborn cerebral white matter assessed in vivo by diffusion tensor magnetic resonance imaging. Pediatr Res. 1998; 44(4):584–590. [PubMed: 9773850]

Huttenlocher PR. Synaptic density in human frontal cortex - developmental changes and effects of aging. Brain Res. 1979; 163(2):195–205. [PubMed: 427544]

Huttenlocher PR. Morphometric study of human cerebral cortex development. Neuropsychologia. 1990; 28(6):517–527. [PubMed: 2203993]

Huttenlocher PR, Dabholkar AS. Regional differences in synaptogenesis in human cerebral cortex. J Comp Neurol. 1997; 387(2):167–178. [PubMed: 9336221]

Johnston JM, Vaishnavi SN, et al. Loss of resting interhemispheric functional connectivity after complete section of the corpus callosum. J Neurosci. 2008; 28(25):6453–6458. [PubMed: 18562616]

Jones RA, Palasis S, et al. MRI of the neonatal brain: optimization of spin-echo parameters. AJR Am J Roentgenol. 2004; 182(2):367–372. [PubMed: 14736664]

Kang HC, Burgund ED, et al. Comparison of functional activation foci in children and adults using a common stereotactic space. Neuroimage. 2003; 19(1):16–28. [PubMed: 12781724]

Kanold PO, Luhmann HJ. The subplate and early cortical circuits. Annu Rev Neurosci. 2010; 33:23–48. [PubMed: 20201645]

Kastrup A, Kruger G, et al. Assessment of cerebral oxidative metabolism with breath holding and fMRI. Magn Reson Med. 1999; 42(3):608–611. [PubMed: 10467308]

Kazemi K, Moghaddam HA, et al. A neonatal atlas template for spatial normalization of whole-brain magnetic resonance images of newborns: preliminary results. Neuroimage. 2007; 37(2):463–473. [PubMed: 17560795]

Kelly AM, Di Martino A, et al. Development of anterior cingulate functional connectivity from late childhood to early adulthood. Cereb Cortex. 2009; 19(3):640–657. [PubMed: 18653667]

Kiviniemi V, Jauhiainen J, et al. Slow vasomotor fluctuation in fMRI of anesthetized child brain. Magn Reson Med. 2000; 44(3):373–378. [PubMed: 10975887]

Kiviniemi V, Ruohonen J, et al. Separation of physiological very low frequency fluctuation from aliasing by switched sampling interval fMRI scans. Magn Reson Imaging. 2005; 23(1):41–46. [PubMed: 15733787]

Kiviniemi VJ, Haanpaa H, et al. Midazolam sedation increases fluctuation and synchrony of the resting brain BOLD signal. Magn Reson Imaging. 2005; 23(4):531–537. [PubMed: 15919598]

Koch MA, Norris DG, et al. An investigation of functional and anatomical connectivity using magnetic resonance imaging. Neuroimage. 2002; 16(1):241–250. [PubMed: 11969331]

Kokkonen SM, Nikkinen J, et al. Preoperative localization of the sensorimotor area using independent component analysis of resting-state fMRI. Magn Reson Imaging. 2009; 27(6):733–740. [PubMed: 19110394]

Kostovic I, Jovanov-Milosevic N. The development of cerebral connections during the first 20–45 weeks' gestation. Semin Fetal Neonatal Med. 2006; 11(6):415–422. [PubMed: 16962836]

Kostovic I, Judas M, et al. Ontogenesis of goal-directed behavior: anatomo-functional considerations. Int J Psychophysiol. 1995; 19(2):85–102. [PubMed: 7622411]

Kostovic I, Rakic P. Developmental history of the transient subplate zone in the visual and somatosensory cortex of the macaque monkey and human brain. J Comp Neurol. 1990; 297(3): 441–470. [PubMed: 2398142]

Lagercrantz H, Changeux JP. The emergence of human consciousness: from fetal to neonatal life. Pediatr Res. 2009; 65(3):255–260. [PubMed: 19092726]

Larson-Prior LJ, Zempel JM, et al. Cortical network functional connectivity in the descent to sleep. Proc Natl Acad Sci U S A. 2009; 106(11):4489–4494. [PubMed: 19255447]

Le Be JV, Markram H. Spontaneous and evoked synaptic rewiring in the neonatal neocortex. Proc Natl Acad Sci U S A. 2006; 103(35):13214–13219. [PubMed: 16924105]

Levitt P. Structural and functional maturation of the developing primate brain. J Pediatr. 2003; 143(4 Suppl):S35–S45. [PubMed: 14597912]

Lin W, Zhu Q, et al. Functional connectivity MR imaging reveals cortical functional connectivity in the developing brain. AJNR Am J Neuroradiol. 2008; 29(10):1883–1889. [PubMed: 18784212]

Liu H, Buckner RL, et al. Task-free presurgical mapping using functional magnetic resonance imaging intrinsic activity. J Neurosurg. 2009; 111(4):746–754. [PubMed: 19361264]

Liu WC, Flax JF, et al. Functional connectivity of the sensorimotor area in naturally sleeping infants. Brain Res. 2008; 1223:42–49. [PubMed: 18599026]

Logothetis NK. The underpinnings of the BOLD functional magnetic resonance imaging signal. J Neurosci. 2003; 23(10):3963–3971. [PubMed: 12764080]

