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Brain Dynamics Underlying Cognitive Flexibility in Typical and Atypical Development

A dissertation submitted in partial satisfaction of the requirements for the Degree of Philosophy in Neuroscience

by

Lauren Breanna Kupis

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ABSTRACT OF THE DISSERTATION

Brain Dynamics Underlying Cognitive Flexibility in

Typical and Atypical Development

by

Lauren Breanna Kupis

Doctor of Philosophy in Neuroscience

University of California, Los Angeles 2024

Professor Lucina Qazi Uddin, Chair

Cognitive and behavioral flexibility necessitate mental and behavioral changes in response to changing environmental demands. Cognitive flexibility, or the mental ability to change from one thought to another, supports positive life outcomes. Individuals diagnosed with autism spectrum disorder (ASD) often face challenges with cognitive flexibility as exhibited by a core symptom of restricted and repetitive interests and behaviors (RRBs) and having poor adaptive behavior skills. ASD is rising each year, with one in 36 children diagnosed annually. Early diagnosis and treatment are critical for improving life outcomes. This makes it even more important to investigate the neural underpinnings of cognitive flexibility during early stages of development and across the lifespan to improve treatments and interventions. This dissertation seeks to understand the neural mechanisms underlying healthy cognitive flexibility development

and the cognitive inflexibility observed in ASD. Chapter 1 includes an introduction to the research involved in the following chapters, and provides a review of cognitive flexibility and its neural correlates in typical development and in ASD. Chapter 2 includes a study exploring brain dynamics and cognitive flexibility across the lifespan in a neurotypical population. Using a co-activation pattern analysis method to investigate brain dynamics, brain networks including the salience (SN), default mode (DMN), and central executive networks (CEN) underlie changes in cognitive flexibility across the lifespan. Chapter 3 then explores the neural correlates of shifting in children with and without autism during a task and resting state fMRI. Results of this study further revealed brain dynamics among the SN, DMN and CEN were involved in shifting abilities and abnormalities in these dynamics may underlie differences observed in children with ASD. Chapter 4 then examines early brain dynamics in toddlers with and without ASD and their relationship with flexible behaviors. Findings reveal differences in brain dynamics among the SN, DMN, and CEN in toddlers later diagnosed with ASD compared with non-ASD toddlers. Additionally, brain dynamics among the SN, DMN, and CEN were associated with RRBs and real-world measures of flexible behaviors across all toddlers. Chapter 5 discusses the overall findings of this dissertation, limitations, and future directions. Overall, these data indicate the importance of investigating brain dynamics associated with cognitive flexibility across the lifespan. Further, it has potential implications for autism research, implying that brain network dynamics may be an early indicator for behavioral symptoms.

The dissertation of Lauren Kupis is approved.

Lucina Qazi Uddin

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2024

DEDICATION

I dedicate this work to my parents, Teresa and Richard, who always believed me. My sisters, Catherine and Alicia, who have been here for me throughout this journey. And to my mentor, friends, and other loved ones that helped me through this journey. Ultimately, God who guided me throughout the PhD, and who kept me alive in the last few months prior to defending my thesis when I faced a near-death experience.

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Kupis LB, Uddin LQ (2023): Developmental Neuroimaging of Cognitive Flexibility: Update and Future Directions. *Annual Review of Developmental Psychology* 5: 263–284.

Bainter SA, McCauley TG, Fahmy MM, Goodman ZT, **Kupis LB**, Rao JS (2023): Comparing Bayesian Variable Selection to Lasso Approaches for Applications in Psychology. *Psychometrika* 88: 1032–1055.

Xiao, Y., Wen, T.H., **Kupis, L**. et al. Atypical functional connectivity of temporal cortex with precuneus and visual regions may be an early-age signature of ASD. Molecular Autism 14, 11 (2023). https://doi.org/10.1186/s13229-023-00543-8

Romero C, **Kupis L**, Goodman ZT, Dirks B, Baez A, Beaumont AL, *et al.* (2023): Pre-pandemic Executive Function Protects Against Pandemic Anxiety in Children with and Without Autism Spectrum Disorder. *J Autism Dev Disord*. https://doi.org/10.1007/s10803-023-06175-4

Xiao Y, Wen TH, **Kupis L**, Eyler LT, Goel D, Vaux K, et al. (2022): Neural responses to affective speech, including motherese, map onto clinical and social eye tracking profiles in toddlers with ASD. Nat Hum Behav 6: 443–454.

Kupis L, Goodman ZT, Kornfeld S, Hoang S, Romero C, Dirks B, *et al.* (2021): Brain Dynamics Underlying Cognitive Flexibility Across the Lifespan. *Cereb Cortex* 31: 5263–5274

Kupis L, Goodman ZT, Kornfeld S, Romero C, Dirks B, Kircher L, *et al.* (2021): Body mass index moderates brain dynamics and executive function: A structural equation modeling approach. *Aperture Neuro* 2021. https://doi.org/10.52294/8944e106-c54b-40d7-a620-925f7b074f99

Kupis L, Goodman ZT, Kircher L, Romero C, Dirks B, Chang C, *et al.* (2021): Altered patterns of brain dynamics linked with body mass index in youth with autism. *Autism Res* 14: 873–886.

Molnar-Szakacs, I, **Kupis, L**, Uddin, LQ, Neuroimaging Markers of Risk and Pathways to Resilience in Autism Spectrum Disorder, Biological Psychiatry: Cognitive Neuroscience and Neuroimaging, Volume 6, Issue 2, 2021, Pages 200-210, ISSN 2451-9022, https://doi.org/10.1016/j.bpsc.2020.06.017.

Kupis L, Romero C, Dirks B, Hoang S, Parladé MV, Beaumont AL, *et al.* (2020): Evoked and intrinsic brain network dynamics in children with autism spectrum disorder. *Neuroimage Clin* 28: 102396.

Dirks B, Romero C, Voorhies W, **Kupis L**, Nomi JS, Dajani DR, *et al.* (2020): Neural Responses to a Putative Set-shifting Task in Children with Autism Spectrum Disorder. *Autism Res* 13: 1501–1515.

Marshall E, Nomi JS, Dirks B, Romero C, **Kupis L**, Chang C, Uddin LQ (2020): Co-activation pattern analysis reveals altered salience network dynamics in children with autism spectrum disorder. *Network Neuroscience* 1–16.

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Sigar, P., Kathrein, N., Kupis, L., Uddin, LQ. (2024). Age related changes in brain signal variability in autism spectrum disorder. Molecular Autism (Under Review).

Bainter, A., Sierra, McCauley, G., Thomas, Fahmy, M., Mahmoud, Goodman, T., Zachary, **Kupis, B. Lauren**, Rao, J. Sunil. (2022). Advantages of Bayesian Variable Selection for Applications in Psychology. Psychological Methods (Under Review).

Bainter, A., Sierra, McCauley, G., **Kupis, B. Lauren**, Thomas, Fahmy, M., Mahmoud, Goodman, T., Zachary, Rao, J. Sunil. (In Preparation). Neural predictors of OCD using Bayesian Variable Selection.

CHAPTER 1:

Developmental Neuroimaging of Cognitive Flexibility: Update and Future Directions

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ANNUAL REVIEWS

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cognitive flexibility, neurodevelopment, executive function, task fMRI

Abstract

Cognitive flexibility, or the ability to mentally switch between tasks according to changing environmental demands, supports optimal life outcomes, making it an important executive function to study across development. Here we review the literature examining the development of cognitive flexibility, with an emphasis on studies using task-based functional magnetic resonance imaging (fMRI). The neuroimaging literature suggests that key brain regions important for cognitive flexibility include the inferior frontal junction and regions within the midcingulo-insular network, including the insular and dorsal anterior cingulate cortices. We further discuss challenges surrounding studying cognitive flexibility during neurodevelopment, including inconsistent terminology, the diversity of fMRI task paradigms, difficulties with isolating cognitive flexibility from other executive functions, and accounting for developmental changes in cognitive strategy. Future directions include assessing how developmental changes in brain network dynamics enable cognitive flexibility and examining potential modulators of cognitive flexibility including physical activity and bilingualism.

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INTRODUCTION

What Is Cognitive Flexibility?

Cognitive flexibility is a core aspect of executive function that facilitates adaptive responses to changing environmental demands throughout the life span (Dajani & Uddin 2015, Diamond 2013, Logue & Gould 2014). Examples of cognitive flexibility include the ability to think differently about a situation and to quickly switch between tasks or strategies to solve a problem. Being flexible ultimately aids creativity (Lu et al. 2017, 2019), problem-solving (Ionescu 2012), learning (Kehagia et al. 2010), and resilience to negative life events (Genet & Siemer 2011). Cognitive flexibility benefits adaptation via the ability to quickly transition between different activities or change perspective, making it an important feature of daily functioning. Cognitive flexibility is also associated with positive life outcomes including academic achievement, such as stronger reading and math skills (Yeniad et al. 2013), and the successful transition into adulthood (Burt & Paysnick 2012). Conversely, cognitive inflexibility may be a risk factor for repetitive or ruminative thoughts (Deveney & Deldin 2006, Genet et al. 2013, Whitmer & Banich 2007), which underlie psychological disorders including anxiety, depression, obsessive-compulsive disorder, and autism spectrum disorder (Keenan et al. 2018, McDougle et al. 1995, van Steensel et al. 2011). Ultimately, cognitive flexibility is important during childhood and adolescence because these developmental periods are marked by learning, susceptibility to psychological disorders [e.g., anxiety and depression (Côté et al. 2009)], and increased risk for substance use initiation (Rose et al. 2019). In all cases, cognitive flexibility may buffer against negative mental health or cognitive outcomes.

Although cognitive flexibility contributes to positive adaptation and learning across development, the underlying brain regions supporting the developmental changes of cognitive flexibility are not yet fully understood. Characterizing the brain regions involved in cognitive flexibility across development may clarify the mechanisms underlying its age-related improvement. The frontoparietal network (FPN) has been found to be important in the development of executive function broadly and in adult levels of cognitive flexibility specifically (Dajani & Uddin 2015).

Additional work examining brain network dynamics in adults demonstrates that flexible interactions among brain regions and neural networks may contribute to greater cognitive flexibility (Nomi et al. 2017a). Findings from studies primarily conducted in adults reflect the neural processes associated with peak cognitive flexibility. A small but growing body of neuroimaging work focuses on the development of cognitive flexibility from childhood through adolescence into adulthood. Better understanding of the neural mechanisms underlying the development of cognitive flexibility can lead to the creation of treatments for neurodevelopmental disorders, tailored classroom curricula, and preventative targeted and individualized measures for psychological disorders or substance misuse (Uddin 2021a). To identify gaps in the literature and guide future studies, the current review synthesizes findings from neurodevelopmental functional magnetic resonance imaging (fMRI) studies utilizing a range of cognitive flexibility tasks.

We begin with summarizing common issues that arise when studying cognitive flexibility: (*a*) the multiplicity of terms used to describe cognitive flexibility, (*b*) the variety of fMRI task paradigms used to assess cognitive flexibility, (*c*) difficulties in isolating cognitive flexibility from the other core executive functions (i.e., inhibition and working memory), and (*d*) accounting for cognitive strategy differences with age and development. These challenges each have important implications for the neuroimaging findings discussed in this review. We conclude the review of neurodevelopmental literature on cognitive flexibility with a discussion of the limitations of these studies and proposed future directions.

CONCEPTUAL MODELS OF COGNITIVE FLEXIBILITY

Terminology

A major challenge to studying cognitive flexibility in developmental fMRI studies is that a variety of terminologies are used to describe the construct. Here, we outline terminology in the context of task paradigms commonly used in developmental neuroimaging studies (**Figure 1**). This overview is not meant to redefine cognitive flexibility but rather to illustrate the types of studies that are referred to throughout the review. Cognitive flexibility was first considered to be the umbrella term used to describe studies using set-shifting and task-switching tasks (Konishi et al. 1998, Monsell 2003). Relatedly, cognitive flexibility is referred to as shifting in latent models of executive

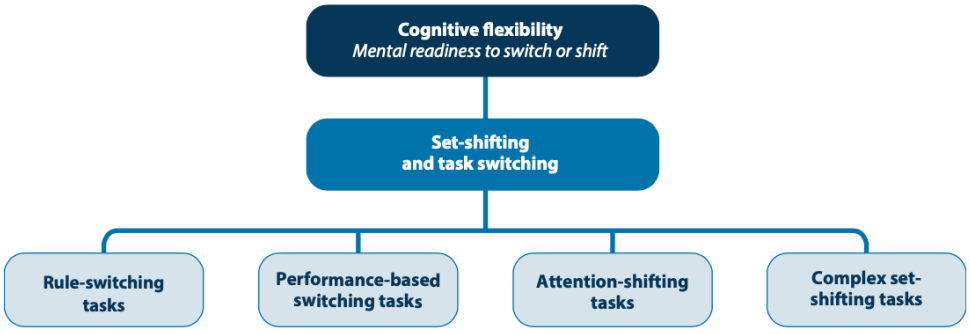

Figure 1

Overview of task paradigms typically used to assess cognitive flexibility, which is the mental readiness to flexibly shift in response to dynamic environmental demands. Task-switching and set-shifting tasks are often used to elicit cognitive flexibility. Variants of these paradigms include rule-switching tasks, performance-based switching tasks, attention-shifting tasks, and complex set-shifting tasks.

function (Miyake et al. 2000). The various terminologies used to describe cognitive flexibility and the tasks used to study it are important to clearly delineate, as different behavioral or neural processes may be associated with the different terms.

Two types of experimental paradigms often used to assess cognitive flexibility are task switching and set-shifting. Task-switching paradigms involve alternating between tasks; performance is measured by a switch cost, the difference in task performance (e.g., reaction time) between switching and nonswitching task blocks (Monsell 2003, Vandierendonck et al. 2010). An example of task switching is in rule-switching tasks, which require participants to switch their response selection (or task) based on the presented rule (Wendelken et al. 2012). Set-shifting, on the other hand, involves shifting or switching within a task (Dajani & Uddin 2015). For example, a commonly used set-shifting task requires participants to shift attention between color and shape dimensions and choose a unique attribute (Casey et al. 2004) (**Figure 2*a***). Although these types of paradigms differ in terms of switching within versus between tasks, they are both thought to engage cognitive flexibility. However, these subtle differences may have important implications in the neural processes underlying the different shifting processes occurring.

Task-Switching and Set-Shifting Paradigms

There are different types of task-switching and set-shifting paradigms used to study cognitive flexibility in developmental neuroimaging studies (**Figure 1**). Broadly, these tasks include attention-shifting, complex set-shifting, rule-switching, and performance-based switching (**Figure 2**). These fMRI-based tasks are often adaptations of common behavioral test batteries that assess cognitive flexibility such as the Dimensional Change Card Sort (DCCS) (Zelazo 2006), the Wisconsin Card Sorting Task (WCST) (Heaton et al. 1993), and shifting tasks within the Delis-Kaplan Executive Function System (D-KEFS) (Delis et al. 2001). In the DCCS, children are presented with two cards (e.g., blue rabbit and red boat) and are asked to match a series of cards to those two based on one dimension, such as color or shape (e.g., blue boat is matched with blue rabbit) (Zelazo 2006). The WCST can be administered to individuals between 6.5 and 89 years of age and requires cards to be sorted based on one of three dimensions (color, shape, or number). Individuals must update the sorting criteria based on the experimenter's feedback (Heaton et al. 1993). Lastly, the D-KEFS consists of several tasks (color–word interference, card sorting, design fluency, and verbal fluency tasks) that can be used to assess shifting abilities across 8–89-year-olds (Delis et al. 2001).

Attention-shifting. In attention-shifting tasks (**Figure 2*a***), participants shift attention between stimulus dimensions (e.g., color, shape). Attention-shifting tasks are the primary cognitive flexibility task utilized in developmental neuroimaging studies (Casey et al. 2004, Dirks et al. 2020, Morton et al. 2009, Yerys et al. 2015). Attention-shifting tasks are often an adaptation of the DCCS test battery (Ezekiel et al. 2013, Morton et al. 2009) and therefore can be given to children as young as 3 years (Hanania & Smith 2010).

Set-shifting. In complex set-shifting tasks (**Figure 2*b***), participants are still required to shift within a trial but utilize more cognitive abilities than simply shifting attention (Yasumura et al. 2015). One example of a complex set-shifting task is the flexible item selection task (FIST) (Jacques & Zelazo 2001). In one version, participants are presented with three cards and are instructed to select two of them that match on one dimension (e.g., number, color, size) and then shift by selecting another matching pair based on a different dimension (Jacques & Zelazo 2001). The FIST is similar to the DCCS and WCST but can vary in difficulty by including more abstract or complex dimensional shifts, which can be further altered by increasing dimensions or shifts within a

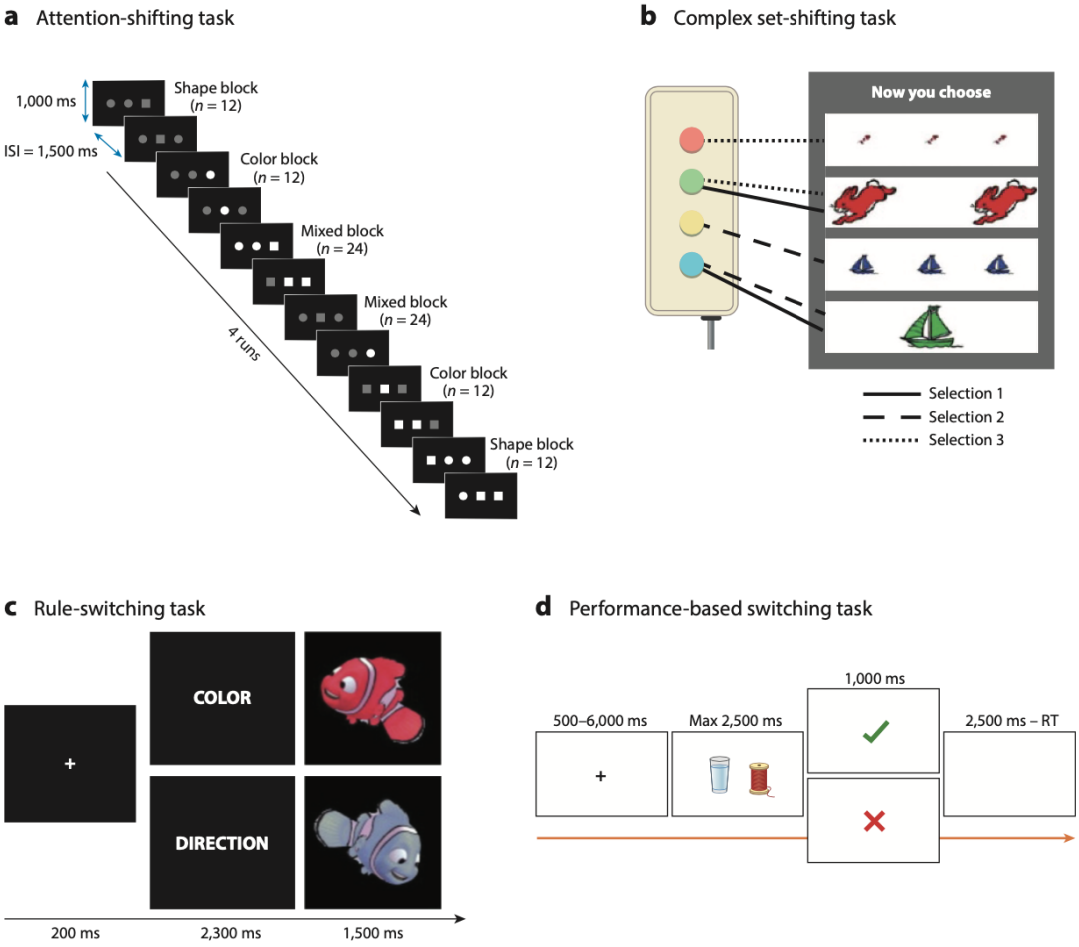

Figure 2

An overview of commonly used cognitive flexibility tasks in developmental cognitive neuroscience. Four general categories of commonly used fMRI tasks to elicit cognitive flexibility are (*a*) attention-shifting, (*b*) complex set-shifting, (*c*) rule-switching, and (*d*) performance-based switching tasks. (*a*) In attention-shifting tasks, participants shift attention among stimuli. As shown in the example, participants are presented with three shapes and must shift their attention to the shape that is different from the other two in one way (color or shape). (*b*) In more complex set-shifting tasks, participants may need to utilize more executive functions or cognitive abilities to complete the task. For the example in this panel, participants must select two items that are similar in one way (color, shape, size, or number). Then they are asked to make two more selections within the same trial. (*c*) In rule-switching tasks, participants are required to make a selection based on a rule (e.g., color or direction), and these rules switch randomly throughout the run. (*d*) In performance-based switching tasks, participants make selections based on a probability and feedback from the experimenter. Participants choose one stimulus and receive positive or negative feedback based on probabilistic rules (e.g., the first object receives positive feedback 80% of the time whereas the other object receives positive feedback 20% of the time). Abbreviations: fMRI, functional magnetic resonance imaging; ISI, interstimulus intervals; RT, reaction time.

trial (Dajani et al. 2020, Dick 2014). As in the FIST, some complex set-shifting tasks appear to be more difficult for children based on their behavioral performance (Dajani et al. 2020, Dick 2014) compared with attention-shifting tasks. Therefore, complex set-shifting tasks may require greater cognitive demands compared with attention-shifting tasks.