Logothetis NK, Pauls J, et al. Neurophysiological investigation of the basis of the fMRI signal. Nature. 2001; 412(6843):150–157. [PubMed: 11449264]

Long XY, Zuo XN, et al. Default mode network as revealed with multiple methods for resting-state functional MRI analysis. J Neurosci Methods. 2008; 171(2):349–355. [PubMed: 18486233]

Lowe MJ, Beall EB, et al. Resting state sensorimotor functional connectivity in multiple sclerosis inversely correlates with transcallosal motor pathway transverse diffusivity. Hum Brain Mapp. 2008; 29(7):818–827. [PubMed: 18438889]

Lowe MJ, Mock BJ, et al. Functional connectivity in single and multislice echoplanar imaging using resting-state fluctuations. Neuroimage. 1998; 7(2):119–132. [PubMed: 9558644]

Lund TE, Madsen KH, et al. Non-white noise in fMRI: does modelling have an impact? Neuroimage. 2006; 29(1):54–66. [PubMed: 16099175]

Marin-Padilla M. Early prenatal ontogenesis of the cerebral cortex (neocortex) of the cat (Felis domestica). A Golgi study. I. The primordial neocortical organization. Z Anat Entwicklungsgesch. 1971; 134(2):117–145. [PubMed: 4932608]

Martuzzi R, Ramani R, et al. Functional connectivity and alterations in baseline brain state in humans. Neuroimage. 2010; 49(1):823–834. [PubMed: 19631277]

Mathur AM, Neil JJ, et al. Transport, monitoring, and successful brain MR imaging in unsedated neonates. Pediatr Radiol. 2008; 38(3):260–264. [PubMed: 18175110]

Ment LR, Constable RT. Injury and recovery in the developing brain: evidence from functional MRI studies of prematurely born children. Nat Clin Pract Neurol. 2007; 3(10):558–571. [PubMed: 17914344]

Ment LR, Hirtz D, et al. Imaging biomarkers of outcome in the developing preterm brain. Lancet Neurol. 2009; 8(11):1042–1055. [PubMed: 19800293]

Merchant N, Groves A, et al. A patient care system for early 3.0 Tesla magnetic resonance imaging of very low birth weight infants. Early Hum Dev. 2009; 85(12):779–783. [PubMed: 19926413]

Mhuircheartaigh RN, Rosenorn-Lanng D, et al. Cortical and subcortical connectivity changes during decreasing levels of consciousness in humans: a functional magnetic resonance imaging study using propofol. J Neurosci. 2010; 30(27):9095–9102. [PubMed: 20610743]

Morishita H, Hensch TK. Critical period revisited: impact on vision. Curr Opin Neurobiol. 2008; 18(1):101–107. [PubMed: 18534841]

Murphy K, Birn RM, et al. The impact of global signal regression on resting state correlations: are anti-correlated networks introduced? Neuroimage. 2009; 44(3):893–905. [PubMed: 18976716]

Muzik O, Chugani DC, et al. Statistical parametric mapping: assessment of application in children. Neuroimage. 2000; 12(5):538–549. [PubMed: 11034861]

Neil J, Miller J, et al. Diffusion tensor imaging of normal and injured developing human brain - a technical review. NMR Biomed. 2002; 15(7–8):543–552. [PubMed: 12489100]

Niedermeyer E. Ultrafast EEG activities and their significance. Clin EEG Neurosci. 2005; 36(4):257–262. [PubMed: 16296442]

O'Shaughnessy ES, Berl MM, et al. Pediatric functional magnetic resonance imaging (fMRI): issues and applications. J Child Neurol. 2008; 23(7):791–801. [PubMed: 18281625]

Partridge SC, Mukherjee P, et al. Diffusion tensor imaging: serial quantitation of white matter tract maturity in premature newborns. Neuroimage. 2004; 22(3):1302–1314. [PubMed: 15219602]

Penn AA, Shatz CJ. Brain waves and brain wiring: the role of endogenous and sensory-driven neural activity in development. Pediatr Res. 1999; 45(4 Pt 1):447–458. [PubMed: 10203134]

Prastawa M, Gilmore JH, et al. Automatic segmentation of MR images of the developing newborn brain. Med Image Anal. 2005; 9(5):457–466. [PubMed: 16019252]

Quigley M, Cordes D, et al. Role of the corpus callosum in functional connectivity. AJNR Am J Neuroradiol. 2003; 24(2):208–212. [PubMed: 12591635]

Redcay E, Kennedy DP, et al. fMRI during natural sleep as a method to study brain function during early childhood. Neuroimage. 2007; 38(4):696–707. [PubMed: 17904385]

Rivkin MJ, Wolraich D, et al. Prolonged T*2 values in newborn versus adult brain: Implications for fMRI studies of newborns. Magn Reson Med. 2004; 51(6):1287–1291. [PubMed: 15170852]

Rutherford M, Jiang S, et al. MR imaging methods for assessing fetal brain development. Dev Neurobiol. 2008; 68(6):700–711. [PubMed: 18383541]

Salvador R, Martinez A, et al. A simple view of the brain through a frequency-specific functional connectivity measure. Neuroimage. 2008; 39(1):279–289. [PubMed: 17919927]