Rule-switching. Rule-switching tasks (**Figure 2*c***) require participants to flexibly respond to a stimulus on the basis of the given rule (e.g., to respond based on color or direction) (Wendelken et al. 2012). Rule-switching tasks sometimes have high working memory demands, as participants must mentally maintain the rules (Dajani & Uddin 2015). Although other shifting tasks may also draw on working memory skills to complete the task, rule-switching tasks sometimes require extended maintenance of the rule, rule retrieval, and suppression of previous rules, adding to the difficulty of the task.

Performance-based switching. Lastly, performance-based switching tasks (**Figure 2*d***) typically involve (*a*) an adaptation of the WCST, where participants have to spatially switch between three possible response rules (Crone et al. 2008), and (*b*) a probabilistic task (Hauser et al. 2015, van den Bos et al. 2012), where participants have to switch their stimulus choice based on probabilistic feedback (e.g., choosing stimulus A receives positive feedback 80% of the time whereas choosing B results in positive feedback only 20% of the time). Variations of this task exist, such as implementing reversal learning within a probabilistic framework. Reversal learning in humans is primarily probabilistic and requires participants to change their behavioral response when the reward contingency previously learned is reversed (Izquierdo et al. 2017).

Task-Dependent Brain Regions: Implications from Adult Findings

Understanding task-related brain activity in adult neuroimaging studies may highlight key findings that could have implications for neurodevelopment. fMRI studies of adults using age-adjusted versions of these tasks have shown the core brain regions and networks underlying cognitive flexibility (Dajani & Uddin 2015) as well as task-dependent brain regions (Kim et al. 2011, 2012) (**Figure 3**). Brain regions found to be involved in core cognitive flexibility across all task types include the ventrolateral prefrontal cortex (PFC), dorsolateral PFC (dlPFC), anterior cingulate, right anterior insula (AI), inferior frontal junction (IFJ), premotor cortex (PMC), inferior and superior parietal cortices, inferior temporal cortex, occipital cortex, and subcortical structures such as the caudate nucleus and thalamus (Dajani & Uddin 2015, Kim et al. 2012, Niendam et al. 2012). **Figure 3** includes the core regions involved in cognitive flexibility; note that not all brain regions listed above are included in the figure. Brain regions involved in cognitive flexibility further differentiate depending on the type of task (Kim et al. 2011, 2012). More abstract switching (e.g., complex set-shifting tasks with more dimensions or stimuli) recruits anterior PFC regions, moderately abstract switching (e.g., complex set-shifting tasks with fewer dimensions or stimuli) recruits mid-PFC regions, and constrained switching (i.e., attention-shifting tasks) recruits posterior PFC regions (Kim et al. 2011). Results from Kim and colleagues' (2012) meta-analysis further revealed region-specific activation during attention-shifting tasks in the dorsal portion of the PMC and during set-shifting tasks in the frontopolar cortex. Due to findings of task-dependent neural and behavioral responses associated with cognitive flexibility, it has been suggested there may be different types of cognitive flexibility (Eslinger & Grattan 1993, Kim et al. 2011, Kraft et al. 2020). Overall, the findings from adult neuroimaging studies of cognitive flexibility reveal core brain regions and task-specific regions involved in this function. The difference between core and task-dependent brain regions associated with cognitive flexibility is not yet well established in children.

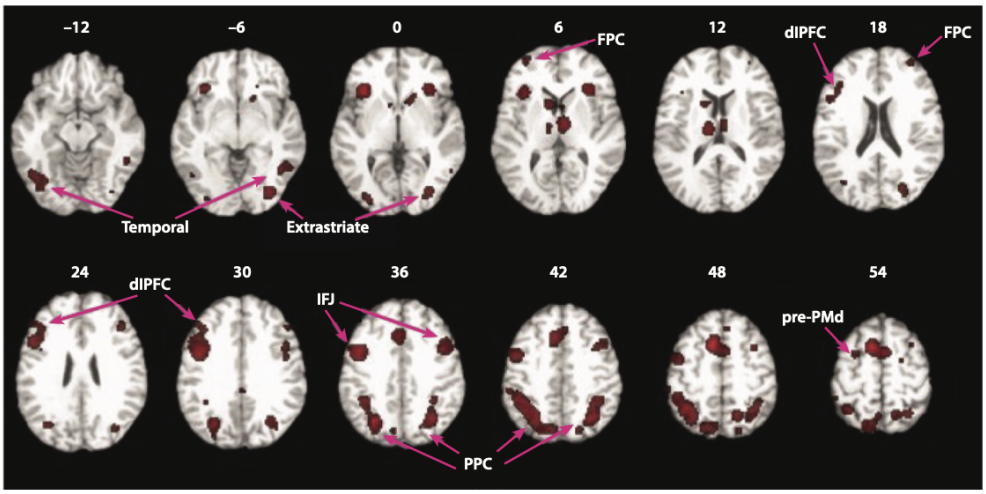

Figure 3

Summary of core brain regions involved in cognitive flexibility across task types in adults. Switching is associated with activation in the medial prefrontal cortex, lateral prefrontal cortex, and parietal, temporal, and occipital cortices, as well as subcortical structures such as the caudate nucleus and thalamus. Abbreviations: dlPFC, dorsolateral prefrontal cortex; FPC, frontopolar cortex; IFJ, inferior frontal junction; PMd, dorsal premotor cortex; PPC, posterior parietal cortex. Figure reproduced with permission from Kim et al. (2011).

Isolating Cognitive Flexibility from Other Executive Functions in Neuroimaging Studies

Another challenge involves isolating cognitive flexibility from other core executive functions (i.e., inhibition and working memory). This may impact our understanding of the brain regions underlying cognitive flexibility specifically across development. Executive functions enable goal-oriented behaviors, and the extant literature suggests that core executive functions are independent yet highly interrelated constructs (Miyake et al. 2000). Therefore, isolating executive functions from each other becomes difficult because many of these processes may in some cases depend on each other. For example, many of the flexibility tasks described above involve working memory to keep the task rules in mind and inhibition to prevent repetition of previously answered responses. Additionally, in adult studies of the neural substrates underlying cognitive flexibility, there are brain regions activated solely during cognitive flexibility tasks and brain regions activated across tasks of executive functions more broadly (for a meta-analysis, see Rodríguez-Nieto et al. 2022). Thus, fMRI studies of cognitive flexibility during childhood and adolescence may not always be testing pure cognitive flexibility. Because of this, multiple considerations need to be made when assessing the results from the developmental fMRI studies of cognitive flexibility, including the type of task used, executive functions required to complete the task, and the developmental stage of the participants, since executive functions have different developmental trajectories.

Accounting for Age Differences in the Context of Cognitive Flexibility

Another challenge of studying cognitive flexibility involves accounting for age differences associated with the development of cognitive flexibility. Cognitive flexibility and other core executive functions develop differently across childhood (Anderson 2002). Cognitive flexibility may be demonstrated as early as infancy (Shinya et al. 2022) but is typically first studied in preschool

children as young as 3 years (Hughes 1998) in lower-order forms, such as shifting. Evidence of the emergence of cognitive flexibility is supported by the ability to perform well on shifting tasks, such as the DCCS (Diamond et al. 2005) and A-not-B tasks (Johansson et al. 2015). Other executive functions such as inhibition and working memory also emerge in toddlerhood and mature throughout adolescence (Dajani & Uddin 2015, Luna 2009). However, children show difficulty with integrating multiple executive functions at once, as revealed by poorer performance on tasks requiring the integration of various executive functions (Diamond et al. 2005). For example, 4.5-year-old children successfully perform the DCCS task but perform poorly on the WCST (Chelune & Baer 1986, Dick 2014), suggesting that cognitive flexibility can be altered by task difficulty and that higher-order flexibility takes longer to develop. Through elementary school age, cognitive flexibility undergoes a prolonged development compared with other executive functions (Davidson et al. 2006) and begins to rapidly increase around 10 years of age (Chelune & Baer 1986, Dick 2014, Rosselli & Ardila 1993, Welsh et al. 1991). However, cognitive flexibility skills continue to develop into adolescence and adulthood (Anderson 2002), peaking between 21 and 30 years (Cepeda et al. 2001). Overall, considering the behavioral progression of cognitive flexibility across development may be important for understanding the neural changes underlying the development of cognitive flexibility across childhood and into adulthood.

NEURAL MARKERS OF COGNITIVE FLEXIBILITY ACROSS DEVELOPMENT

Neurodevelopmental Findings

Developmental neuroimaging studies suggest that cognitive flexibility undergoes a prolonged trajectory, as paralleled in behavioral findings (**Table 1**). Prolonged development is observed in structural findings among the brain regions supporting cognitive flexibility, mainly in the FPN (Gogtay et al. 2004), suggesting the gray matter in those regions mature and thin later than in other regions (Casey et al. 2000, Giedd et al. 1999). Similar to findings in adults, brain regions commonly activated in children across switching tasks include the FPN [the dlPFC, IFJ, and pre-supplementary motor area (SMA)] (Morton et al. 2009, Wendelken et al. 2012) and the basal ganglia (Casey et al. 2004, Crone et al. 2006b, Rubia et al. 2006). However, connectivity among the FPN and insula increases in activation strength across development, and these regions activate most strongly in adults, suggesting that cognitive flexibility strengthens as these regions develop (Kupis et al. 2021b, Rubia et al. 2006, Taylor et al. 2012, Wendelken et al. 2012).

Attention-shifting. Much like in the behavioral findings, cognitive flexibility–related brain activation can also be differentiated by switching or shifting tasks. In attention-shifting tasks, children and adults share common brain activation in regions including the superior parietal cortex, dlPFC, IFJ, pre-SMA (Morton et al. 2009), and caudate nucleus (Casey et al. 2004). Key developmental differences between children and adults, however, are seen during attention-shifting tasks. In a study by Morton et al. (2009), children (11–13 years) exhibited unique activation in the right superior frontal sulcus, whereas adults exhibited unique activation in the left superior parietal cortex and right thalamus. This finding implies that children may have different switching strategies compared with adults, resulting in differing brain activation patterns. The stronger parietal activation in adults versus stronger frontal activation in children during the task may result from adults relying on more attentional efforts to complete the shift, whereas children may be relying on more executive efforts. Casey et al. (2004) observed more prefrontal and parietal regions activated in adults compared with children (7–11 years), suggesting greater recruitment of these cortical regions across development. In Dirks et al. (2020), the same task from Casey et al. (2004)

Table 1 Developmental cognitive flexibility neuroimaging studies

Study	Number of subjects	Age range (years)	Neuroimaging modality	Cognitive flexibility task	Behavioral results	Contrast	Brain region differences between children and adults	Brain regions activated in all groups
Casey et al. 2004	7 children, 7 adults	7–11, 18–23	fMRI	Attention-shifting	Adults were more accurate than children Adults responded faster than children	None	Adults had greater activation than children in R superior frontal gyrus, L superior parietal lobule, R fusiform gyrus, L/R precuneus, R middle temporal gyrus	R inferior caudate nucleus
Rubia et al. 2006	27 children/ adolescents, 22 adults	10–17, 20–43	fMRI	Attention-shifting	Adults had fewer errors than children/ adolescents during switch trials	None	Adults had greater activation than children and adolescents in R anterior cingulate/putamen, R inferior prefrontal cortex/insula, L inferior parietal cortex	None
Morton et al. 2009	14 children, 13 adults	11–13, 19–25	fMRI	Attention-shifting	No differences in switch cost Adults were more accurate than children Children had more switch cost errors than adults	None	Children, but not adults, had activation in the R superior frontal sulcus Adults, but not children, had activation in the L superior parietal cortex and R thalamus	Superior parietal cortex, dlPFC, pre-SMA, IFJ, fusiform gyrus
Moriguchi & Hiraki 2009	15 3-year-old children, 11 5-year-old children, 10 adults	37–47 months, 61–74 months, 21–31 years	fNIRS	Attention-shifting	3-year-olds were less accurate than 5-year-olds and adults in post-switch accuracy 3-year-olds were slower than 5-year-olds and adults pre- and post-switch	Pre-switch > control Post-switch > control	3-year-olds without perseverative errors were similar to 5-year-olds and adults in oxy-Hb changes in the inferior prefrontal cortex Adults had increased oxy-Hb in inferior prefrontal regions in pre- and post-switching > control trials 5-year-olds had increased oxy-Hb in prefrontal regions in post-switching > control trials 3-year-olds (no perseverative errors) had increased oxy-Hb in prefrontal regions in post-switching > control trials 3-year-olds (with perseverative errors) had decreases in oxy-Hb in inferior prefrontal regions in pre- and post-switching > control trials	Inferior prefrontal cortex

(*Continued*)

Table 1 (*Continued*)

Study	Number of subjects	Age range (years)	Neuroimaging modality	Cognitive flexibility task	Behavioral results	Contrast	Brain region differences between children and adults	Brain regions activated in all groups
Christakou et al. 2009	63	13–38	fMRI	Attention-shifting	Adults and adolescents both had switch costs on mean RT during switch trial	None	NA	Medial frontal cortex, anterior and posterior cingulate gyrus, bilateral inferior parietal and superior temporal gyrus, left putamen and thalamus
Ezekiel et al. 2013	14 children, 13 adults	12 (mean), 23.9 (mean)	fMRI	Attention-shifting	Adults and children had a similar accuracy and RT Both children and adults were slower and had more errors during switch trials	Adults > children, children > adults	Adults had greater connections than children in the bilateral dorsolateral prefrontal cortex, right inferior frontal gyrus, anterior cingulate/medial prefrontal cortex, bilateral inferior parietal cortex, and ventral tegmental area Children had greater connections than adults in regions more strongly connected with CCN: anterior extent of the superior and middle frontal gyri, frontal pole, right anterior insula, and left posterior temporal cortex Age-related decreases in FC of frontopolar, insular, and temporal cortex with CCN	None
Rodehacke et al. 2014	185 adolescents, 28 adults	13–15, 20–50	fMRI	Attention-shifting	Adults had fewer errors than adolescents	None	When comparing adolescents to adults, adolescents had greater activation during switch > repeat trials in regions including the inferior temporal lobe, superior temporal gyrus, cuneus, lingual gyrus, inferior occipital gyrus, cingulate gyrus, anterior cingulate gyrus, and middle frontal gyrus	Switch > repeat trials in inferior parietal lobe, superior frontal gyrus, inferior parietal lobe, fusiform gyrus, temporal lobe subgyral, cerebellum

(*Continued*)

Table 1 (*Continued*)

Study	Number of subjects	Age range (years)	Neuroimaging modality	Cognitive flexibility task	Behavioral results	Contrast	Brain region differences between children and adults	Brain regions activated in all groups
Yerys et al. 2015	19 NT children	7–14	fMRI	Set-shifting (spatial)	Accuracy was greater than 95% in children	NA	NA	MFG/frontal pole, right lateral SFG/precentral gyrus, bilateral putamen, mdACC/SMA, medial SFG, insula, bilateral superior parietal lobule, precuneus, cuneus, cerebellum, L lateral SFG/precentral gyrus, L hippocampus, L fusiform/inferior temporal/ parahippocampal area, occipital regions
Mogadam et al. 2018	22 NT children	8–15	MEG	Set-shifting	No age differences in accuracy of RT, accuracy > 89%	Shifting > control	SMG (supramarginal gyrus) showed a negative correlation between peak latency and age	Superior/inferior R parietal, L parietal, R prefrontal association cortex, L prefrontal areas (dl/vlPFC, Broca's area), insula, supplementary and premotor cortices, R PCC, L temporal pole
Dirks et al. 2020	36 NT children	7–12	fMRI	Attention-shifting	Accuracy was greater than 85% in children	None	TD children showed activation in the posterior SMG/ANG during shift trials	Posterior SMG/ANG
Wendelken et al. 2012	20 children, 16 adults	8–13, 20-27	fMRI	Rule-switching	Adults were more accurate than children Adults were faster than children	None	Adults had greater activation than children in pre-SMA, L PMC, R PMC, L IPL, L MOG, R MOG, and R MOG Children had greater activation than adults in the IPL, cingulate gyrus In the group × switch interaction, children engaged regions of L STG and R MTG during switch trials	dlPFC, L PPC, and pre-SMA

(*Continued*)

Table 1 (*Continued*)

Study	Number of subjects	Age range (years)	Neuroimaging modality	Cognitive flexibility task	Behavioral results	Contrast	Brain region differences between children and adults	Brain regions activated in all groups
Crone et al. 2006a	17 children, 17 adolescents, 20 adults	8–12, 13–17, 18–25	fMRI	Rule-switching	Adults were faster and had fewer errors than adolescents and children Adolescents were faster and had fewer errors than children All groups had similar univalent switch costs (univalent switches minus univalent repetition) Adults had fewer bivalent switch costs than adolescents and children and adolescents had fewer than children (bivalent switches minus bivalent repetition)	None	Children had greater vlPFC activation during switch trials Adults had similar activation to adolescents and both groups had greater activation than children in pre-SMA/SMA during bivalent switch greater than in repetition trials	vlPFC, pre-SMA/SMA, L superior parietal (ROI)
Dibbets et al. 2006	7 TD children	6 years, 10 months (mean)	fMRI	Switch task for children	Children were slower on the switch than nonswitch condition	None	NA	R temporal areas, R frontal areas, L postcentral gyrus, L parietal lobe, L hippocampus
Nelson et al. 2007	17 TD adolescents	8–17	fMRI	Change task	52.1% change trial accuracy	Change trials > go trials Change trials > baseline fixation	NA	Deactivated dlPFC, activated motor cortex
Taylor et al. 2012	26 TD children	7–14	fMRI	Set-shifting	NA	None	NA	Right insula activation increased with age
Crone et al. 2008	17 children, 20 adolescents, 20 adults	8–11, 14–15, 18–24	fMRI	Performance-based switching	Children had fewer rule shifts, fewer efficient errors, and more performance errors than adolescents and adults Adolescents had similar performance as adults but had more performance errors	None	Adults had similar activation to adolescents in the lateral OFC/insula, anterior prefrontal cortex, medial PFC/ACC, and superior and inferior parietal cortex Children had fewer areas of activation than adults/adolescents	dlPFC, medial PFC/ACC, lateral OFC/insula, superior and inferior parietal cortex

(*Continued*)

Table 1 (*Continued*)

Study	Number of subjects	Age range (years)	Neuroimaging modality	Cognitive flexibility task	Behavioral results	Contrast	Brain region differences between children and adults	Brain regions activated in all groups
van den Bos et al. 2012	18 children, 27 adolescents, 22 adults	8–11, 13–16, 18–22	fMRI	Performance-based switching	Age-related decrease in learning rates from negative feedback Marginal age-related increase in learning rates from positive feedback	Positive > negative feedback	Age-related increases in FC of the ventral striatum with mPFC	Enhanced FC between the bilateral ventral striatum seed and mPFC
Hauser et al. 2015	19 adolescents, 17 adults	12–16, 20–29	fMRI	Performance-based switching	Adults performed similarly to adolescents on task Adolescents had a greater learning rate for negative RPEs than adults	None	Adolescents had greater activation for decreasing RPEs in the anterior insula	L putamen, L vmPFC, amygdala, L PCC, R precentral gyrus, L SFG, L IPL, L mPFC
D'Cruz et al. 2016	23 NT children	8.4 years (mean)	fMRI	Performance-based switching	Perseverative errors equaled 0.5 (mean) Failures to maintain set equaled 3.0 (mean)	Four-choice reversal learning task	NA	Ventral striatum, thalamus, insula, motor, and affective subdivision of anterior cingulate, dlPFC, premotor cortex, pre-SMA, PPC, primary visual cortex, lateral extrastriate cortex and precuneus, L cognitive subdivision of ACC, L caudate, L orbitofrontal cortex

Studies were identified through systematic searches in PubMed and Google Scholar with the search term "'shift' OR 'switch' AND 'children' OR 'adult' AND 'cognitive flexibility' OR 'cognitive control' AND 'fMRI'." Studies comparing children or adolescents to adults, only children or adolescents, or NT children to children with a neurodevelopmental disorder were included. However, only the results for NT children are depicted if provided. Additionally, studies using various neuroimaging modalities such as fMRI, MEG, and fNIRS were included. Abbreviations: ACC, anterior cingulate cortex; ANG, angular gyrus; CCN, cognitive control network; dlPFC, dorsolateral prefrontal cortex; FC, frontal cortex; fMRI, functional magnetic resonance imaging; fNIRS, functional near-infrared spectroscopy; IFJ, inferior frontal junction; IPL, inferior parietal lobe; L, left; mdACC, mid-dorsal anterior cingulate cortex; MEG, magnetoencephalography; MFG, middle frontal gyrus; MOG, middle occipital gyrus; mPFC, medial prefrontal cortex; MTG, middle temporal gyrus; NA, not applicable; NT, neurotypical; OFC, orbitofrontal cortex; oxy-Hb, oxyhemoglobin; PCC, posterior cingulate cortex; PFC, prefrontal cortex; PMC, premotor cortex; PPC, posterior parietal cortex; R, right; ROI, region of interest; RPEs, reward prediction errors; RT, reaction time; SFG, superior frontal gyrus; SMA, supplementary motor area; SMG, supramarginal gyrus; STG, superior temporal gyrus; TD, typical development; vlPFC, ventrolateral prefrontal cortex; vmPFC, ventromedial prefrontal cortex.

was used. The authors found typically developing children (7–12 years) had brain activation in the L posterior supramarginal gyrus/angular gyrus during shift trials. The discrepancies among the studies involving attentional-shift tasks may be due to small sample sizes and differences in the shifting tasks used. It has been suggested that these types of simple attention-shifting tasks may not strongly engage cognitive flexibility (Dirks et al. 2020).

Complex set-shifting. Complex set-shifting tasks require different brain regions to complete compared with lower-level shifting tasks. To our knowledge, only one study to date has examined set-shifting abilities in children using a WCST task. Children showed activation in the right insula, a region important for switching between brain networks to enable flexible behavior (Menon & Uddin 2010), with increasing activation with age (Taylor et al. 2012). Only one study in adults has used the FIST, a complex version of a set-shifting task that requires subjects to abstract a matching dimension and switch flexibly to a new matching dimension (Dajani et al. 2020, Dick 2014). Although the neural correlates of the FIST have been studied only in adults, behavioral evidence in children reveals age-related changes until at least 10 years of age, suggesting that there may be age-related neural differences in set-shifting at least until 10 years (Dick 2014).

Rule-switching. Rule-switching has also been explored in few developmental neuroimaging studies to date. The earliest study found adolescents had similar brain activation as adults among regions of the pre-SMA/SMA during rule-switching (Crone et al. 2006a). Wendelken and colleagues (2012) utilized a task that required participants to switch flexibly from one task rule to another. In children (8–13 years) and adults, brain regions of the left dlPFC, left posterior parietal cortex (PPC), and pre-SMA regions were active during rule-switching trials. The dlPFC and SMA were similarly activated as in other cognitive flexibility studies. Further, there were key developmental differences seen between children and adults in brain activation during the task. Adults had greater activation overall and among the pre-SMA, PMC, and left PPC. Children also had greater activation than adults in the right inferior parietal lobe and the cingulate gyrus. Additionally, children, but not adults, engaged the left superior temporal gyrus and the right middle temporal gyrus more in the switch trials as compared with the non-switch trials. Children had more initial dlPFC activation driven by the previous trial rule, suggesting children have more difficulties with rule-switching in particular (Wendelken et al. 2012). Rule-switching may therefore create greater cognitive demands and may be more difficult for children compared with adults, accounting for the observed neural differences.

Performance-based switching. Performance-based switching studies have revealed many behavioral and neurodevelopmental differences between children and adults (Crone et al. 2008, Hauser et al. 2015, van den Bos et al. 2012). The insula appears to be an important contributor to performance-based switching, as indicated by activation during the WCST-adapted task (Crone et al. 2008), especially in adolescents, where greater activation occurred in the anterior insula for reward prediction errors prior to switching. This is also in line with findings from a meta-analysis of developmental executive functions, where the right anterior insular cortex was shown to have age-related involvement among inhibition, switching, and working memory (Houdé et al. 2010). Other regions were important during performance-switching including the dlPFC, medial PFC (mPFC), anterior cingulate cortex (ACC), striatum, ventromedial PFC, amygdala, left posterior cingulate cortex, left putamen, right precentral gyrus, left superior frontal gyrus, and left inferior parietal lobe. Activation in the mPFC was commonly observed across the three developmental studies, consistent with reports of the role of the PFC in cognitive flexibility (Rougier et al. 2005).

Summary of neurodevelopmental findings. The neurodevelopmental studies of cognitive flexibility provide initial insight into the developmental processes surrounding switching and shifting, abilities important for life outcomes. The brain regions commonly implicated during switching or shifting across task types in children include the dlPFC and the pre-SMA/SMA. The dlPFC is one region of the FPN involved in switching in adults and is primarily thought to be involved with working memory (Thomason et al. 2009) and, specifically, generating the executive response guided by the working memory (Barbey et al. 2013, D'Esposito & Postle 1999). This indicates that children rely on working memory to complete flexibility tasks. Moreover, in cognitive flexibility tasks with higher loads of working memory, children typically perform less accurately than adults (Thomason et al. 2009, Wendelken et al. 2012), suggesting that the integration of working memory and switching processes is not fully developed during childhood. The pre-SMA/SMA was also discovered to be activated in children across multiple developmental cognitive flexibility studies. The pre-SMA is commonly activated during set-shifting tasks (Barber & Carter 2005) in adults and, more broadly, during executive functioning tasks in general (Duncan & Owen 2000). Overall, the pre-SMA is thought to be involved in task-set reconfiguration, suppression of previous responses, and error likelihood estimation (Morton et al. 2009).

Another set of regions commonly observed in flexibility tasks in adults includes the dorsal ACC and AI, comprising the midcingulo-insular network (M-CIN). There were mixed findings surrounding the M-CIN among developmental studies, with some instances of increased activation of the insula in children (Ezekiel et al. 2013, Mogadam et al. 2018, Rodehacke et al. 2014) and, conversely, some instances of decreased or no activation of the insula during switching (Crone et al. 2006a, 2008; Dibbets et al. 2006; Dirks et al. 2020; Nelson et al. 2007; Taylor et al. 2012; Wendelken et al. 2012). The M-CIN supports coordination among large-scale networks and is thought to enable flexible behavior (Uddin et al. 2015). The mixed findings regarding the M-CIN brain regions in children may be due to the various tasks (i.e., attention shifting versus performance switching) or varying age groups across the studies (i.e., from 3 years to 17 years). In most cases, children and adolescents were separately grouped, and generally, adolescents showed more insula activation than children, suggesting an age-related increase in activation of the insula (Taylor et al. 2012) associated with better behavioral performance (Chen et al. 2016). The insula plays an important role in executive function and shifting broadly. The AI in particular is thought to coordinate shifting between the default mode and executive control networks and serves as a gatekeeper of executive control (Molnar-Szakacs & Uddin 2022). Overall, the studies reviewed here support the notion that the brain regions involved in the salience (M-CIN) and the executive control (FPN) networks may support the development and maturation of cognitive flexibility (**Figure 4**).

The developmental neuroimaging studies reviewed also provided insight into the regions that were not commonly activated in children during cognitive flexibility tasks. For example, only one study found evidence of activation of the IFJ in both children and adults (Morton et al. 2009). The IFJ has consistently been found to be activated across various cognitive flexibility tasks in adults (Dajani et al. 2020, Kim et al. 2011). The IFJ was additionally found to coordinate switches among brain regions during cognitive flexibility task performance (Dajani et al. 2020). As demonstrated by Dajani and colleagues (2020), the IFJ gets activated first and leads to activations in other regions involved in cognitive flexibility. As this region was not commonly activated in cognitive flexibility studies of children, this suggests that the ability to coordinate switching undergoes developmental changes. Taken together, these studies provide valuable insight into the development of cognitive flexibility. Still, many questions remain to be answered, including how the functional organization of these brain regions enables changes in cognitive flexibility across development.

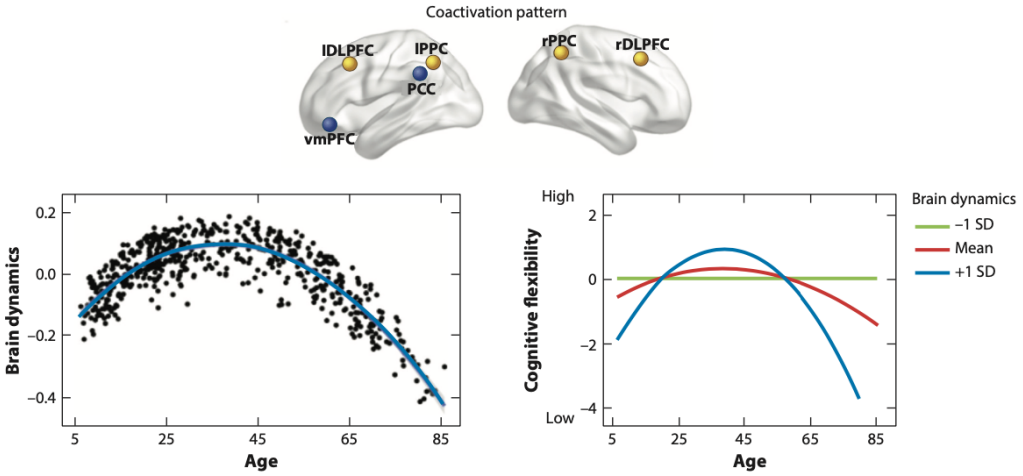

Figure 4

Relationships between brain dynamics and cognitive flexibility across the life span. A coactivation pattern involving the default mode and FPN shows increasing frequency of occurrence into young adulthood, decreasing again during older adulthood. Brain dynamics were differentially associated with cognitive flexibility across the life span such that children and older adults who had fewer brain state transitions showed poorer cognitive flexibility, whereas middle-aged adults had optimal cognitive flexibility at average and fewer brain state transitions (Kupis et al. 2021b, Molnar-Szakacs & Uddin 2022). Abbreviations: FPN, frontoparietal network; lDLPFC, left dorsolateral prefrontal cortex; lPPC, left posterior parietal cortex; PCC, posterior cingulate cortex; rDLPFC, right dorsolateral prefrontal cortex; rPPC; right posterior parietal cortex; SD, standard deviation; vmPFC, ventromedial prefrontal cortex.

Limitations of Neurodevelopmental Studies of Cognitive Flexibility: Four Challenges

Although the neurodevelopmental studies reviewed here provide initial insight into the neural mechanisms underlying cognitive flexibility, caution should be used when drawing conclusions from the results. First, the idea that executive functioning constructs are separable yet highly correlated (Miyake & Friedman 2012, Rodríguez-Nieto et al. 2022) makes the study of cognitive flexibility difficult, as it is uncertain if the studies reviewed strictly captured cognitive flexibility, other components of executive functions (e.g., working memory), or executive function as a whole. Additionally, many brain regions are active across various executive functions (Nowrangi et al. 2014), thereby making interpretations of what is uniquely accounted for by cognitive flexibility difficult. Similarly, since various tasks elicit cognitive flexibility, it makes studying the construct difficult. For example, both attention-shifting and rule-shifting are thought to elicit cognitive flexibility; however, some think attention-shifting is a poor indicator of cognitive flexibility (Dirks et al. 2020). In addition, many of the studies reviewed above have small sample sizes and potentially confounding task designs that do not consistently isolate cognitive flexibility due to poorly designed control tasks (Marek et al. 2022, Zhao et al. 2023). Additionally, the tasks differ on the degree of cognitive effort required, often engaging varying levels of working memory demands. Lastly, there is controversy regarding the ecological validity of some of the cognitive flexibility tasks used as they do not always correlate with informant reports (Dang et al. 2020), suggesting they may not be representative of real-world situations (Kenworthy et al. 2008). Overall, the studies reviewed provide initial insight into the development of cognitive flexibility, but these findings should be considered in the context of these limitations.

FUTURE DIRECTIONS

Many questions remain, including how dynamic brain network organization or neural flexibility enables changes in cognitive flexibility across development. Future directions include studying cognitive flexibility across development using tasks that better capture the cognitive flexibility construct and have higher ecological validity. Additionally, it will be necessary to further investigate different potential forms of cognitive flexibility to disentangle the brain regions involved with each type. It will also be necessary to study larger samples in future work to arrive at reproducible results. Future studies may leverage large, public databases, such as the Adolescent Brain Cognitive Development (ABCD) Study, which includes task and resting-state data and out-of-scanner executive control measures to better characterize neural changes associated with developing cognitive flexibility (Casey et al. 2018). As the ABCD Study provides longitudinal data from over 10,000 children, it will be critical to leverage in order to gain insight into individual differences in cognitive flexibility as they relate to the development of key brain networks. Further, examination of additional factors that may modulate the relationship between cognitive flexibility and brain functioning should be explored, such as health factors (Cserjési et al. 2007), exercise (Bae & Masaki 2019), bilingualism (Becker et al. 2016, Christoffels et al. 2015), and neuropsychiatric disorders (Uddin 2021b). There is evidence that these factors may modulate or interact with executive function, yet the impacts on shifting and flexibility (specifically) are not fully understood. Further, studying modulators of shifting and cognitive flexibility skills may provide insight into novel intervention targets. Additionally, adolescence is a unique period where cognitive flexibility is heightened, the brain is undergoing many changes (Hauser et al. 2015), and executive function skills are stabilizing (Friedman et al. 2016) (see **Figure 4**), making it a critical time period to study.

Another important future direction is the examination of intrinsic functional connectivity dynamics and developmental changes within and across brain networks supporting cognitive flexibility. Studies have begun to examine how brain networks dynamically act together to support neural and cognitive flexibility (Kupis et al. 2020, 2021a,b; Uddin 2021a,b). Evidence from these studies supports the idea that connectivity within the salience (M-CIN) and executive control (FPN) networks may underlie the development of cognitive flexibility into young adulthood (Kupis et al. 2021b) (**Figure 4**). Further, certain brain dynamic patterns measured by the WCST have been associated with cognitive flexibility such that individuals exhibiting greater episodes of more frequently occurring brain states showed greater cognitive flexibility (Nomi et al. 2017b). Brain dynamics within the M-CIN and between the medial and left FPN have also been associated with cognitive flexibility (for a review see Uddin 2021a). Future investigation needs to include task-based fMRI approaches to map brain dynamics during a cognitive flexibility task and changes across development.

Overall, neurodevelopmental findings of cognitive flexibility have various limitations, and many questions are left to be answered regarding brain network flexibility and moderators of cognitive flexibility and brain functioning across development.

CONCLUSION

Cognitive flexibility is challenging to study, as various tasks are used to elicit it. Emerging evidence suggests that different brain regions are associated with different types of cognitive flexibility tasks. Different brain regions are also engaged as a result of varying degrees of difficulty across task types and inconsistent task instructions across studies. However, key brain regions support the development of cognitive flexibility broadly, including the dlPFC, pre-SMA/SMA, IFJ, and the insula. Large-scale brain networks such as the salience (M-CIN), executive (FPN), and default mode networks additionally support the development of cognitive flexibility. These core networks enhance

behavioral performance through salience detection, performance monitoring, attention, and control (Menon & Uddin 2010). Initial work suggests that structural and functional maturation of the M-CIN underlies the development of flexibility (Uddin et al. 2011). Future work is needed to expand upon cognitive flexibility as a construct, determine the neural impacts of its potential modulators, and investigate brain dynamics associated with shifting abilities across development.