Scafidi J, Fagel DM, et al. Modeling premature brain injury and recovery. Int J Dev Neurosci. 2009; 27(8):863–871. [PubMed: 19482072]

Schenck JF. Physical interactions of static magnetic fields with living tissues. Prog Biophys Mol Biol. 2005; 87(2–3):185–204. [PubMed: 15556658]

Seghier ML, Huppi PS. The role of functional magnetic resonance imaging in the study of brain development, injury, and recovery in the newborn. Semin Perinatol. 2010; 34(1):79–86. [PubMed: 20109975]

Seghier ML, Lazeyras F, et al. Functional MRI of the newborn. Semin Fetal Neonatal Med. 2006; 11(6):479–488. [PubMed: 17000141]

Shatz CJ. Emergence of order in visual system development. Proc Natl Acad Sci U S A. 1996; 93(2): 602–608. [PubMed: 8570602]

Shimony JS, Zhang D, et al. Resting-state spontaneous fluctuations in brain activity: a new paradigm for presurgical planning using fMRI. Acad Radiol. 2009; 16(5):578–583. [PubMed: 19345899]

Skudlarski P, Jagannathan K, et al. Measuring brain connectivity: diffusion tensor imaging validates resting state temporal correlations. Neuroimage. 2008; 43(3):554–561. [PubMed: 18771736]

Smith SM, Fox PT, et al. Correspondence of the brain's functional architecture during activation and rest. Proc Natl Acad Sci U S A. 2009; 106(31):13040–13045. [PubMed: 19620724]

Smyser, C.; Lambeth, J., et al. Pediatric Academic Societies. Vancouver, Canada: 2010. Perinatal Factors Affecting Development of Neural Networks in Preterm Infants; p. 4150E-PAS2010

Smyser CD, Inder TE, et al. Longitudinal Analysis of Neural Network Development in Preterm Infants. Cereb Cortex. 2010

Souweidane MM, Kim KH, et al. Brain mapping in sedated infants and young children with passive-functional magnetic resonance imaging. Pediatr Neurosurg. 1999; 30(2):86–92. [PubMed: 10325564]

Stewart GR, Pearlman AL. Fibronectin-like immunoreactivity in the developing cerebral cortex. J Neurosci. 1987; 7(10):3325–3333. [PubMed: 3668630]

Supekar K, Uddin LQ, et al. Development of functional and structural connectivity within the default mode network in young children. Neuroimage. 2010; 52(1):290–301. [PubMed: 20385244]

Super H, Soriano E, et al. The functions of the preplate in development and evolution of the neocortex and hippocampus. Brain Res Brain Res Rev. 1998; 27(1):40–64. [PubMed: 9639671]

Tau GZ, Peterson BS. Normal development of brain circuits. Neuropsychopharmacology. 2010; 35(1): 147–168. [PubMed: 19794405]

Thesen S, Heid O, et al. Prospective acquisition correction for head motion with image-based tracking for real-time fMRI. Magn Reson Med. 2000; 44(3):457–465. [PubMed: 10975899]

Thomason ME, Chang CE, et al. Default-mode function and task-induced deactivation have overlapping brain substrates in children. Neuroimage. 2008; 41(4):1493–1503. [PubMed: 18482851]

Uddin LQ, Mooshagian E, et al. Residual functional connectivity in the split-brain revealed with resting-state functional MRI. Neuroreport. 2008; 19(7):703–709. [PubMed: 18418243]

Uddin LQ, Supekar K, et al. Typical and atypical development of functional human brain networks: insights from resting-state FMRI. Front Syst Neurosci. 2010; 4:21. [PubMed: 20577585]

van den Heuvel M, Mandl R, et al. Microstructural organization of the cingulum tract and the level of default mode functional connectivity. J Neurosci. 2008; 28(43):10844–10851. [PubMed: 18945892]

van den Heuvel MP, Hulshoff Pol HE. Exploring the brain network: a review on resting-state fMRI functional connectivity. Eur Neuropsychopharmacol. 2010; 20(8):519–534. [PubMed: 20471808]

Van Dijk KR, Hedden T, et al. Intrinsic functional connectivity as a tool for human connectomics: theory, properties, and optimization. J Neurophysiol. 2010; 103(1):297–321. [PubMed: 19889849]

van Wezel-Meijler G, Leijser LM, et al. Magnetic resonance imaging of the brain in newborn infants: practical aspects. Early Hum Dev. 2009; 85(2):85–92. [PubMed: 19138830]

Vanhatalo S, Kaila K. Development of neonatal EEG activity: from phenomenology to physiology. Semin Fetal Neonatal Med. 2006; 11(6):471–478. [PubMed: 17018268]

Vincent JL, Patel GH, et al. Intrinsic functional architecture in the anaesthetized monkey brain. Nature. 2007; 447(7140):83–86. [PubMed: 17476267]

Wang L, Yu C, et al. Dynamic functional reorganization of the motor execution network after stroke. Brain. 2010; 133(Pt 4):1224–1238. [PubMed: 20354002]

Weissenbacher A, Kasess C, et al. Correlations and anticorrelations in resting-state functional connectivity MRI: a quantitative comparison of preprocessing strategies. Neuroimage. 2009; 47(4):1408–1416. [PubMed: 19442749]