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LITERATURE CITED

Anderson P. 2002. Assessment and development of executive function (EF) during childhood. *Child Neuropsychol.* 8(2):71–82

Bae S, Masaki H. 2019. Effects of acute aerobic exercise on cognitive flexibility required during task-switching paradigm. *Front. Hum. Neurosci.* 13:260

Barber AD, Carter CS. 2005. Cognitive control involved in overcoming prepotent response tendencies and switching between tasks. *Cereb. Cortex* 15(7):899–912

Barbey AK, Koenigs M, Grafman J. 2013. Dorsolateral prefrontal contributions to human working memory. *Cortex* 49(5):1195–205

Becker TM, Prat CS, Stocco A. 2016. A network-level analysis of cognitive flexibility reveals a differential influence of the anterior cingulate cortex in bilinguals versus monolinguals. *Neuropsychologia* 85:62–73

Burt KB, Paysnick AA. 2012. Resilience in the transition to adulthood. *Dev. Psychopathol.* 24(2):493–505

Casey BJ, Cannonier T, Conley MI, Cohen AO, Barch DM, et al. 2018. The Adolescent Brain Cognitive Development (ABCD) study: imaging acquisition across 21 sites. *Dev. Cogn. Neurosci.* 32:43–54

Casey BJ, Davidson MC, Hara Y, Thomas KM, Martinez A, et al. 2004. Early development of subcortical regions involved in non-cued attention switching. *Dev. Sci.* 7(5):534–42

Casey BJ, Giedd JN, Thomas KM. 2000. Structural and functional brain development and its relation to cognitive development. *Biol. Psychol.* 54(1–3):241–57

Cepeda NJ, Kramer AF, Gonzalez de Sather JC. 2001. Changes in executive control across the life span: examination of task-switching performance. *Dev. Psychol.* 37(5):715–30

Chelune GJ, Baer RA. 1986. Developmental norms for the Wisconsin Card Sorting test. *J. Clin. Exp. Neuropsychol.* 8(3):219–28

Chen T, Cai W, Ryali S, Supekar K, Menon V. 2016. Distinct global brain dynamics and spatiotemporal organization of the salience network. *PLOS Biol.* 14(6):e1002469

Christakou A, Halari R, Smith AB, Ifkovits E, Brammer M, Rubia K. 2009. Sex-dependent age modulation of frontostriatal and temporo-parietal activation during cognitive control. *NeuroImage* 48(1):233–36

Christoffels IK, de Haan AM, Steenbergen L, van den Wildenberg WPM, Colzato LS. 2015. Two is better than one: Bilingual education promotes the flexible mind. *Psychol. Res.* 79:371–79

Côté SM, Boivin M, Liu X, Nagin DS, Zoccolillo M, Tremblay RE. 2009. Depression and anxiety symptoms: onset, developmental course and risk factors during early childhood. *J. Child Psychol. Psychiatry* 50(10):1201–8

Crone EA, Donohue SE, Honomichl R, Wendelken C, Bunge SA. 2006a. Brain regions mediating flexible rule use during development. *J. Neurosci.* 26(43):11239–47

Crone EA, Wendelken C, Donohue SE, Bunge SA. 2006b. Neural evidence for dissociable components of task-switching. *Cereb. Cortex.* 16(4):475–86

Crone EA, Zanolie K, Van Leijenhorst L, Westenberg PM, Rombouts SARB. 2008. Neural mechanisms supporting flexible performance adjustment during development. *Cogn. Affect. Behav. Neurosci.* 8(2):165–77

Cserjési R, Molnár D, Luminet O, Lénárd L. 2007. Is there any relationship between obesity and mental flexibility in children? *Appetite* 49(3):675–78

Dajani DR, Odriozola P, Winters M, Voorhies W, Marcano S, et al. 2020. Measuring cognitive flexibility with the flexible item selection task: from fMRI adaptation to individual connectome mapping. *J. Cogn. Neurosci.* 32(6):1026–45

Dajani DR, Uddin LQ. 2015. Demystifying cognitive flexibility: implications for clinical and developmental neuroscience. *Trends Neurosci.* 38(9):571–78

Dang J, King KM, Inzlicht M. 2020. Why are self-report and behavioral measures weakly correlated? *Trends Cogn. Sci.* 24(4):267–69

Davidson MC, Amso D, Anderson LC, Diamond A. 2006. Development of cognitive control and executive functions from 4 to 13 years: evidence from manipulations of memory, inhibition, and task switching. *Neuropsychologia* 44(11):2037–78

D'Cruz A-M, Mosconi MW, Ragozzino ME, Cook EH, Sweeney JA. 2016. Alterations in the functional neural circuitry supporting flexible choice behavior in autism spectrum disorders. *Transl. Psychiatry* 6(10):e916

Delis DC, Kaplan E, Kramer JH. 2001. *Delis-Kaplan Executive Function System*. San Antonio, TX: Psychol. Corp.

D'Esposito M, Postle BR. 1999. The dependence of span and delayed-response performance on prefrontal cortex. *Neuropsychologia* 37(11):1303–15

Deveney CM, Deldin PJ. 2006. A preliminary investigation of cognitive flexibility for emotional information in major depressive disorder and non-psychiatric controls. *Emotion* 6(3):429–37

Diamond A. 2013. Executive functions. *Annu. Rev. Psychol.* 64:135–68

Diamond A, Carlson SM, Beck DM. 2005. Preschool children's performance in task switching on the dimensional change card sort task: separating the dimensions aids the ability to switch. *Dev. Neuropsychol.* 28(2):689–729

Dibbets P, Bakker K, Jolles J. 2006. Functional MRI of task switching in children with Specific Language Impairment (SLI). *Neurocase* 12(1):71–79

Dick AS. 2014. The development of cognitive flexibility beyond the preschool period: an investigation using a modified Flexible Item Selection Task. *J. Exp. Child Psychol.* 125:13–34

Dirks B, Romero C, Voorhies W, Kupis L, Nomi JS, et al. 2020. Neural responses to a putative set-shifting task in children with Autism Spectrum Disorder. *Autism Res.* 13(9):1501–15

Duncan J, Owen AM. 2000. Common regions of the human frontal lobe recruited by diverse cognitive demands. *Trends Neurosci.* 23(10):475–83

Eslinger PJ, Grattan LM. 1993. Frontal lobe and frontal-striatal substrates for different forms of human cognitive flexibility. *Neuropsychologia* 31(1):17–28

Ezekiel F, Bosma R, Morton JB. 2013. Dimensional Change Card Sort performance associated with age-related differences in functional connectivity of lateral prefrontal cortex. *Dev. Cogn. Neurosci.* 5:40–50

Friedman NP, Miyake A, Altamirano LJ, Corley RP, Young SE, et al. 2016. Stability and change in executive function abilities from late adolescence to early adulthood: a longitudinal twin study. *Dev. Psychol.* 52(2):326–40

Genet JJ, Malooly AM, Siemer M. 2013. Flexibility is not always adaptive: affective flexibility and inflexibility predict rumination use in everyday life. *Cogn. Emot.* 27(4):685–95

Genet JJ, Siemer M. 2011. Flexible control in processing affective and non-affective material predicts individual differences in trait resilience. *Cogn. Emot.* 25(2):380–88

Giedd JN, Blumenthal J, Jeffries NO, Castellanos FX, Liu H, et al. 1999. Brain development during childhood and adolescence: a longitudinal MRI study. *Nat. Neurosci.* 2:861–63

Gogtay N, Giedd JN, Lusk L, Hayashi KM, Greenstein D, et al. 2004. Dynamic mapping of human cortical development during childhood through early adulthood. *PNAS* 101(21):8174–79

Hanania R, Smith LB. 2010. Selective attention and attention switching: towards a unified developmental approach. *Dev. Sci.* 13(4):622–35

Hauser TU, Iannaccone R, Walitza S, Brandeis D, Brem S. 2015. Cognitive flexibility in adolescence: neural and behavioral mechanisms of reward prediction error processing in adaptive decision making during development. *NeuroImage* 104:347–54
Heaton RK, Chelune GJ, Talley JL, Kay GG, Curtiss G. 1993. *Wisconsin Card Sorting Test Manual: Revised and Expanded*. Odessa, FL: Psychol. Assess.
Houdé O, Rossi S, Lubin A, Joliot M. 2010. Mapping numerical processing, reading, and executive functions in the developing brain: an fMRI meta-analysis of 52 studies including 842 children. *Dev. Sci.* 13(6):876–85
Hughes C. 1998. Executive function in preschoolers: links with theory of mind and verbal ability. *Br. J. Dev. Psychol.* 16(2):233–53
Ionescu T. 2012. Exploring the nature of cognitive flexibility. *New Ideas Psychol.* 30(2):190–200
Izquierdo A, Brigman JL, Radke AK, Rudebeck PH, Holmes A. 2017. The neural basis of reversal learning: an updated perspective. *Neuroscience* 345:12–26
Jacques S, Zelazo PD. 2001. The Flexible Item Selection Task (FIST): a measure of executive function in preschoolers. *Dev. Neuropsychol.* 20(3):573–91
Johansson M, Marciszko C, Gredebäck G, Nyström P, Bohlin G. 2015. Sustained attention in infancy as a longitudinal predictor of self-regulatory functions. *Infant Behav. Dev.* 41:1–11
Keenan EG, Gotham K, Lerner MD. 2018. Hooked on a feeling: repetitive cognition and internalizing symptomatology in relation to autism spectrum symptomatology. *Autism* 22(7):814–24
Kehagia AA, Murray GK, Robbins TW. 2010. Learning and cognitive flexibility: frontostriatal function and monoaminergic modulation. *Curr. Opin. Neurobiol.* 20(2):199–204
Kenworthy L, Yerys BE, Anthony LG, Wallace GL. 2008. Understanding executive control in autism spectrum disorders in the lab and in the real world. *Neuropsychol. Rev.* 18(4):320–38
Kim C, Cilles SE, Johnson NF, Gold BT. 2012. Domain general and domain preferential brain regions associated with different types of task switching: a meta-analysis. *Hum. Brain Mapp.* 33(1):130–42
Kim C, Johnson NF, Cilles SE, Gold BT. 2011. Common and distinct mechanisms of cognitive flexibility in prefrontal cortex. *J. Neurosci.* 31(13):4771–79
Kraft D, Rademacher L, Eckart C, Fiebach C. 2020. Cognitive, affective, and feedback-based flexibility—disentangling shared and different aspects of three facets of psychological flexibility. *J. Cogn.* 3(1):21
Konishi S, Nakajima K, Uchida I, Kameyama M, Nakahara K, et al. 1998. Transient activation of inferior prefrontal cortex during cognitive set shifting. *Nat. Neurosci.* 1(1):80–84
Kupis L, Goodman ZT, Kircher L, Romero C, Dirks B, et al. 2021a. Altered patterns of brain dynamics linked with body mass index in youth with autism. *Autism Res.* 14(5):873–86
Kupis L, Goodman ZT, Kornfeld S, Hoang S, Romero C, et al. 2021b. Brain dynamics underlying cognitive flexibility across the lifespan. *Cereb. Cortex.* 31(11):5263–74
Kupis L, Romero C, Dirks B, Hoang S, Parladé MV, et al. 2020. Evoked and intrinsic brain network dynamics in children with autism spectrum disorder. *NeuroImage Clin.* 28:102396
Logue SF, Gould TJ. 2014. The neural and genetic basis of executive function: attention, cognitive flexibility, and response inhibition. *Pharmacol. Biochem. Behav.* 123:45–54
Lu JG, Akinola M, Mason MF. 2017. "Switching on" creativity: Task switching can increase creativity by reducing cognitive fixation. *Organ. Behav. Hum. Decis. Process.* 139:63–75
Lu JG, Martin AE, Usova A, Galinsky AD. 2019. Creativity and humor across cultures: where aha meets haha. In *Creativity and Humor*, ed. SR Luria, J Baer, JC Kaufman, pp. 183–203. London: Acad. Press
Luna B. 2009. Developmental changes in cognitive control through adolescence. *Adv. Child Dev. Behav.* 37:233–78
Marek S, Tervo-Clemmens B, Calabro FJ, Montez DF, Kay BP, et al. 2022. Reproducible brain-wide association studies require thousands of individuals. *Nature* 603(7902):654–60
McDougle CJ, Kresch LE, Goodman WK, Naylor ST, Volkmar FR, et al. 1995. A case-controlled study of repetitive thoughts and behavior in adults with autistic disorder and obsessive-compulsive disorder. *Am. J. Psychiatry* 152(5):772–77
Menon V, Uddin LQ. 2010. Saliency, switching, attention and control: a network model of insula function. *Brain Struct. Funct.* 214(5–6):655–67
Miyake A, Friedman NP. 2012. The nature and organization of individual differences in executive functions: four general conclusions. *Curr. Dir. Psychol. Sci.* 21(1):8–14

Miyake A, Friedman NP, Emerson MJ, Witzki AH, Howerter A, Wager TD. 2000. The unity and diversity of executive functions and their contributions to complex "frontal lobe" tasks: a latent variable analysis. *Cogn. Psychol.* 41(1):49–100

Mogadam A, Keller AE, Taylor MJ, Lerch JP, Anagnostou E, Pang EW. 2018. Mental flexibility: an MEG investigation in typically developing children. *Brain Cogn*. 120:58–66

Molnar-Szakacs I, Uddin LQ. 2022. Anterior insula as a gatekeeper of executive control. *Neurosci. Biobehav. Rev.* 139:104736

Monsell S. 2003. Task switching. *Trends Cogn. Sci.* 7(3):134–40

Moriguchi Y, Hiraki K. 2009. Neural origin of cognitive shifting in young children. *PNAS* 106(14):6017–21

Morton JB, Bosma R, Ansari D. 2009. Age-related changes in brain activation associated with dimensional shifts of attention: an fMRI study. *NeuroImage* 46(1):249–56

Nelson EE, Vinton DT, Berghorst L, Towbin KE, Hommer RE, et al. 2007. Brain systems underlying response flexibility in healthy and bipolar adolescents: an event-related fMRI study. *Bipolar Disord.* 9(8):810–19

Niendam TA, Laird AR, Ray KL, Dean YM, Glahn DC, Carter CS. 2012. Meta-analytic evidence for a superordinate cognitive control network subserving diverse executive functions. *Cogn. Affect. Behav. Neurosci.* 12(2):241–68

Nomi JS, Bolt TS, Ezie CEC, Uddin LQ, Heller AS. 2017a. Moment-to-moment BOLD signal variability reflects regional changes in neural flexibility across the lifespan. *J. Neurosci.* 37(22):5539–48

Nomi JS, Vij SG, Dajani DR, Steimke R, Damaraju E, et al. 2017b. Chronnectomic patterns and neural flexibility underlie executive function. *NeuroImage* 147:861–71

Nowrangi MA, Lyketsos C, Rao V, Munro CA. 2014. Systematic review of neuroimaging correlates of executive functioning: converging evidence from different clinical populations. *J. Neuropsychiatry Clin. Neurosci.* 26(2):114–25

Rodehacke S, Mennigen E, Müller KU, Ripke S, Jacob MJ, et al. 2014. Interindividual differences in mid-adolescents in error monitoring and post-error adjustment. *PLOS ONE* 9(2):e88957

Rodríguez-Nieto G, Seer C, Sidlauskaite J, Vleugels L, Van Roy A, et al. 2022. Inhibition, shifting and updating: inter and intra-domain commonalities and differences from an executive functions activation likelihood estimation meta-analysis. *NeuroImage* 264:119665

Rose EJ, Picci G, Fishbein DH. 2019. Neurocognitive precursors of substance misuse corresponding to risk, resistance, and resilience pathways: implications for prevention science. *Front. Psychiatry* 10:399

Rosselli M, Ardila A. 1993. Developmental norms for the Wisconsin Card Sorting Test in 5-to 12-year-old children. *Clin. Neuropsychol.* 7(2):145–54

Rougier NP, Noelle DC, Braver TS, Cohen JD, O'Reilly RC. 2005. Prefrontal cortex and flexible cognitive control: rules without symbols. *PNAS* 102(20):7338–43

Rubia K, Smith AB, Woolley J, Nosarti C, Heyman I, et al. 2006. Progressive increase of frontostriatal brain activation from childhood to adulthood during event-related tasks of cognitive control. *Hum. Brain Mapp.* 27(12):973–93

Shinya Y, Kawai M, Niwa F, Kanakogi Y, Imafuku M, Myowa M. 2022. Cognitive flexibility in 12-month-old preterm and term infants is associated with neurobehavioural development in 18-month-olds. *Sci. Rep.* 12(1):3

Taylor MJ, Donner EJ, Pang EW. 2012. fMRI and MEG in the study of typical and atypical cognitive development. *Neurophysiol. Clin.* 42(1–2):19–25

Thomason ME, Race E, Burrows B, Whitfield-Gabrieli S, Glover GH, Gabrieli JDE. 2009. Development of spatial and verbal working memory capacity in the human brain. *J. Cogn. Neurosci.* 21(2):316–32

Uddin LQ. 2021a. Cognitive and behavioural flexibility: neural mechanisms and clinical considerations. *Nat. Rev. Neurosci.* 22(3):167–79

Uddin LQ. 2021b. Brain mechanisms supporting flexible cognition and behavior in adolescents with Autism Spectrum Disorder. *Biol. Psychiatry* 89(2):172–83

Uddin LQ, Supekar K, Lynch CJ, Cheng KM, Odriozola P, et al. 2015. Brain state differentiation and behavioral inflexibility in autism. *Cereb. Cortex.* 25(12):4740–47

Uddin LQ, Supekar KS, Ryali S, Menon V. 2011. Dynamic reconfiguration of structural and functional connectivity across core neurocognitive brain networks with development. *J. Neurosci.* 31(50):18578–89

van den Bos W, Cohen MX, Kahnt T, Crone EA. 2012. Striatum-medial prefrontal cortex connectivity predicts developmental changes in reinforcement learning. *Cereb. Cortex*. 22(6):1247–55

van Steensel FJA, Bögels SM, Perrin S. 2011. Anxiety disorders in children and adolescents with autistic spectrum disorders: a meta-analysis. *Clin. Child Fam. Psychol. Rev.* 14(3):302–17

Vandierendonck A, Liefooghe B, Verbruggen F. 2010. Task switching: interplay of reconfiguration and interference control. *Psychol. Bull.* 136(4):601–26

Welsh MC, Pennington BF, Groisser DB. 1991. A normative-developmental study of executive function: a window on prefrontal function in children. *Dev. Neuropsychol.* 7(2):131–49

Wendelken C, Munakata Y, Baym C, Souza M, Bunge SA. 2012. Flexible rule use: common neural substrates in children and adults. *Dev. Cogn. Neurosci.* 2(3):329–39

Whitmer AJ, Banich MT. 2007. Inhibition versus switching deficits in different forms of rumination. *Psychol. Sci.* 18(6):546–53

Yasumura A, Yamamoto H, Yasumura Y, Moriguchi Y, Hiraki K, et al. 2015. Cognitive shifting in children with attention-deficit hyperactivity disorder: a near infrared spectroscopy study. *Afr. J. Psychiatry* 18(1):196

Yeniad N, Malda M, Mesman J, van IJzendoorn MH, Pieper S. 2013. Shifting ability predicts math and reading performance in children: a meta-analytical study. *Learn. Individ. Differ.* 23:1–9

Yerys BE, Antezana L, Weinblatt R, Jankowski KF, Strang J, et al. 2015. Neural correlates of set-shifting in children with autism. *Autism Res.* 8(4):386–97

Zelazo PD. 2006. The Dimensional Change Card Sort (DCCS): a method of assessing executive function in children. *Nat. Protoc.* 1(1):297–301

Zhao W, Makowski C, Hagler DJ, Garavan HP, Thompson WK, et al. 2023. Task fMRI paradigms may capture more behaviorally relevant information than resting-state functional connectivity. *NeuroImage* 270:119946

Autism and Cognitive Inflexibility

Autism spectrum disorder (ASD) is a neurodevelopmental disorder characterized by social deficits and restricted and repetitive behaviors (RRBs). RRBs are an understudied core symptom of ASD, yet negatively affect vital abilities such as social interactions, learning new skills, and completing daily activities (Aqdassi et al. 2021; Sadeghi and Pouretemad 2022). Autistic individuals additionally face greater cognitive flexibility difficulties (Lage, Smith, and Lawson 2024), and this dysfunction is thought to underlie RRBs (Gioia et al. 2002; Lopez et al. 2005). Cognitive inflexibility poses a great challenge in daily living in children with ASD such as creating difficulties with transitioning between activities (Craig et al. 2016), emotion regulation (Gardiner and Iarocci 2017), and adaptive behavior (Gilotty et al. 2002). Cognitive inflexibility is vital to understand in early life as it can have detrimental impacts later in life, as manifested by lower rates of employment and independent living (Anderson et al. 2014), quality of living (Mason et al. 2018), and fewer relationships (Farley et al. 2009).

Early identification of ASD and early cognitive flexibility intervention holds promise for improving life outcomes. There is abundant evidence that early diagnosis and intervention improves life outcomes of children diagnosed with ASD (Vivanti et al. 2014; Okoye et al. 2023), yet one in 36 children are diagnosed with ASD and the median age of diagnosis is not until 4 years of age (Maenner et al. 2023). Due to a delayed diagnosis, children are missing out on critical treatment and intervention during critical years of formative brain development (Tierney and Nelson 2009a). There is also evidence that cognitive-focused intervention improves cognitive flexibility as demonstrated in improved classroom behavior, behavior flexibility, and

problem solving skills (Kenworthy et al. 2014). Therefore, early detection and intervention should be prioritized in ASD to improve cognitive flexibility, symptoms, and life outcomes. There are early behavioral indicators of ASD (Barbaro and Dissanayake 2009), but these signs are typically observed in the social domain (Ozonoff et al. 2010) and are not reliable enough to establish diagnosis and can lead to biases (Guthrie et al. 2019; Pierce et al. 2019; Jarquin et al. 2011). Brain biomarkers hold promise in identifying early markers of risk for ASD in concert with overt behavioral signs (I. Molnar-Szakacs, Kupis, and Uddin 2021). Additionally, early brain markers of cognitive inflexibility may help improve intervention targets. Overall, identifying early brain markers of ASD and cognitive inflexibility is imperative to reduce the delay to diagnosis and treatment, and to improve outcomes for those affected.

Early Detection and Methods

Behavioral signs of ASD are rarely observable during the first year of life, and in subsequent years they are difficult to distinguish from both typical development and from other disorders (Istvan Molnar-Szakacs, Kupis, and Uddin 2020). However, risk of ASD appears to affect multiple brain systems prior to detectable behavioral signs of ASD (Eric Courchesne, Gazestani, and Lewis 2020; Xiao et al. 2021). Non-invasive techniques such as magnetic resonance imaging (MRI) are useful tools to identify early biomarkers for ASD and predict symptom severity to guide intervention and treatments (Muhle et al. 2018). Sleep MRI - that is MRI conducted during natural sleep - is a safe and effective method for examining the brain's functional architecture, reducing the concerns related to participant head motion while obviating the need for anesthesia (Power et al. 2012). Sleep MRI has already been used to examine brain

activation patterns in toddlers with ASD (Redcay and Courchesne 2008; Lombardo et al. 2015; Eyler, Pierce, and Courchesne 2012) and brain network dynamics during sleep ("Connectivity Dynamics from Wakefulness to Sleep" 2020). Additionally, resting-state fMRI methods - that is MRI conducted with no stimuli - are used to investigate the intrinsic functional architecture of the brain and hold promise of mapping brain function with behavior (Lee, Smyser, and Shimony 2013) and have been used in children and adults with ASD. Task fMRI - MRI conducted while participants perform a task during the scan - is commonly used to understand the blood-oxygen level dependent (BOLD) changes (Dichter 2012) and uncover brain regions involved in a cognitive process and has been used across the lifespan. MRI methods paired with detection of early behavioral signs of a developmental delay hold potential to reduce the delay to diagnosis and advance targeted treatments of cognitive flexibility.

Brain markers of autism and cognitive inflexibility: The Triple Network Model

The use of resting-state and task fMRI methods has uncovered potential brain biomarkers for ASD and cognitive inflexibility in children and adults with ASD. Previous studies have found that children and adults with ASD show atypical brain patterns during both task performance and resting-states (Marshall et al. 2020a; Kupis, Goodman, Kircher, et al. 2021; "Brain Mechanisms Supporting Flexible Cognition and Behavior in Adolescents With Autism Spectrum Disorder" 2021; Lau, Leung, and Lau 2019; Hull et al. 2016; Dichter 2012). Most studies point towards patterns of hyper and hypo functional connectivity, a measure of how brain regions interact with each other, in ASD (Lucina Q. Uddin, Supekar, and Menon 2013) and especially evident during young age (Nomi and Uddin 2015). The broad hyper and hypo connectivity observed in ASD

appears to depend on task demands, brain networks under investigation, age, and method used (Hernandez et al. 2015; Rudie and Dapretto 2013). Despite the widespread findings, many studies consistently highlight dysfunction of three large-scale networks. These networks are the salience network (SN), comprised of the anterior cingulate cortex and anterior insula, involved in interoceptive, affective, attention, and control processes; the central executive network (CEN), comprised of the dorsolateral prefrontal cortex and the anterior inferior parietal cortex, involved in modulating goal-oriented behaviors and decisions; and the default mode network (DMN), comprised of the medial prefrontal cortex, posterior cingulate cortex, and the posterior parietal lobule, involved in self-related and social cognition (Lucina Q. Uddin, Yeo, and Spreng 2019; L. Q. Uddin et al. 2011; Lucina Q. Uddin et al. 2015a). These three large-scale brain networks form the basis for the influential "triple network model" (Vinod Menon and Uddin 2010). This theory and studies support the idea that the SN mediates the interaction between the CEN and DMN, and facilitates switching needed for successful cognitive flexibility (V. Menon 2015b; Chand et al. 2017). There is further evidence that effective and flexible switching among these networks predict cognitive flexibility abilities (Lucina Q. Uddin 2021; "Cognitive Flexibility as the Shifting of Brain Network Flows by Flexible Neural Representations" 2024; Cao et al. 2021). Interestingly, these three networks have been previously associated with RRB symptoms and cognitive flexibility in ASD (Lucina Q. Uddin et al. 2015a; Marshall et al. 2020a; "Brain Mechanisms Supporting Flexible Cognition and Behavior in Adolescents With Autism Spectrum Disorder" 2021; Lucina Q. Uddin et al. 2013a). For example, In older children with ASD, the severity of RRBs were associated with connectivity among these three large-scale brain networks (SN, CEN,

DMN) (Traynor and Hall 2015a; Lucina Q. Uddin et al. 2015a). Another study found static functional connectivity of the SN in 7-12 year old children predict individual differences in RRBs in ASD (Lucina Q. Uddin et al. 2013a). The SN, CEN, and DMN hold promise as potential biomarkers for ASD and early cognitive flexibility skills.

Early neurodevelopment, brain markers of ASD, and cognitive inflexibility

To date, there are few studies that have investigated early brain network development in typical development, early brain markers of ASD, and relationships between early brain development and emerging behavior. From the few studies that have investigated early functional networks in typical development, neural networks are observed in neonates and have even been observed in utero (Thomason et al. 2013; Grayson and Fair 2017; Keunen, Counsell, and Benders 2017). The overall framework of mature brain wiring appears set at the time of birth, and intra and inter network FC continue to develop within the first years of life (Zhang, Shen, and Lin 2019). Formation of FC begins with primary networks followed by higher order networks (Keunen, Counsell, and Benders 2017), such that the visual network and sensorimotor networks emerge first followed by the SN, CEN, and DMN formation (W.-C. Liu et al. 2008; Zhang, Shen, and Lin 2019). The first few years of life are characterized by rapid development with brain volume doubling in the first year and reaching adult volume by age three (E. Courchesne et al. 2000; Grayson and Fair 2017). By two years of age the neural networks appear to be established (Zhang, Shen, and Lin 2019; Grayson and Fair 2017) and both genetics and environment have important contributions to the development of brain functional networks (Gao et al. 2014). Additionally, early brain FC in neurotypical infants have been used to predict learning ability (Zhang et al. 2017),

language performance (Emerson, Gao, and Lin 2016), and higher order cognition at later ages (Alcauter et al. 2014).

The investigation of early brain networks has clinical implications for at-risk populations such as ASD and for uncovering early markers associated with later cognitive inflexibility. The majority of neuroimaging studies examining infants or toddlers at-risk for ASD have examined structural MRI biomarkers that predict later developing ASD and few studies have investigated functional connectivity. Further, most studies mapping brain-behavior relationships have focused on later developing social deficits and language while few have focused on RRBs or flexible cognition and behavior (Istvan Molnar-Szakacs, Kupis, and Uddin 2020). From the studies completed, there is promising evidence of early brain biomarkers predictive of ASD and later developing behaviors (I. Molnar-Szakacs, Kupis, and Uddin 2021). Studies of infants and toddlers at-risk of ASD have revealed alterations in brain volume are predictive of ASD and severity of symptoms (Pote et al. 2019; Hazlett et al. 2012; Shen et al. 2013). Studies have revealed early language areas predictive of later language outcomes in toddlers later diagnosed with ASD (Lombardo et al. 2015). Studies investigating structural connectivity, or white matter tract microstructure (Edward Roberts, Anderson, and Husain 2013), also find promising brain markers predictive of later language development and ASD symptoms (J. Liu et al. 2019; Wolff et al. 2012).

There is additionally a rise of studies investigating FC across all ages of autism (Lucina Q. Uddin, Supekar, and Menon 2013), making it a promising biomarker for investigating in the first years of life. For example, one study investigated FC in 6-month old HR infants and successfully identified subjects who later went on to develop ASD based on the FC measures (Emerson et al.

2017). They additionally found correlations between 6-month FC and 24-month social behavior, language, motor development, and repetitive behavior scores (Emerson et al. 2017). Another study found that alterations of the SN connectivity in 6 week old infants at high risk of ASD predicted subsequent ASD symptoms (Tsang et al. 2024). Another study specifically examined early brain networks associated with RRBs and ASD in infants at high risk of developing ASD (McKinnon et al. 2019). They found alterations in DMN and CEN FC were associated with RRBs at 12 and 24 months of age. These studies provide seminal evidence of potential brain biomarkers of ASD and support the further investigation of brain network alterations during the first years of life in infants and toddlers at risk of ASD and behavioral difficulties.

RRBs and the Relationship with Cognitive Flexibility

Restricted and repetitive behaviors (RRBs) are a hallmark feature of ASD and are associated with cognitive inflexibility making this symptom important to investigate during early life. RRBs are a core symptom of ASD and behaviorally can present as stereotyped movements, insistence on sameness, and circumscribed or perseverative interests (American Psychiatric Association 2021). ASD is, however, heterogeneous (Masi et al. 2017), resulting in a diverse range of RRBs and severity. RRB severity is also predictive of later adaptive behaviors, or the ability to adjust to the environment appropriately and effectively (Troyb et al. 2016). Further, RRBs are not unique to ASD, as they are present in idiopathic neurodevelopmental conditions (e.g., Fragile X),(Lewis and Kim 2009) other neuropsychiatric conditions such as obsessive-compulsive and related disorders (Abramowitz and Jacoby 2015) and even in typical development (Thelen 1980). The heterogeneity of ASD and the overlap of RRBs across diagnostic categories make it difficult

to observationally discern RRBs in ASD from other conditions during early development (Jiujias, Kelley, and Hall 2017). However, RRBs are usually more severe and more frequent in ASD as children develop (Richler et al. 2010a; Harrop et al. 2014). Further, the severity of RRBs is associated with cognitive inflexibility in ASD (Lopez et al. 2005). This highlights the need to further investigate early brain mechanisms underlying RRBs and cognitive inflexibility in ASD. In children and adults with ASD, studies point toward atypical brain patterns of the CEN and SN during cognitive flexibility tasks ("Brain Mechanisms Supporting Flexible Cognition and Behavior in Adolescents With Autism Spectrum Disorder" 2021). However, little is known about early brain network functioning and their association with later developing RRBs and cognitive inflexibility in ASD. This further emphasizes the importance of investigating RRBs and their relationship with cognitive flexibility in ASD especially during the first years of life.

The Research Domain Criteria (RDoC) framework includes psychological and biological 'domains' intended to be studied along a range of functioning from normal to abnormal. RDoC suggests that focusing on symptom domains that cut across diagnostic categories may be beneficial to understanding the heterogeneity of ASD and their behavioral symptoms (Masi et al. 2017). Additionally, RDoC recommends exploring neural substrates linked with dimensions of behavior to inform individual diagnosis and treatment in service of improving well-being (Insel and Wang 2010). Future work is needed to assess both dimensional and categorical brain-behavior relationships at early and longitudinal time-points in infants and toddlers later diagnosed with and without ASD. This approach will elucidate shared neural mechanisms between ASD and typically developing (TD) children and identify unique neural alterations in ASD.

Evolving fMRI methods: Brain Dynamics

Resting-state fMRI (rsfMRI) studies provide insight into functional brain network organization in the absence of task performance (Biswal et al. 1995) and reveal intrinsic brain connectivity patterns associated with behavior and cognition (Smitha et al. 2017). Functional connectivity is the temporal correlation between two distinct brain regions (Friston et al. 1993) averaged over a given time period (typically around 10 minutes or the duration of a resting-state fMRI scan). Recent advances show that functional connectivity patterns are dynamic, and can change over the course of seconds (Chang and Glover 2010). Thus, previous 'static' functional connectivity approaches may be too simplistic to index moment-to-moment changes in resting-state brain activity (Preti, Bolton, and Van De Ville 2017). FC methods, although valuable, rely on many assumptions and arbitrarily collapse data into time and space. Novel dynamic methods, such as co-activation pattern analysis (CAP) and dynamic FC (dFC), account for these changes over time, and in some instances may better capture brain-behavior relationships than static methods (Lurie et al. 2020). The temporal variability as measured using dynamic methods has already shown clinical utility (Damaraju et al. 2014), including in ASD ("Brain Mechanisms Supporting Flexible Cognition and Behavior in Adolescents With Autism Spectrum Disorder" 2021) and have been used to characterize brain network dynamics during sleep ("Connectivity Dynamics from Wakefulness to Sleep" 2020). Brain dynamic methods hold promise in elucidating early brain network dynamics in both ASD and typically developing infants and toddlers.

Co-activation Pattern Analysis (CAP)

Novel dynamic methods such as co-activation pattern (CAP) analysis (X. Liu, Chang, and Duyn 2013; X. Liu et al. 2018) are increasingly utilized (Kupis, Romero, Dirks, Hoang, Parlade, et al. 2020; Kupis, Goodman, Kircher, et al. 2021) because they capture time-varying brain state alterations not otherwise observed using static methods and in some instances reveal more brain and behavior relationships compared with static methods (Lurie et al. 2020). The CAPs method deviates from traditional dynamic methods by accounting for single fMRI volumes at individual time points and focusing on recurring CAPs of the brain. The CAPs overall represent instantaneous brain configurations at single time points (X. Liu et al. 2018). This method is data driven and relies on a clustering algorithm to determine the brain co-activations. This reveals recurring patterns of CAP states with distinct functional and neural relevance. For instance, a CAP could consist of DMN co-activation with subcortical regions such as the hippocampus (X. Liu, Chang, and Duyn 2013; X. Liu et al. 2018). These meaningful CAPs also appear to have important time varying instances that may reflect the dynamic organization of the brain and changes in the CAPs are found to underlie cognition and behavior (X. Liu et al. 2018; Bray et al. 2015). CAP method has also been used to investigate neurodiverse populations (Marshall et al. 2020b; Goodman et al. 2021; "Reproducible Coactivation Patterns of Functional Brain Networks Reveal the Aberrant Dynamic State Transition in Schizophrenia" 2021). The CAP method is promising for investigating brain dynamics across all ages, and in neurotypical and diverse populations. Additionally, since it is data driven and relies on independent component analysis and a clustering method (X. Liu et al.

2018), it holds promise to elucidate early brain biomarkers in infants and toddlers with and without ASD.

Dynamic Functional Connectivity (DFC)

Another commonly used dynamic brain method is dynamic functional connectivity (dFC) using the sliding window approach (Hutchison et al. 2013). In this method, a fixed length of time (e.g., 30s, 60s, or 120s) is chosen and data points within that period of time are used to calculate the FC in that specific window. The window is then shifted over by a fixed number of data points allowing for a certain amount of overlap between each window. A FC measure is calculated for each window of time, and then a clustering method such as *k*-means clustering is applied to the data set and used to group the FC matrices based on their similarity. This results in averaged clusters or brain states representing FC patterns that recur throughout the duration of the fMRI scan. These repeating patterns provide insights into how the brain dynamically shifts and reorganizes itself throughout a task or rest state and holds promising cognitive and behavioral significance (Hutchison et al. 2013; Cohen 2018).

Brain Dynamics in Autism and Cognitive Flexibility

Brain dynamic methods such as CAP and dFC are emerging in the field of ASD research and in the study of flexible cognition and behavior (Lurie et al. 2020; L. Q. Uddin and Karlsgodt 2018). Dynamic methods have already revealed neural underpinnings of brain inflexibility, and cognitive and behavioral inflexibility in ASD ("Brain Mechanisms Supporting Flexible Cognition and Behavior in Adolescents With Autism Spectrum Disorder" 2021). Studies have revealed aberrant dynamic interactions among the SN, CEN, and DMN in ASD. For example,

the frequency of the SN was altered in ASD children compared with neurotypical children (Marshall et al. 2020b). Other studies report reduced transitions between brain states in children and adults with ASD (de Lacy et al. 2017; Watanabe and Rees 2017). Another study investigating 774 6-10 year old children found a longer dwell time in a state characterized by global disconnection was associated with higher levels of ASD traits and ASD diagnosis (Rashid et al. 2018). These findings suggest an earlier neurodevelopmental origin and support investigating brain network dynamics in ASD and typical developmental in early life.