Wilke M, Schmithorst VJ, et al. Assessment of spatial normalization of whole-brain magnetic resonance images in children. Hum Brain Mapp. 2002; 17(1):48–60. [PubMed: 12203688]

Wise RG, Ide K, et al. Resting fluctuations in arterial carbon dioxide induce significant low frequency variations in BOLD signal. Neuroimage. 2004; 21(4):1652–1664. [PubMed: 15050588]

Wu CW, Gu H, et al. Frequency specificity of functional connectivity in brain networks. Neuroimage. 2008; 42(3):1047–1055. [PubMed: 18632288]

Yamada H, Sadato N, et al. A rapid brain metabolic change in infants detected by fMRI. Neuroreport. 1997; 8(17):3775–3778. [PubMed: 9427369]

Zhang D, Johnston JM, et al. Preoperative sensorimotor mapping in brain tumor patients using spontaneous fluctuations in neuronal activity imaged with functional magnetic resonance imaging: initial experience. Neurosurgery. 2009; 65(6 Suppl):226–236. [PubMed: 19934999]

Zhang D, Raichle ME. Disease and the brain's dark energy. Nat Rev Neurol. 2010; 6(1):15–28. [PubMed: 20057496]

Zhang D, Snyder AZ, et al. Intrinsic functional relations between human cerebral cortex and thalamus. J Neurophysiol. 2008; 100(4):1740–1748. [PubMed: 18701759]

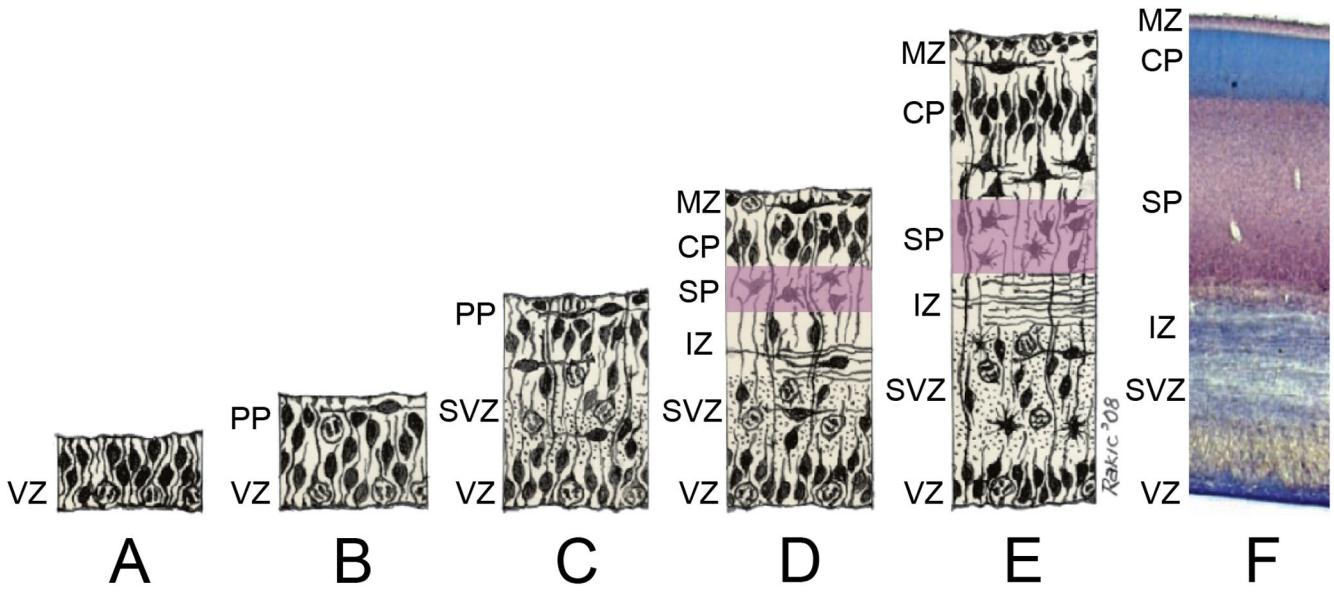

Figure 1. Histological changes in the neocortex during prenatal development

The developing neocortex is composed of a series of embryonic cellular zones, with appearance (and in some cases subsequent resolution) dependent upon gestational age. Neurons are generated in the ventricular zone (VZ). The preplate (PP) is formed by migration of post-mitotic neurons from this region. Accumulation of intermediate progenitors creates the subventricular zone (SVZ). The cortical plate (CP) forms via ensuing neurogenesis and migration in an inside-out sequence, with cortical layers 2–6 emerging through subsequent rounds of cell division. The neurons divide the preplate into the marginal zone (MZ) (below the pial surface) and the subplate zone (SP) *via* radial migration. The marginal zone becomes layer 1 of mature cortex. The intermediate zone (IZ) lies between the proliferating layers and post-migratory neurons, providing the foundation for subsequent white matter development (Bystron, Blakemore et al. 2008). Images are schematic representations of the developing neocortex at approximately **(A)** embryonic day (E) 30, **(B)** E31–32, **(C)** E45, **(D)** E55, **(E)** gestational week 14, and **(F)** gestational week 18. Adapted with permission from Annual Reviews from (Kanold and Luhmann 2010).