CHAPTER 2:

Brain Dynamics Underlying Cognitive Flexibility Across the Lifespan

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ORIGINAL ARTICLE

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Abstract

The neural mechanisms contributing to flexible cognition and behavior and how they change with development and aging are incompletely understood. The current study explored intrinsic brain dynamics across the lifespan using resting-state fMRI data ($n = 601$, 6–85 years) and examined the interactions between age and brain dynamics among three neurocognitive networks (midcingulo-insular network, M-CIN; medial frontoparietal network, M-FPN; and lateral frontoparietal network, L-FPN) in relation to behavioral measures of cognitive flexibility. Hierarchical multiple regression analysis revealed brain dynamics among a brain state characterized by co-activation of the L-FPN and M-FPN, and brain state transitions, moderated the relationship between quadratic effects of age and cognitive flexibility as measured by scores on the Delis-Kaplan Executive Function System (D-KEFS) test. Furthermore, simple slope analyses of significant interactions revealed children and older adults were more likely to exhibit brain dynamic patterns associated with poorer cognitive flexibility compared with younger adults. Our findings link changes in cognitive flexibility observed with age with the underlying brain dynamics supporting these changes. Preventative and intervention measures should prioritize targeting these networks with cognitive flexibility training to promote optimal outcomes across the lifespan.

Key words: aging, central executive network, default mode network, executive function, salience network

Introduction

Flexible brain dynamics support cognition and behavior (Grady and Garrett 2014; Jia et al. 2014). However, little is known regarding brain dynamic changes across the lifespan associated with cognitive flexibility, a component of executive function (Diamond 2013) that supports the ability to adapt behavior to an ever-changing environment (Dajani and Uddin 2015). Cognitive flexibility is associated with positive academic, occupational, and social outcomes throughout life (Davis et al. 2010; Genet and Siemer 2011; Burt and Paysnick 2012; Yeniad et al. 2013; Colé et al. 2014). Understanding age-related changes in brain dynamics and their relationship with cognitive flexibility is crucial to identifying neural markers of risk and resilience across development and aging.

Across the lifespan, greater dynamic brain flexibility is increasingly being associated with younger adulthood and

enhanced cognitive performance (Jia et al. 2014; Braun et al. 2015; Nomi et al. 2017a; Xia et al. 2019; Battaglia et al. 2020). A greater number of transitions among certain brain states has been found in younger adults compared with older adults (Xia et al. 2019) and children (Hutchison and Morton 2015). The dwell time, or the time spent within a brain state, has also been shown to differ across age, with shorter dwell times in certain states in young adulthood (Hutchison and Morton 2015) potentially underlying efficient cognitive control. Dwell time increases with older age (Xia et al. 2019), potentially underlying cognitive changes and reduced cognitive efficiency (i.e., perseveration) (Ridderinkhof et al. 2002). Lastly, the frequency of occurrence of highly variable brain states has also been associated with better performance on behavioral measures of executive function including cognitive flexibility (Nomi et al. 2017b). Although greater dynamic brain flexibility is increasingly being associated with younger age and enhanced cognitive performance, there is little known about variability in brain dynamics supporting cognition across age. For example, growing evidence suggests individuals have varying "brain ages," resulting in differences in functional brain maturity among age-matched individuals (Dosenbach et al. 2010). Therefore, age-related changes associated with brain network dynamic variability and cognitive flexibility require further investigation (Cohen 2018) as they may provide potential markers of risk for, and resilience to, age-related cognitive decline across the lifespan.

Within and between network connectivity among the midcingulo-insular network (M-CIN; also known as salience), medial frontoparietal network (M-FPN; also known as default), and lateral frontoparietal network (L-FPN; also known as executive control) (Uddin et al. 2019) has also been shown to be important for aging (Ryali et al. 2016; Chand et al. 2017), and cognitive and neural flexibility (Uddin et al. 2011; Chen et al. 2016). The M-CIN is involved in interoceptive, affective, attention, and control processes associated with subjective salience; the L-PFN is involved in executive control and modulating goal-oriented behaviors and decisions; and the M-FPN is involved in self-related processes and social cognition (Uddin et al. 2019). Together, these networks support various functions important for adaptation across the lifespan (Masten and Obradovic 2006; Touroutoglou et al. 2018). A longer dwell time within certain states of the M-CIN, M-FPN, and L-FPN has been associated with less flexibility in children's brain dynamic repertoires compared with young adults (Ryali et al. 2016). Greater flexibility within these networks may therefore account for improved behavioral performance across development. In older age, extant literature suggests weaker modulation occurs among the M-FPN and L-FPN, resulting in the greater reliance on crystallized knowledge, and weaker fluency skills (Turner and Nathan Spreng 2015; Spreng et al. 2018). Furthermore, temporal variability specifically of the M-CIN has been shown to uniquely predict individual differences in cognitive flexibility in young adults (Chen et al. 2016). Conversely, higher M-FPN and L-FPN functional dynamics during the resting-state have been associated with poorer cognitive flexibility (Douw et al. 2016). Overall, dynamic relationships among the M-CIN, M-FPN, and L-FPN appear to be important contributors to cognitive flexibility across the lifespan.

Despite its importance to optimal lifespan development, no previous studies have characterized brain network dynamics supporting cognitive flexibility from childhood to older adulthood. This study provides a novel framework for understanding the relationship between brain dynamics and cognitive flexibility and may lend insight into neuropsychiatric disorders and resilience in typical development and aging. Previous studies have found both linear and quadratic relationships across the lifespan related to cognitive flexibility and brain dynamics when examining within- and between-network associations (Grady et al. 2006; Wang et al. 2012; Betzel et al. 2014; Cao et al. 2014; Nomi et al. 2017a). To extend previous findings, we examined the hypotheses that between-network dynamics among the M-CIN, M-FPN, and L-FPN exhibit a quadratic trajectory across the lifespan. To examine if varying levels of brain dynamics supports optimal cognitive flexibility across the lifespan, we also tested the hypothesis that brain dynamics among these three large-scale networks interact with age to enable cognitive flexibility changes associated with healthy aging. Specifically, we hypothesized that greater brain dynamic flexibility as indexed by dwell time, frequency of occurrence, and transitions between states would be associated with greater cognitive flexibility across the lifespan.

Table 1 Participant Demographics

	N = 601; mean ± sd (min—max)
Age (year)	37.22 ± 20.73 (6.18–85.62)
Gender	239 M 361 F 1 NR
Mean FD (mm)	0.25 ± 0.09 (0.08–0.50)
Ethnicity	514 (not Hispanic or Latino) 86 (Hispanic or Latino) 1 NR
Race	4 (1) 46 (2) 116 (3) 1 (4) 417 (5) 16 (6) 1(NR)
CWIT inhibition/switching total completion time	62.80 ± 17.89 (32–146)
CWIT inhibition/switching total errors	1.92 ± 2.24 (0–22)
TMT number-letter switching total completion time	81.70 ± 38.79 (25–240)
VF switching total correct	13.49 ± 3.20 (4–23)

Note: SD, standard deviation; M, male; F, female; NR: no response; 1: American Indian or Native Alaskan; 2: Asian; 3: Black or African American; 4: Native Hawaiian or Other Pacific Islander; 5: White; 6: Other Race; CWIT, Color-Word Interference Test; TMT, Trail Making Test; VF, Verbal Fluency.

Methods

Neuroimaging, phenotypic, and behavioral data collected from 601 healthy adult participants were downloaded from the Enhanced Nathan Kline Institute.

(NKI)-dataset (http://fcon_1000.projects.nitrc.org/indi/enhanced/). Participants were selected according to the following inclusion criteria: 1) availability of neuroimaging and behavioral data, 2) no current or past DSM-diagnosis for psychiatric disorders and/or attention deficit hyperactivity disorder, and (3) resting-state fMRI data head motion <0.5 mm. See Table 1 for participant information and Supplementary Figure S1 for information about the age distribution included in this study. The study was approved by the NKI institutional review board and all participants provided informed consent. Written consent and assent was collected from child participants and their legal guardian (Nooner et al. 2012)

MRI and Behavior Protocol

Participants were assessed during a 1- or 2-day examination by trained experts. Details of the MRI and behavioral assessment

procedures can be found at http://fcon_1000.projects.nitrc.org/indi/enhanced/mri_protocol.html, and http://fcon_1000.projects.nitrc.org/indi/enhanced/assessments.html, respectively. Some participants were missing behavioral data for certain measures and were omitted when necessary. Additionally, children below the age of 8 years ($n = 7$) were not administered the executive function tests, as the test battery is only valid in 8–89 year olds (Delis et al. 2001). These children were excluded from the analyses with behavioral measures of executive function.

Cognitive Flexibility Measures

Participants were administered the Delis-Kaplan Executive Function System (D-KEFS), a series of neuropsychological tests designed to measure executive functions in children and adults between the ages of 8–89 (Delis et al. 2001). The commonly used cognitive flexibility tests within the D-KEFS include the Color-Word Interference Task (CWIT), the Trail Making Test (TMT), and the Verbal Fluency (VF) Task.

The CWIT is a modified Stroop task (Stroop and Ridley Stroop 1992) and consists of four conditions. The first two conditions are similar to the Stroop interference task, and the last condition involves Inhibition/Switching and is a commonly used cognitive flexibility task (Bohnen et al. 1992; Mattson et al. 1999). In the Inhibition/Switching condition, participants are presented with a page containing the words "red," "green," and "blue," written in red, green, or blue ink. Some of the words are contained in a box and the subject must switch between saying the color of the ink (word is not inside a box) or the color of the word (word inside a box). Participants are told to complete the task as quickly as possible. Raw scores include the time to complete the Inhibition/Switching condition in seconds and the total number of errors made during the task. Higher scores indicate poorer cognitive flexibility.

The TMT was created to isolate set-shifting abilities by including baseline conditions such as visual scanning, number sequencing, letter sequencing and motor speed (Fine et al. 2011). TMT also includes a Number-Letter Switching condition, a commonly used cognitive flexibility task (Kleinhans et al. 2005; Mcdonald et al. 2005; Yochim et al. 2007). During the Number-Letter Switching condition, participants switch back and forth between connecting numbers and letters (i.e., 1, A, 2, B etc.,) (Yochim et al. 2007). They are instructed to connect the numbers and letters as quickly as possible. The raw score measure for the Number-Letter Switching task is the total time to complete the task in seconds. Higher scores indicate poorer cognitive flexibility.

The VF test requires participants to generate words beginning with a letter (phonemic fluency) or from a category (category fluency). The VF task also includes a Category Switching condition where participants alternate between saying words from two different semantic categories. The Category Switching condition is a commonly used task to study cognitive flexibility (de Paula et al. 2015; Ramanan et al. 2015). In the switching condition, participants are told to produce as many words within 60 seconds. The VF category switching raw score is the total correct number of responses and a higher score indicates better cognitive flexibility.

MRI Data Acquisition

A Siemens Trio 3.0 T scanner was used to obtain the functional images. Multiband (factor of 4) echo-planarimage (EPI) sequenced resting-state images (rsfMRI; TR = 1400 ms, TE = 30 ms, flip angle 65°, field of view (FOV) 224 mm, voxel size = 2x2x2 mm, 64 interleaved slices, 404 volumes) were applied for the acquisition of the functional images. Participants were instructed to keep their eyes open and fixate on a cross in the center of the screen during the 9-min 19-s rsfMRI scan. For detailed MRI protocol see: http://fcon_1000.projects.nitrc.org/indi/enhanced/mri_protocol.html.

Neuroimaging Data Preprocessing and Postprocessing

The resting-state fMRI data were preprocessed using the Data Preprocessing Assistant for Resting-State fMRI Advanced edition (DPARSF-A, Yan and Zang, 2016), which uses FSL, SPM-12 (https://www.fil.ion.ucl.ac.uk/spm/software/spm12/), and AFNI https://afni.nimh.nih.gov (Cox 1996). The preprocessing steps were the following: removal of the first 5 volumes to allow scanner signal to reach equilibrium, despiked using AFNI 3dDespike, realignment, normalization to 3 mm MNI template, and smoothing (6 mm FWHM) (Espinoza et al. 2019).

Independent component analysis (ICA) was conducted using FSL's MELODIC by means of automatic dimensionality estimation (Nomi et al. 2017a; Espinoza et al. 2019). The ICA-FIX classifier was trained on hand-classified independent components separated into noise and non-noise categories using randomly chosen participants ($n = 24$) across the lifespan (Griffanti et al. 2014; Nomi et al. 2017b). The ICA-FIX classification algorithm was applied to the data (FSL's ICA-FIX; (Griffanti et al. 2014) to classify noise and non-noise components from individual subject data before conducting nuisance regression of classified noise components from the resting-state scans in MNI space. The ICA-FIX fMRI data then underwent nuisance covariance regression (linear detrend, Friston 24 motion parameters (6 motion parameters of each volume, the preceding volume, and the 12 corresponding squared items) (Friston et al. 1996), global mean signal, followed by bandpass filtering (0.01–0.10 Hz) (Damoiseaux et al. 2006). Preprocessing and postprocessing were additionally conducted without global mean signal regression (GSR) to assess the effect of this step on subsequently derived metrics, as there is yet no consensus regarding the extent to which this step removes neural signal in addition to noise (Uddin 2020a).

Nine regions-of-interest (ROIs) representing the three large-scale networks (Uddin et al. 2011) were selected (Table 2), including the right and left fronto-insular cortex (rFIC) and anterior cingulate cortex (ACC) of the M-CIN; right and left dorsolateral prefrontal cortex (rDLPFC) and right and left posterior parietal cortex (rPPC) of the L-FPN; and the ventromedial prefrontal cortex (VMPFC) and posterior cingulate cortex (PCC) of the M-FPN. These networks and regions were chosen because of previous work demonstrating their functional roles in flexible cognition (Uddin et al. 2011) and aging (Ryali et al. 2016; Chand et al. 2017). Additionally, these ROIs have long been recognized as critical nodes in the three neural networks (Seeley et al. 2007; Menon and Uddin 2010; Chand et al. 2017) and as evidenced by recent ICA group analyses (Marshall et al. 2020; Kupis et al. 2021). A trained research assistant examined all ROIs in older participants ($\geq$ 70–85 years), the years where the most marked changes in brain atrophy can occur (Scahill et al. 2003), to ensure the masks were within the cerebral cortex for each individual subject.

Co-Activation Pattern Analysis

For each individual subject, time series extracted from the nine ROIs were converted to z-statistics and then concatenated into

Table 2 Coordinates of M-CIN, M-FPN, and L-FPN regions

Network	Region	BA	Peak MNI coordinates (mm)
M-CIN	rFIC	47	39, 23, −4
	lFIC	47	−34, 20, −8
	ACC	24	6, 24, 32
L-FPN	rDLPFC	9	46, 20, 44
	lDLPFC	9	−46, 20, 44
	rPPC	40	52, −52, 50
	lPPC	40	−40, −56, 44
M-FPN	VMPFC	11	−2, 38, −12
	PCC	23/30	−6, −44, 34

one matrix containing all subjects [(399 TR × 601 subjects) × 9 ROIs], following previous studies (Hutchison and Morton 2015; Kupis et al. 2020). Both children and adults were included due to prior evidence suggesting the brain's repertoire of states are generally preserved across age (Hutchison and Morton 2015). The matrix was then subjected to *k*-means clustering to determine the optimal number of clusters. The elbow criterion was applied to the cluster validity index (the ratio between within-cluster to between-cluster distance) for values of $k = 2$–20 to determine the optimal value of $k = 5$ (Supplementary Figure S2) (Liu et al. 2013).

K-means clustering using a squared Euclidean distance was then applied to the matrix using the optimal $k = 5$ to produce 5 co-activation pattern (CAP) "brain states." The CAP metrics included: a) dwell time, calculated as the average number of continuous TRs that a participant stayed in a given brain state, b) frequency of occurrence of brain states, calculated as an overall percentage that the brain state occurred throughout the duration of the scan compared with other brain states, and c) the number of transitions, calculated as the number of switches between any two brain states.

In the processing pipeline including the data without GSR, *k*-means analysis was again conducted to obtain the optimal *k*, determined to be $k = 5$.

Statistical Analysis

To test our first hypothesis that the dynamic network integration among networks important for cognitive flexibility differs across age, linear and quadratic regressions were conducted with Age and Age^2, predicting the dynamic brain state metric (dwell time, frequency, and transitions) for each CAP. Covariates included head motion and sex. Age^2 was included due to prior evidence revealing age has a quadratic or curvilinear relationship with certain brain regions and networks (DuPre and Nathan Spreng 2017; Chen et al. 2018). Overall, this model was conducted to extend prior "static" results by using dynamic brain network states.

$$\hat{Y} = B_0 + B_1(\text{Age}) + B_2(\text{Age}^2) + B_n(\text{Covariates}).$$

To test our second hypothesis that brain dynamics moderate the relationship between age and cognitive flexibility, hierarchical multiple regressions were conducted. Hierarchical multiple regression analysis includes adding variables into the model in separate steps (Francis et al. 1975). In the first step, Age and Age^2 were included as predictors of cognitive flexibility, with sex and mean FD included as covariates. This tested for quadratic relationships between age and cognitive flexibility before the moderation analysis were conducted. In the second step, the brain dynamic metric (dwell time, frequency, and transitions) for each CAP was included as a predictor. In the last step, the interaction between Age and the dynamic metric and the interaction between Age^2 and the dynamic metric were included into the regression analysis. Brain dynamics were tested as the moderator in this study due to the idea that there may be variability in brain functioning among subjects of the same age (Dosenbach et al. 2010). This approach supports assessing variability in brain dynamics associated with cognitive flexibility across the lifespan, while still revealing age-related changes. The cognitive flexibility measures used were the CWIT Inhibition/Switching, the TMT Color/Number Switching, and the VF Category Switching raw scores. Following significant interactions, the simple slopes were examined to aid interpretation. Simple slopes were computed to explore the effect of Age^2 on the cognitive flexibility measure at three different levels of the moderator as represented by the brain dynamic metric (i.e., at −1 *SD* below the mean, at the mean, and at +1 *SD* above the mean). All analyses were conducted using R (Computing and Others 2013) (https://www.R-project.org/) and all analyses are publicly available (https://github.com/lkupis/lifespan_Dynamics). Additional analyses were also conducted with more ROIs using the Schaefer parcellation (Schaefer et al. 2018), and are available in the Supplementary Materials.

$$\hat{Y} = B_0 + B_1(\text{Age}) + B_2(\text{Age}^2) + B_n(\text{Covariates}) \quad \text{[Step 1]}$$

$$\hat{Y} = B_0 + B_1(\text{Age}) + B_2(\text{Age}^2) + B_3(\text{Brain Dynamic}) + B_n(\text{Covariates}) \quad \text{[Step 2]}$$

$$\hat{Y} = B_0 + B_1(\text{Age}) + B_2(\text{Age}^2) + B_3(\text{Brain Dynamic}) + B_1(\text{Age} \times \text{Brain Dynamic}) + B_2(\text{Age}^2 \times \text{Brain Dynamic}) + B_n(\text{Covariates}) \quad \text{[Step 3]}$$

Results

Recurrent CAP Analysis

Results from the CAP analysis among the M-CIN, L-FPN, and M-FPN are presented in Figure 1. The first brain state (CAP 1) was characterized by stronger co-activation among the M-FPN nodes relative to the L-FPN and M-CIN. The second brain state (CAP 2) was characterized by co-activation among the M-CIN nodes. The third brain state (CAP 3) was characterized by co-activation among the M-CIN and the M-FPN. The fourth brain state (CAP 4) was characterized by co-activation among the L-FPN and M-CIN. The last brain state (CAP 5) was characterized by co-activation among the L-FPN and M-FPN.