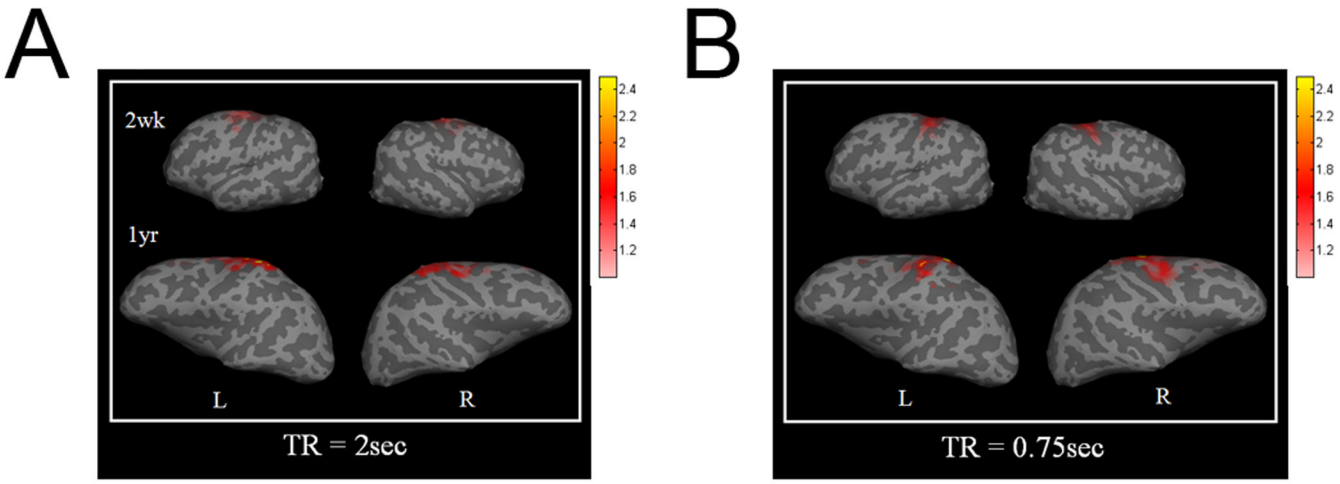

Figure 2. Consistent fcMRI results in infants using varied acquisition parameters

Selection of image acquisition parameters can significantly affect fcMRI results, and limited investigation of the effect of these differences in this population has been performed. Lin *et al* used differing repetition time (TR) settings to acquire fcMRI data in subjects 0–2 years of age. Comparable results were identified for each group, suggesting minimal effects for TR time. **(A, B)** fcMRI correlation maps generated using seeds located in the right (R) and left (L) sensorimotor cortices for infants age two weeks and one year from data obtained using TR time of **(A)** 2 seconds and **(B)** 0.75 seconds. Note similar patterns of interhemispheric correlation between homotopic counterparts for each group. Z-score values are indicated by color bar on right (threshold value = 1.0). Adapted with permission from Williams and Wilkins Co. from (Lin, Zhu et al. 2008).

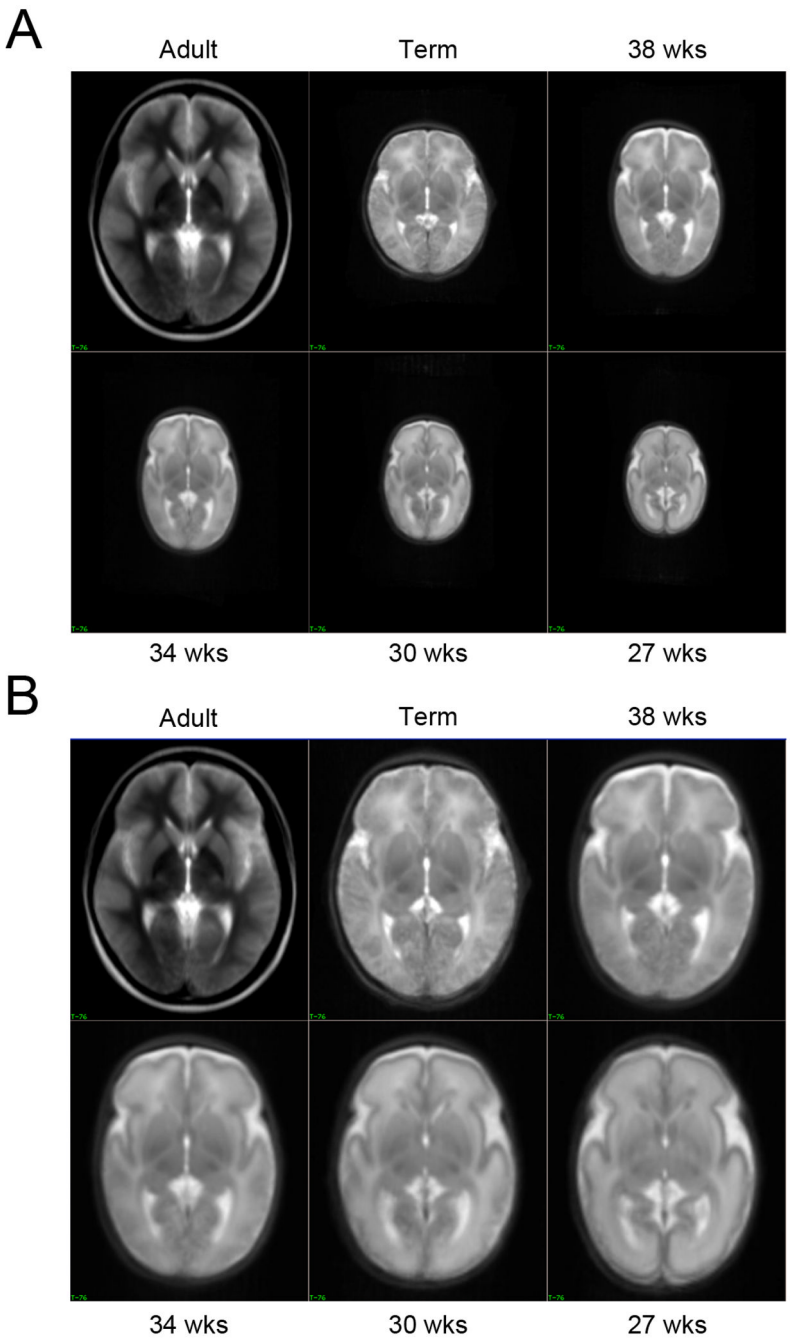

Figure 3. Gestational age specific atlases enable improved image registration

Spatial normalization to a common stereotactic space allows statistical comparison of inter- and intra-individual functional neuroimaging results. This procedure can be problematic in infants due to the rapid, regional-specific changes in cerebral anatomy which occur during early development. Use of atlas targets specific to narrow periods of development limits the impact of these effects, improving image registration and normalization. **(A)** Post menstrual age (PMA) specific targets generated for designated gestational age categories (27, 30, 34, and 38 weeks PMA and term-born infants; adult target provided for comparison) using a previously described iterative algorithm (Buckner, Head et al. 2004). Note progressive change in cerebral volume and gyration and sulcation patterns with advancing age. **(B)**

Results transformed to common (adult) stereotactic space through application of age-specific, logarithmically averaged stretch. Adapted with permission from Oxford University Press from (Smyser, Inder et al. 2010).