CAP Analysis without Global Signal Regression

Results from the CAP analysis using data without GSR are presented in Supplementary Figure S3. The resulting CAPs revealed the influence of the global signal, notably in CAPs 1 and 2. CAP 1 shows all nodes with inactivity and CAP 2 shows all nodes with activity representing the global signal across all nodes. Prior work suggests that the decision to remove the global signal or not depends on the scientific question, and should be considered when interpreting the results (Murphy and Fox 2017). The removal of the global signal as a preprocessing step significantly mitigates artifacts from a variety of sources (Power et al. 2017; Ciric et al. 2018). Although in some cases the global signal can represent neuronal signal (Hyder and Rothman 2010; Schölvinck et al. 2010); in the current dataset, removal of the global signal was beneficial to revealing CAPs associated with cognition. Therefore, all statistical analyses and results presented are derived from data that was preprocessed with GSR.

Associations between Brain Dynamics and Quadratic Effects of Age

Curvilinear regressions were conducted with Age and Age^2 predicting the brain dynamic metric for each brain state (CAP 1–5), while controlling for sex and mean FD. There was a positive quadratic effect of age when predicting the frequency of CAP 3, characterized by co-activation among the M-CIN and M-FPN, β = 0.42, $b = < 0.001$, $SE = < 0.001$, $P = 0.030$, uncorrected. CAP 3 occurred less frequently as age increased, but increased in occurrence with older age (see Fig. 2A). There was also a negative quadratic effect of age when predicting the frequency of CAP 5, characterized by co-activation among the L-FPN and M-FPN, $\beta = -0.40$, $b = < 0.001$, $SE = < 0.001$, $P = 0.037$, uncorrected. CAP 5 occurred more frequently as age increased; however, it decreased with older age (see Fig. 2B). Lastly, there was a positive quadratic effect of age predicting the dwell time of CAP 4, characterized by co-activation among the M-CIN and L-FPN, $\beta = 0.37$, $b = < 0.001$, $SE = < 0.001$, $P = 0.053$, uncorrected. The dwell time of CAP 4 decreased with age, and increased with older age (see Fig. 2C).

Main Effects of age ^{2} and Brain Dynamics Predicting Cognitive Flexibility

Age, Age^2, and the brain dynamic metric were included in steps 1 and 2 of the hierarchical regression analyses. There were significant quadratic effects of age for the cognitive flexibility measures including the CWIT total errors raw score, TMT completion time raw score, and VF total correct number of responses raw score (P's < 0.001), but not for CWIT completion time raw score (P's > 0.05). The brain dynamic metrics were not significant predictors of cognitive flexibility when included into the regression equations (P's > 0.05).

Interactions between Age and Brain Dynamics Predicting Cognitive Flexibility

There were multiple significant interactions between the dynamic brain states and Age^2 predicting cognitive flexibility (Supplementary Table S1). Only the significant interactions that survived Bonferroni correction ((.05/10) = 0.005) will be discussed. The dwell time of CAP 5, characterized by co-activation among the L-FPN and M-FPN, moderated the relationship between the quadratic effect of age and cognitive flexibility (TMT switching completion time), $b = 0.02$, $SE = 0.01$, $P = 0.002$. Simple slope analyses indicated there was a significant slope between Age^2 and TMT switching completion time at low (−1 SD), $b = 0.02$, $SE = 0.01$, $P = 0.003$, average, $b = 0.03$, $SE = 0.004$, $P = < 0.001$, and high (+1 SD), $b = 0.04$, $SE = 0.01$, $P < 0.001$ CAP 5 dwell times. A low CAP 5 dwell time was associated with improved cognitive flexibility across the lifespan; Average CAP 5 dwell time consisted of slightly poorer cognitive flexibility at young and older ages and improved cognitive flexibility mid age. A higher CAP 5 dwell time was associated with poorer cognitive flexibility at younger and older ages and improved cognitive flexibility performance during mid-age. Although the simple slopes were significant at low, average, and high levels of CAP 5 dwell time, examination of the slopes in Figure 3A further revealed the effect was minimal at a low level (Li 2018). Overall, the dynamics of a brain state consisting of co-activation among the L-FPN and M-FPN moderated the relationship between cognitive flexibility with Age^2 (see Fig. 3A).

The number of brain state transitions also moderated the relationship between the quadratic effect of age and cognitive flexibility for TMT switching completion time, $b = -0.001$, $SE = < 0.001$, $P = 0.005$. Simple slope analyses indicated there was a significant slope between Age^2 and TMT switching completion time at low (−1 SD), $b = 0.04$, $SE = 0.01$, $P = < 0.001$, average, $b = 0.03$, $SE = 0.004$, $P = < 0.001$, and high (+1 SD), $b = 0.02$, $SE = 0.01$, $P = 0.001$, transitions. Simple slopes analyses indicated that greater numbers of transitions were associated with stable/-good cognitive flexibility throughout the lifespan, with a reduction in cognitive flexibility around mid-age. In both average and low transitions, cognitive flexibility was poorer in younger and older ages, but peaked during mid-age. Overall, transitions moderated the relationship between cognitive flexibility and Age^2 (see Fig. 3B).

Discussion

Cognitive flexibility is an important executive function enabling optimal outcomes in academic achievement, transitions into adulthood, quality of life, and resilience to negative life events (Uddin 2021). Examining brain dynamic changes across the lifespan aids the understanding of the neural mechanisms underlying optimal and flexible cognition (Grady and Garrett 2014) and may inform studies of cognitive (Zhang et al. 2020a) and neuropsychiatric disorders (Rabany et al. 2019; Uddin 2020b). The large-scale networks known as the M-CIN (salience), L-FPN (executive), and M-FPN (default), are thought to be important for flexible cognition (Uddin et al. 2011; Qin et al. 2015) across aging (Chand et al. 2017; Adnan et al. 2019a). The present study examined brain dynamics among the M-CIN, L-FPN, and M-FPN as they relate to lifespan development, and as a moderator between age and cognitive flexibility.

The present study revealed five recurring CAP (CAPs or "brain states") involving the M-CIN, L-FPN, and M-FPN across the lifespan. Quadratic relationships were observed between age and the brain dynamic metrics, primarily within hybrid brain states characterized by between-network coupling. Furthermore, brain dynamics moderated the relationship between a quadratic effect of age and cognitive flexibility. We demonstrate differences in intrinsic brain network dynamics across aging associated with cognitive flexibility, specifically within the M-FPN/L-FPN co-activation state (CAP 5), and brain

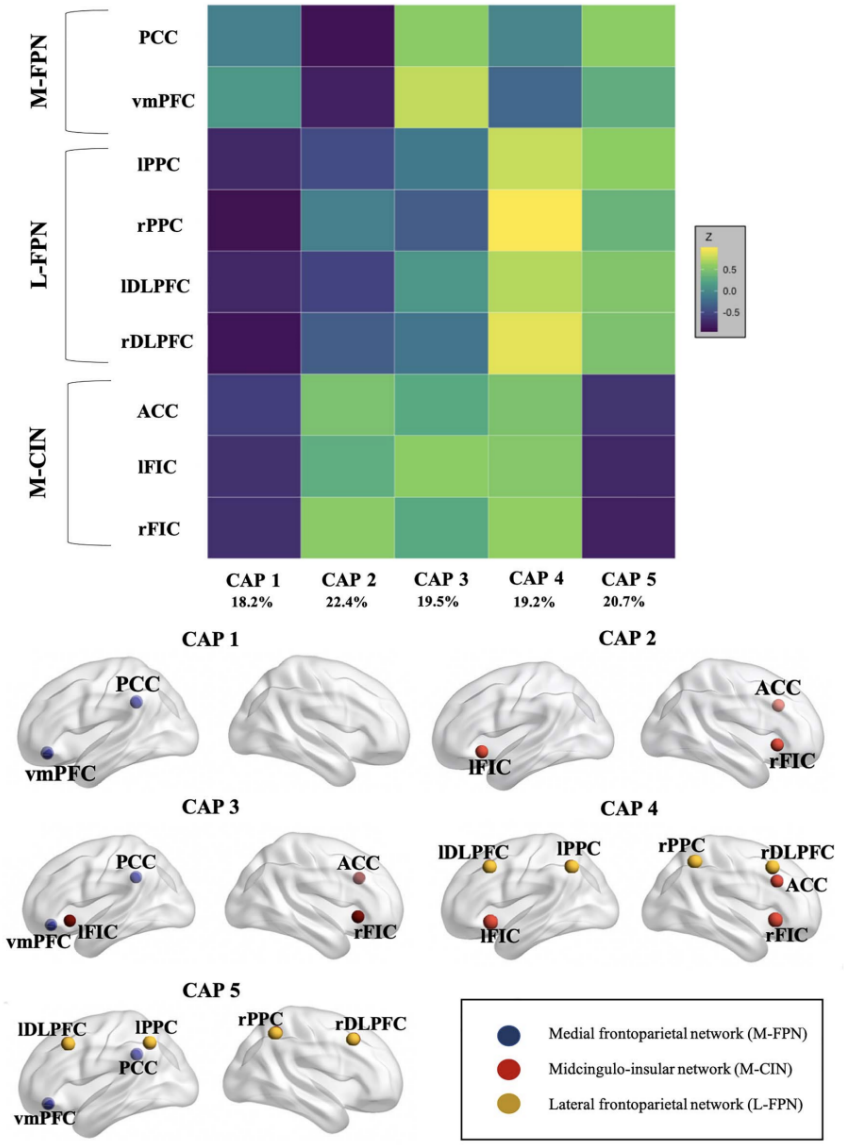

Figure 1. *Top*: CAPs or brain states from dynamic CAP analysis. CAP 1 was characterized by stronger co-activation among the M-FPN nodes relative to the L-FPN and M-CIN. CAP 2 was characterized by co-activation among the M-CIN nodes. CAP 3 was characterized by co-activation among the M-CIN and the M-FPN. CAP 4 was characterized by co-activation among the L-FPN and M-CIN. Lastly, CAP 5 was characterized by co-activation among the L-FPN and M-FPN. *Bottom*: Graphical brain representation of each CAP as demonstrated by the ROIs. Note: PCC, posterior cingulate cortex; vmPFC, ventromedial prefrontal cortex; lPPC, left posterior parietal cortex; rPPC, right posterior parietal cortex; lDLPFC, left dorsolateral prefrontal cortex; rDLPFC, right dorsolateral prefrontal cortex; ACC, anterior cingulate cortex; lFIC, left fronto-insular cortex; rFIC, right fronto-insular cortex.

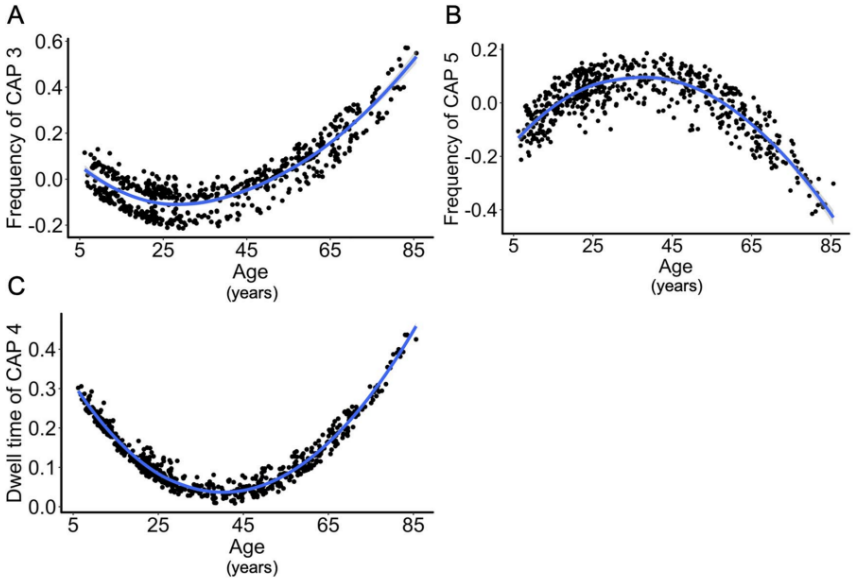

Figure 2. Positive and negative quadratic effects of age (years) predicting dynamic brain metrics for specific CAPs. For all graphs, regression coefficients from the regression lines of quadratic effects of age predicting each dynamic brain state were plotted. Y-axes were z-scored to facilitate interpretation across graphs. For A and B, a negative value represents lower frequency of occurrence compared with the average, whereas positive values represent greater frequency of occurrence compared with the average. (*A*) A positive quadratic relationship among age and CAP 3 frequency of occurrence. CAP 3 occurred frequently during childhood, decreased in frequency during young adulthood, and increased in frequency again throughout middle- to older adulthood. CAP 3 consisted of co-activation among the M-CIN (salience) and the M-FPN (default). (*B*) A negative quadratic relationship among age and CAP 5 frequency of occurrence. CAP 5 occurred less frequently during childhood, increased in frequency during young- and middle-adulthood, and decreased again in frequency in older adulthood. CAP 5 consisted of co-activation among the L-FPN (executive) and M-CIN. Lastly, (*C*) A positive quadratic relationship among age and CAP 4 dwell time. CAP 4 exhibited longer dwell times during childhood, shorter dwell times during young- and middle-adulthood, and longer dwell times again in older adulthood. CAP 4 consisted of co-activation among the L-FPN and M-FPN. In *A*, *B*, and *C*, children and older adults had similar brain dynamic patterns for each CAP, whereas young adults had different brain dynamic patterns. For example, in *B*, CAP 5 occurred less frequently in early childhood and older adulthood, but occurred more frequently in early adulthood.

network transitions. We found that a greater M-FPN/L-FPN dwell time in children and older adults was associated with poorer cognitive flexibility. Furthermore, greater brain state transitions in children and older adults was associated with better cognitive flexibility, consistent with prior observations (Grady and Garrett 2014; Battaglia et al. 2020). Mid-adulthood, however, was associated with different dynamic patterns associated with optimal cognitive flexibility. This age represents a change in cognition from greater fluid to semantic abilities (Park et al. 2001). Our findings suggest children and older adults are most vulnerable to cognitive flexibility deficits, however, a "deficit" in children is defined by having worse cognitive flexibility compared with age-matched peers, with the potential of improvement in adulthood. Cognitive inflexibility in children and adults was associated with brain dynamic alterations among the M-CIN, M-FPN, and L-FPN based on time spent in the hybrid M-FPN/L-FPN state and variability in state transitions.

U-Shaped Trajectories of between-Network Dynamics

Previous studies have demonstrated quadratic effects of age associated with between-network connections (Betzel et al. 2014; Cao et al. 2014). Prior studies are consistent with our findings of quadratic or U-shaped trajectories in between-network dynamics among three large-scale brain networks of the M-CIN, L-FPN, and M-FPN (Chen et al. 2018). We found the brain state consisting of co-activation of the M-CIN and M-FPN (CAP 3) decreased in frequency of occurrence during middle adulthood but increased during both childhood and older adulthood. Functional connectivity between the M-CIN and M-FPN has been previously shown to be associated with greater cognitive control (Jilka et al. 2014), behavioral performance on cognitive tasks (Putcha et al. 2016), and memory in older adults (Zhang et al. 2020b). Additionally, there is evidence that coupling between the M-FPN and M-CIN may be an intermediary "switching mechanism" prior to later M-FPN and L-FPN coupling (Beaty et al. 2016), potentially underlying greater use of semantic or crystallized knowledge (Spreng and Turner 2019).

Previous work examined M-CIN and M-FPN connections using static functional connectivity approaches, whereas we explored the relationship using dynamic or time-varying methods. Therefore, dynamic interactions between the M-CIN and M-FPN may be critical to further assess in relation to previous static functional connectivity findings. Furthermore, we expand upon previous findings by demonstrating increased

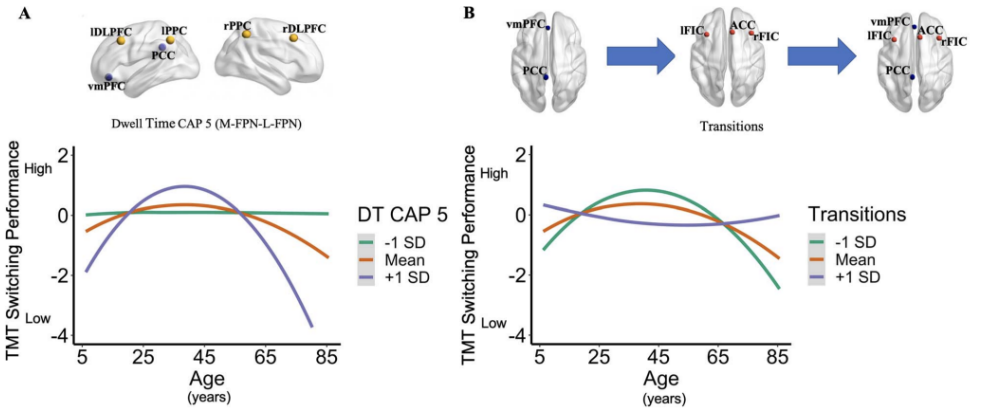

Figure 3. Brain dynamics moderate the relationship between age and cognitive flexibility: simple slopes. The interactions presented in *A* and *B* were between Age^2 and the brain dynamic metrics for CAP 5 and brain state transitions; however, they are presented across age (years) for visual purposes. Additionally, the simple slopes for both interactions are presented to visually determine the effect of age on the cognitive flexibility measure across three different levels of the moderator as represented by the brain dynamic metric (i.e., −1 SD below the mean, at the mean, and +1 SD above the mean). Additionally, the *y*-axes were reversed and standardized so better cognitive flexibility is at higher ends (the top) and poorer cognitive flexibility is at lower ends (the bottom) of the *y*-axes. (**A**) The CAP 5 dwell time (DT) moderated the relationship between Age^2 and the TMT Switching condition (total time to complete the task) as represented by the simple slopes. CAP 5 is characterized by co-activation among the L-FPN (executive control) and M-FPN (default). Children and older adults who spent a longer time in CAP 5 had poorer cognitive flexibility, whereas younger adults had optimal cognitive flexibility regardless of their CAP 5 brain dynamics. Similar findings were seen at average levels of CAP 5 dwell time. Children and older adults who spent less time in CAP 5 had optimal cognitive flexibility relative to those with average and greater time spent in CAP 5, whereas younger adults had poorer, yet still optimal, cognitive flexibility. (**B**) The number of transitions moderated the relationship between Age^2 and the TMT switching condition (total time to complete the task) as represented by the simple slopes. Children and older adults who had fewer brain state transitions had poorer cognitive flexibility, whereas younger adults had optimal cognitive flexibility at average and fewer transitions. Similar findings were seen across individuals with average numbers of brain state transitions. Children and older adults with greater brain state transitions had optimal cognitive flexibility relative to those with average and fewer brain state transitions, whereas young adults had poorer cognitive flexibility.

dynamic interactions or frequency of occurrence of the M-CIN and M-FPN state is associated with older age and development. This may be due to its role as an intermediary switching mechanism prior to M-FPN and L-FPN connections, which is greater in older adults (Spreng and Turner 2019). Thus, M-CIN/M-FPN coupling may occur more frequently prior to M-FPN/L-FPN coupling. Within- and between-brain network integration increases with age, therefore, brain network variability between certain brain networks may be greater in children due to less integration (Gu et al. 2015; Kundu et al. 2018). Furthermore, connectivity with the M-FPN is important for brain network development (Dosenbach et al. 2010). Together, the M-CIN/M-FPN hybrid state exhibits a quadratic trend across the lifespan, and children and older adults may be more likely to enter this state prior to engaging other functional configurations.