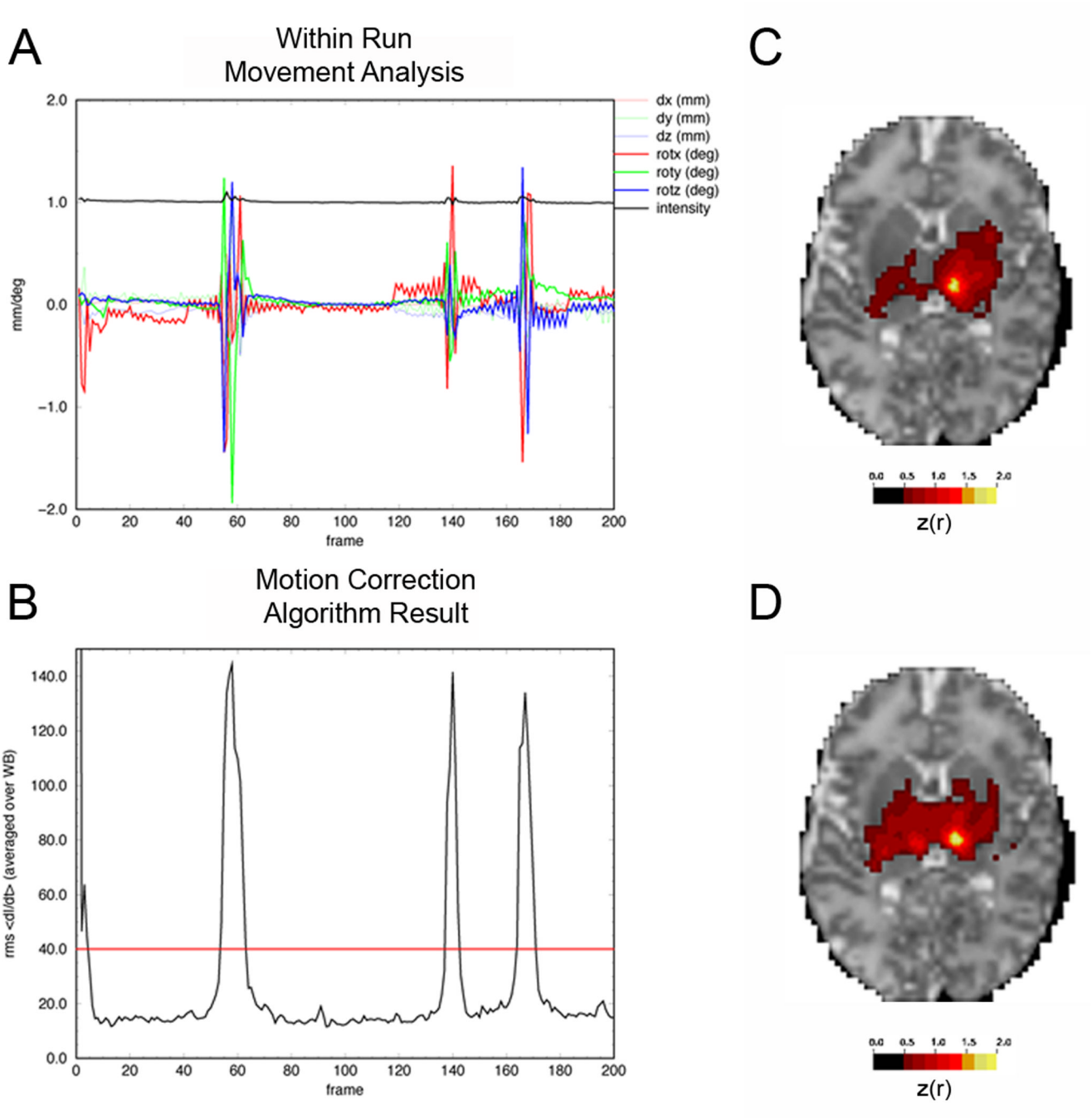

Figure 4. Detection of intermittent head motion allowing exclusion of affected volumes with increased correlation strength

Subject motion generates artifact that cannot be corrected post-acquisition. This is of significant concern in investigations of infants who are prone to movement. However, head motion tends to occur sporadically in sleeping subjects and manifests as intermittent change (usually loss) of signal intensity. **(A)** Linear and rotational head movement during acquisition for a typical subject. Changes in mean signal intensity (most likely due to intermittent head motion) are reflected in the black trace, which reflects the reciprocal of the imaged mean whole brain intensity. **(B)** Results generated by head motion correction algorithm for same fMRI data. Frames exceeding an empirically determined rms tolerance

(solid red line) are ignored in the correlation analysis. **(C, D)** fcMRI correlation maps generated using right thalamic seed for same subject without **(C)** and with **(D)** inclusion of results obtained from motion correction algorithm. Note increased interhemispheric connection strength with correction for subject movement. **(C)** and **(D)** show Fisher *z*-transformed correlation coefficients (color threshold value = 0.5). Adapted with permission from Oxford University Press from (Smyser, Inder et al. 2010).

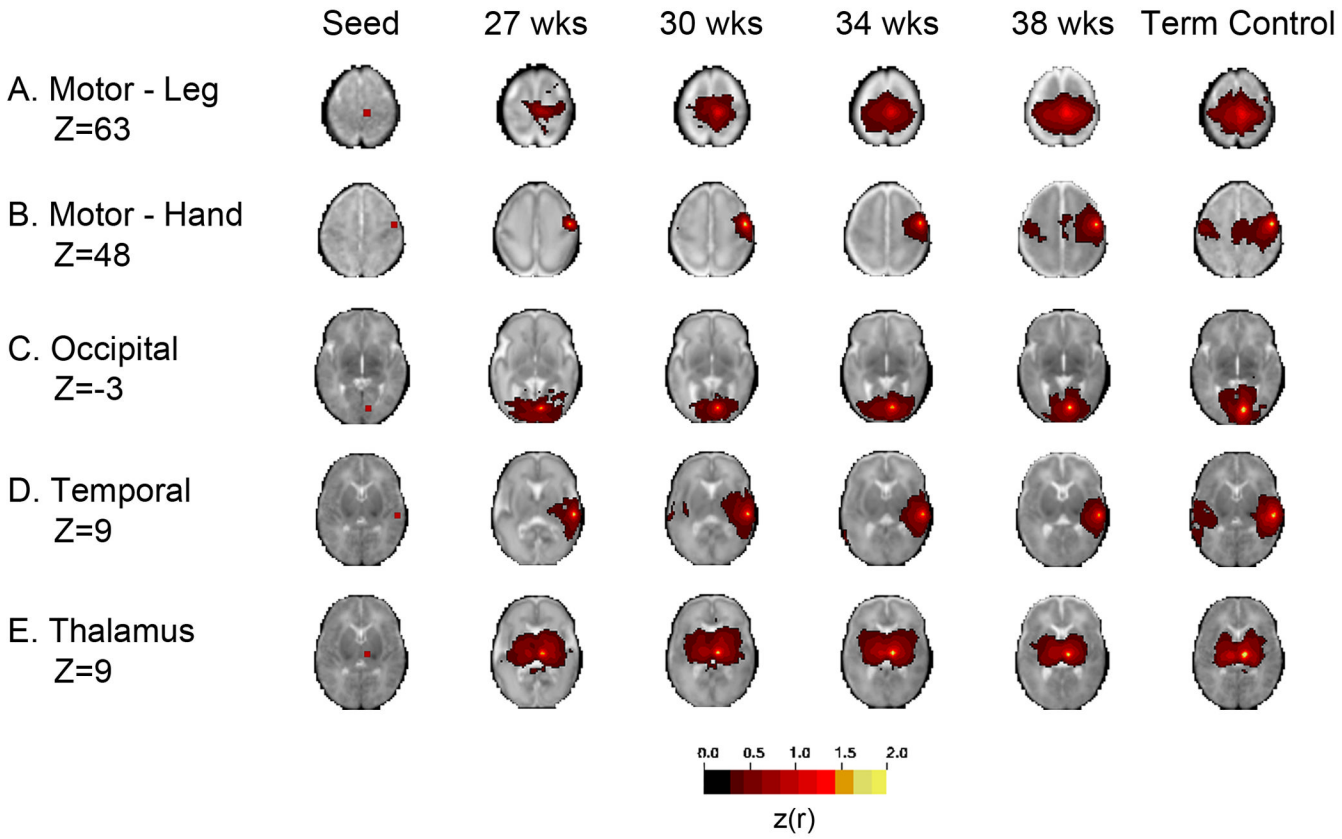

Figure 5. Longitudinal neural network development in preterm infants

Average fcMRI correlation maps corresponding to varied seed locations. The images are organized in columns corresponding to PMA at time of imaging. The illustrated quantity is the group mean Fisher *z*-transformed correlation coefficient (color threshold value = 0.3) overlaid on the gestational age specific atlas. Each row shows the axial slice at the level of the seed region. Note age dependent network maturation. Included are representative networks identified utilizing seeds located in the **(A)** motor – leg (Z=63), **(B)** motor – hand (Z=48), **(C)** occipital (Z=−3), and **(D)** temporal (Z=9) cortices and **(E)** thalamus (Z=9). The right side of the image corresponds to the right side of the brain. Adapted with permission from Oxford University Press from (Smyser, Inder et al. 2010).

TABLE 1

Previous investigations of resting-state network development in infants using fcMRI

Authors	Population	Field Strength	Parameters	RF Coil	Sedation	Duration (minutes)	Analysis Method	Neural Networks Identified
Fransson, et al. (2007)	12 former preterm infants Mean PMA at birth: 25.6 wks Mean PMA at scan: 41.0 wks	1.5T	TE=50 ms TR=2000 ms Voxel=2.8 ×2.8×4.5mm^3	8-channel receive only head	CH[a]	10	ICA[b]	1. Multiple bilateral cortical networks 2. 'Proto-DMN[c]'
Fransson, et al. (2009)	19 term infants Mean PMA at birth: 38.5 wks Mean PMA at scan: 40.3 wks	1.5T	TE=50 ms TR=2000 ms Voxel=2.8 × 2.8×4.5mm^3	6-channel receive only head	Sleep	10	ICA	1. Multiple bilateral cortical networks 2. Basal ganglia network 3. 'Proto-DMN'
Fransson, et al. (2010)	18 healthy, term infants Mean PMA at birth: 38.5 wks Mean PMA at scan: 39.2 wks	1.5T	TE=50 ms TR=2000 ms Voxel=2.8×2.8 ×4.5mm^3	6-channel receive only head	Sleep	10	Graph Theory	1. Multiple bilateral cortical networks, predominantly in sensory cortices
Smyser, et al. (2010)	53 preterm infants including longitudinal investigations 10 infants PMA at scan: 27.9 wks 16 infants PMA at scan: 30.9 wks 36 infants PMA at scan: 34.2 wks 28 infants PMA at scan: 38.2 wks	3T	TE=28 ms TR=2910 ms Voxel=2.4×2.4 ×2.4mm^3	Infant head	Sleep/ Resting Quietly	10	SCA[d]	1. Multiple bilateral cortical networks (except temporal cortex in TE) 2. Thalamic and cerebellar networks 3. DMN precursor in term infants
Doria, et al. (2010)	10 term infants 62 preterm infants 17 infants PMA at scan: 30.1 wks 21 infants PMA at scan: 33.4 wks 24 infants PMA at scan: 40.4 wks 8 term infants	3T	TE=45 ms TR=1500 ms Voxel=2.5×2.5×3.25mm^3	8-channel phased-array head	CH and Sleep	6.5	ICA SCA	1. Multiple bilateral cortical networks 2. Thalamic and cerebellar networks 3. DMN, frontoparietal, executive control networks
Liu, et al. (2008)	11 healthy, term infants Mean age at scan: 12.8 months	1.5T	TE=40 ms TR=2000 ms Voxel=3.4 ×3.4 ×5mm^3	Adult head	Sleep	5	ICA	1. Unilateral network in sensorimotor cortex in majority of infants
Lin, et al. (2008)	16 late preterm and term infants 12 one-year-old infants 7 two-year-old infants	3T	TE=32 ms TR=2000 ms Voxel=4 ×4 ×4mm^3	N/A	Sleep	5	SCA	1. 2 bilateral cortical networks in sensorimotor and occipital cortices
Gao, et al. (2009)	20 late preterm and term infants 24 one-year-old-infants 27 two-year-old infants	3T	TE=32 ms TR=2000 ms Voxel=4 ×4 ×4mm^3	N/A	Sleep	5	ICA Graph Theory	1. DMN
Damaraju, et al (2010)	16 preterm infants at 18 months 13 preterm infants at 36 months 9 term infants at 18 months 9 term infants at 36 months	3T	TE=29 ms TR=2000 ms Voxel=3.75×3.75×4.45mm^3	N/A	CH and Sleep	5.25	ICA SCA	1. Multiple bilateral cortical networks, including visual, motor, temporal 2. Basal ganglia network 3. DMN

[a] CH = Chloral Hydrate

[b] ICA = Independent Component Analysis

[c] DMN = Default Mode Network

d_{SCA} = Seed Correlation Analysis