Similarly, we found the co-activation between the L-FPN and M-CIN (CAP 4) decreased in dwell time during middle adulthood and increased during childhood and older adulthood. The effect size for this finding was moderate (β = 0.37) (Schäfer and Schwarz 2019). Previous work demonstrates the M-CIN may independently act as a switching mechanism between the M-FPN and L-FPN (Goulden et al. 2014). In children and older adults, a longer time was spent in the L-FPN/M-CIN state during a task-free environment, suggesting the M-CIN related switching mechanism may not be fully developed in children (Uddin et al. 2011), and may be "stickier" or less efficient in older adults. Conversely, middle-aged-adults dwelled less in this state, potentially due to having greater brain state transitions and variability than children and older adults (Grady and Garrett 2014; Ryali et al. 2016; Xia et al. 2019).

Lastly, we found the connection between the L-FPN and M-FPN decreased in frequency of occurrence during childhood and older adulthood and increased during middle adulthood. Although reliance on semantic knowledge and subsequently greater M-FPN/L-FPN connections is not as prevalent in mid-adulthood, evidence suggests mid-adulthood is characterized by the intersection of greater reliance on semantic knowledge while fluency abilities are still retained (Park et al. 2001; Spreng and Turner 2019). Therefore, the M-FPN/L-FPN state may still occur in middle adulthood and may occur more frequently due to there being more flexible brain dynamics compared with older adults and children.

Together, our results demonstrate that hybrid between-network dynamics in certain brain states exhibit quadratic relationships across age, and may underlie the cognitive changes observed through development and aging. Our results are in line with behavioral studies of cognitive flexibility, which reveal cognitive flexibility takes an inverted U-shaped trend across the lifespan (Cepeda et al. 2001; Zelazo et al. 2014). Cognitive flexibility increases throughout childhood and into adulthood, and declines in older age (Cepeda et al. 2001; Zelazo et al. 2014). Although the frontoparietal regions are overall thought to support these changes (Gogtay et al. 2004; Luna et al. 2010), we extend this prior work by revealing between-network dynamic coupling among the M-CIN, M-FPN, and L-FPN

may also facilitate changes associated with cognitive flexibility across aging.

Brain Dynamics as a Moderator of Age and Cognitive Flexibility: L-FPN and M-FPN

We examined brain dynamics as a moderator between quadratic effects of age and cognitive flexibility to directly examine how brain dynamics among networks impact the relationship between aging and cognitive flexibility (Dajani and Uddin 2015). First, the brain state characterized by co-activation among the L-FPN and M-FPN moderated the relationship between the quadratic effect of age and cognitive flexibility as measured by the TMT. This finding was also replicated using more regions of interest within the M-FPN, L-FPN, and M-CIN (see Supplementary Materials). Emerging evidence suggests that greater connectivity between the M-FPN and L-FPN is a central feature of neurocognitive aging (Spreng and Schacter 2012; Turner and Nathan Spreng 2015; Spreng et al. 2018; Adnan et al. 2019a; Adnan et al. 2019b), termed the "default-executive coupling hypothesis of aging" (DECHA) (Turner and Nathan Spreng 2015; Spreng et al. 2018). Relatedly, at each end of the lifespan, behavioral evidence suggests that cognitive flexibility performance is poorer in both childhood and older adulthood (Cepeda et al. 2001; Ridderinkhof et al. 2002; Wasylyshyn et al. 2011; Dajani and Uddin 2015).

Consistent with DECHA and behavioral evidence associated with cognitive flexibility across aging, we show individuals with greater M-FPN/L-FPN dwell time, or individuals with less modulation of the M-FPN and L-FPN, perform worse on cognitive flexibility tasks than older individuals with average or shorter CAP 5 (M-FPN/L-FPN) dwell time. Although the DECHA model has primarily been applied to older individuals, we additionally found evidence that a greater CAP 5 dwell time is associated with cognitive *in*flexibility during childhood. This may contribute to the poorer performance on cognitive flexibility tasks observed during childhood (Dick 2014; Buttelmann and Karbach 2017). Additionally, previous evidence suggests there is less flexibility among the M-CIN, M-FPN, and L-FPN during childhood (Ryali et al. 2016). Our results extend this finding by relating reduced network flexibility (M-FPN and L-FPN) with reduced cognitive flexibility. Furthermore, our findings suggest that older adults are more severely impacted by reduced M-FPN/L-FPN modulation than children. Overall, our findings support the DECHA model of aging, and extend previous work by revealing M-FPN/L-FPN coupling is associated with cognitive flexibility during both childhood and aging.

Furthermore, our results demonstrate different neural patterns associated with cognitive flexibility during mid-adulthood compared with older adults and children. This finding suggests that a greater M-FPN/L-FPN dwell time may be beneficial to cognitive flexibility during mid-adulthood. Additionally, average and reduced M-FPN/L-FPN dwell time during mid-adulthood were also associated with higher levels of cognitive flexibility. Mid-adulthood has previously been shown as a turning point of declining cognitive control and increased reliance on semantic (crystallized) knowledge (Park et al. 2001). However, there is evidence that fluid skills are declining yet intact, while semantic knowledge is increasing, and may actually bolster cognition (Li et al. 2015; Samanez-Larkin and Knutson 2015). Therefore, mid-adulthood has been seen as an optimal period for decision-making (Samanez-Larkin and Knutson 2015; Spreng and Turner 2019) due to the ability to integrate both fluid and semantic knowledge. Overall, our results reflect this idea and demonstrate mid-adulthood is associated with optimal cognitive flexibility that may additionally be aided by semantic knowledge.

Brain Dynamics as a Moderator of Age and Cognitive Flexibility: Transitions

We found that the number of brain state transitions moderated the relationship between a quadratic effect of age and cognitive flexibility. This finding was also replicated using additional regions of interest within the M-FPN, L-FPN, and M-CIN (see Supplementary Materials). Specifically, a greater number of brain state transitions was associated with stable or high cognitive flexibility across the lifespan. As expected, average and lower number of brain state transitions were associated with poorer cognitive flexibility during childhood and older adulthood, consistent with the literature (Hutchison and Morton 2015; Xia et al. 2019; Battaglia et al. 2020). Our findings suggest that the childhood and the older adulthood stages of life are most vulnerable to reduced brain state transitions associated with poorer cognitive flexibility compared with mid-adulthood. This finding has implications for development during both childhood and older adulthood. Overall, our findings demonstrate direct relationships between brain dynamics associated with age and cognitive flexibility changes across the lifespan (Uddin 2021).

Conclusion

Using CAP analysis, we identified brain states characterized by between- and within-network connectivity of neural networks important for cognitive flexibility. We discovered that between-network dynamics of a state characterized by co-activation among the M-FPN and L-FPN, and brain state transitions, moderated the relationship between aging and cognitive flexibility. Our results reveal dynamic brain mechanisms contributing to poorer cognitive flexibility in youth and older individuals. Preventative measures and interventions should prioritize strategies targeting brain dynamics among the M-CIN, M-FPN, and L-FPN, and focus on cognitive flexibility training to promote optimal outcomes across the lifespan.

Supplementary Material

Supplementary material can be found at *Cerebral Cortex* online.

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Notes

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References

Adnan A, Beaty R, Lam J, Nathan Spreng R, Turner GR. 2019a. Intrinsic default—executive coupling of the creative aging brain. *Soc Cogn Affect Neurosci*. 14:291–303.

Adnan A, Beaty R, Silvia P, Spreng RN, Turner GR. 2019b. Creative aging: functional brain networks associated with divergent thinking in older and younger adults. *Neurobiol Aging*. 75:150–158.

Battaglia D, Boudou T, Hansen ECA, Lombardo D, Chettouf S, Daffertshofer A, McIntosh AR, Zimmermann J, Ritter P, Jirsa V. 2020. Dynamic functional connectivity between order and randomness and its evolution across the human adult lifespan. *Neuroimage*. 222:117156.

Beaty RE, Benedek M, Silvia PJ, Schacter DL. 2016. Creative cognition and brain network dynamics. *Trends Cogn Sci*. 20:87–95.

Betzel RF, Byrge L, He Y, Goñi J, Zuo X-N, Sporns O. 2014. Changes in structural and functional connectivity among resting-state networks across the human lifespan. *Neuroimage*. 102(Pt 2):345–357.

Bohnen N, Jolles J, Twijnstra A. 1992. Modification of the stroop color word test improves differentiation between patients with mild head injury and matched controls. *Clin Neuropsychol*. 6:178–184.

Braun U, Schäfer A, Walter H, Erk S, Romanczuk-Seiferth N, Haddad L, Schweiger JI, Grimm O, Heinz A, Tost H, et al. 2015. Dynamic reconfiguration of frontal brain networks during executive cognition in humans. *Proc Natl Acad Sci U S A*. 112:11678–11683.

Burt KB, Paysnick AA. 2012. Resilience in the transition to adulthood. *Dev Psychopathol*. 24:493–505.

Buttelmann F, Karbach J. 2017. Development and plasticity of cognitive flexibility in early and middle childhood. *Front Psychol*. 8:1040.

Cao M, Wang J-H, Dai Z-J, Cao X-Y, Jiang L-L, Fan F-M, Song X-W, Xia M-R, Shu N, Dong Q, et al. 2014. Topological organization of the human brain functional connectome across the lifespan. *Dev Cogn Neurosci*. 7:76–93.

Cepeda NJ, Kramer AF, Gonzalez de Sather JC. 2001. Changes in executive control across the life span: examination of task-switching performance. *Dev Psychol*. 37:715–730.

Chand GB, Wu J, Hajjar I, Qiu D. 2017. Interactions of the salience network and its subsystems with the default-mode and the central-executive networks in normal aging and mild cognitive impairment. *Brain Connect*. 7:401–412.

Chen T, Cai W, Ryali S, Supekar K, Menon V. 2016. Distinct global brain dynamics and spatiotemporal Organization of the Salience Network. *PLoS Biol*. 14:e1002469.

Chen Y, Zhao X, Zhang X, Y'nan L, Zhou P, Ni H, Ma J, Ming D. 2018. Age-related early/late variations of functional connectivity across the human lifespan. *Neuroradiology*. 60:403–412.

Ciric R, Rosen AFG, Erus G, Cieslak M, Adebimpe A, Cook PA, Bassett DS, Davatzikos C, Wolf DH, Satterthwaite TD. 2018. Mitigating head motion artifact in functional connectivity MRI. *Nat Protoc*. 13:2801–2826.

Cohen JR. 2018. The behavioral and cognitive relevance of time-varying, dynamic changes in functional connectivity. *Neuroimage*. 180:515–525.

Colé P, Duncan LG, Blaye A. 2014. Cognitive flexibility predicts early reading skills. *Front Psychol*. 5:565.

Computing R, Others. 2013. *R: A language and environment for statistical computing*. Vienna: R Core Team.

Cox RW. 1996. AFNI: software for analysis and visualization of functional magnetic resonance Neuroimages. *Comput Biomed Res*. 29:162–173.

Dajani DR, Uddin LQ. 2015. Demystifying cognitive flexibility: implications for clinical and developmental neuroscience. *Trends Neurosci*. 38:571–578.

Damoiseaux JS, Rombouts SARB, Barkhof F, Scheltens P, Stam CJ, Smith SM, Beckmann CF. 2006. Consistent resting-state networks across healthy subjects. *Proc Natl Acad Sci U S A*. 103:13848–13853.

Davis JC, Marra CA, Najafzadeh M, Liu-Ambrose T. 2010. The independent contribution of executive functions to health related quality of life in older women. *BMC Geriatr*. 10:8–16.

Delis DC, Kaplan E, Kramer JH. 2001. *Delis-Kaplan Executive Function System*.

de Paula JJ, Paiva GC de C, Costa D de S. 2015. Use of a modified version of the switching verbal fluency test for the assessment of cognitive flexibility. *Dement Neuropsychol*. 9:258–264.

Diamond A. 2013. Executive functions. *Annu Rev Psychol*. 64:135–168.

Dick AS. 2014. The development of cognitive flexibility beyond the preschool period: an investigation using a modified flexible item selection task. *J Exp Child Psychol*. 125:13–34.

Dosenbach NUF, Nardos B, Cohen AL, Fair DA, Power JD, Church JA, Nelson SM, Wig GS, Vogel AC, Lessov-Schlaggar CN, et al. 2010. Prediction of individual brain maturity using fMRI. *Science*. 329:1358–1361.

Douw L, Wakeman DG, Tanaka N, Liu H, Stufflebeam SM. 2016. State-dependent variability of dynamic functional connectivity between frontoparietal and default networks relates to cognitive flexibility. *Neuroscience*. 339:12–21.

DuPre E, Nathan Spreng R. 2017. Structural covariance networks across the life span, from 6 to 94 years of age. *Network Neuroscience*. 1:302–323.

Espinoza FA, Vergara VM, Damaraju E, Henke KG, Faghiri A, Turner JA, Belger AA, Ford JM, McEwen SC, Mathalon DH, et al. 2019. Characterizing whole brain temporal variation of functional connectivity via zero and first order derivatives of sliding window correlations. *Front Neurosci*. 13:634.

Fine EM, Delis DC, Holdnack J. 2011. Normative adjustments to the D-KEFS trail making test: corrections for education and vocabulary level. *Clin Neuropsychol*. 25:1331–1344.

Francis JD, Kerlinger FN, Pedhazur EJ. 1975. Multiple regression in Behavioral research. *Teach Sociol*. 3:103.

Friston KJ, Williams S, Howard R, Frackowiak RSJ, Turner R. 1996. Movement-related effects in fMRI time-series. *Magn Reson Med*. 35:346–355.

Genet JJ, Siemer M. 2011. Flexible control in processing affective and non-affective material predicts individual differences in trait resilience. *Cognit Emot*. 25:380–388.

Gogtay N, Giedd JN, Lusk L, Hayashi KM, Greenstein D, Vaituzis AC, Nugent TF 3rd, Herman DH, Clasen LS, Toga AW, et al. 2004. Dynamic mapping of human cortical development during childhood through early adulthood. *Proc Natl Acad Sci U S A*. 101:8174–8179.

Goulden N, Khusnulina A, Davis NJ, Bracewell RM, Bokde AL, McNulty JP, Mullins PG. 2014. The salience network is responsible for switching between the default mode network and the central executive network: replication from DCM. *Neuroimage*. 99:180–190.

Grady CL, Garrett DD. 2014. Understanding variability in the BOLD signal and why it matters for aging. *Brain Imaging Behav*. 8:274–283.

Grady CL, Springer MV, Hongwanishkul D, McIntosh AR, Winocur G. 2006. Age-related changes in brain activity across the adult lifespan. *J Cogn Neurosci*. 18:227–241.

Griffanti L, Salimi-Khorshidi G, Beckmann CF, Auerbach EJ, Douaud G, Sexton CE, Zsoldos E, Ebmeier KP, Filippini N,

Mackay CE, et al. 2014. ICA-based artefact removal and accelerated fMRI acquisition for improved resting state network imaging. *Neuroimage*. 95:232–247.
Gu S, Satterthwaite TD, Medaglia JD, Yang M, Gur RE, Gur RC, Bassett DS. 2015. Emergence of system roles in normative neurodevelopment. *Proc Natl Acad Sci U S A*. 112:13681–13686.
Hutchison RM, Morton JB. 2015. Tracking the Brain's functional coupling dynamics over development. *J Neurosci*. 35:6849–6859.
Hyder F, Rothman DL. 2010. Neuronal correlate of BOLD signal fluctuations at rest: err on the side of the baseline. *Proc Natl Acad Sci U S A*. 107:10773–10774.
Jia H, Hu X, Deshpande G. 2014. Behavioral relevance of the dynamics of the functional brain connectome. *Brain Connect*. 4:741–759.
Jilka SR, Scott G, Ham T, Pickering A, Bonnelle V, Braga RM, Leech R, Sharp DJ. 2014. Damage to the salience network and interactions with the default mode network. *J Neurosci*. 34:10798–10807.
Kleinhans N, Akshoomoff N, Delis DC. 2005. Executive functions in autism and Asperger's disorder: flexibility, fluency, and inhibition. *Dev Neuropsychol*. 27:379–401.
Kundu P, Benson BE, Rosen D, Frangou S, Leibenluft E, Luh W-M, Bandettini PA, Pine DS, Ernst M. 2018. The integration of functional brain activity from adolescence to adulthood. *J Neurosci*. 38:3559–3570.
Kupis L, Goodman ZT, Kircher L, Romero C, Dirks B, Chang C, Nomi JS, Uddin LQ. 2021. Altered patterns of brain dynamics linked with body mass index in youth with autism. *Autism Res*. 14:873–886.
Kupis L, Romero C, Dirks B, Hoang S, Parlade MV, Beaumont AL, Cardona SM, Alessandri M, Chang C, Nomi JS, et al. 2020. *Evoked and intrinsic brain network dynamics in children with autism spectrum disorder*. Neuroimage: Clinical.
Li JCH. 2018. Curvilinear moderation—a more complete examination of moderation effects in Behavioral sciences. *Frontiers in Applied Mathematics and Statistics*. 4:7.
Liu X, Chang C, Duyn JH. 2013. Decomposition of spontaneous brain activity into distinct fMRI co-activation patterns. *Frontiers in Systems Neuroscience*. 7:101.
Li Y, Gao J, Enkavi AZ, Zaval L, Weber EU, Johnson EJ. 2015. Sound credit scores and financial decisions despite cognitive aging. *Proc Natl Acad Sci U S A*. 112:65–69.
Luna B, Padmanabhan A, O'Hearn K. 2010. What has fMRI told us about the development of cognitive control through adolescence? *Brain Cogn*. 72:101–113.
Marshall E, Nomi JS, Dirks B, Romero C, Kupis L, Chang C, Uddin LQ. 2020. Co-activation pattern analysis reveals altered salience network dynamics in children with autism spectrum disorder. *Network Neuroscience*. 4:1219–1234.
Masten AS, Obradovic J. 2006. Competence and resilience in development. *Ann N Y Acad Sci*. 1094:13–27.
Mattson SN, Goodman AM, Caine C, Delis DC, Riley EP. 1999. Executive functioning in children with heavy prenatal alcohol exposure. *Alcohol Clin Exp Res*. 23:1808–1815.
Mcdonald CR, Delis DC, Norman MA, Tecoma ES, Iragui-Madoz VJ. 2005. Is impairment in set-shifting specific to frontal-lobe dysfunction? Evidence from patients with frontal-lobe or temporal-lobe epilepsy. *J Int Neuropsychol Soc*. 11:477–481.
Menon V, Uddin LQ. 2010. Saliency, switching, attention and control: a network model of insula function. *Brain Struct Funct*. 214:655–667.
Murphy K, Fox MD. 2017. Towards a consensus regarding global signal regression for resting state functional connectivity MRI. *Neuroimage*. 154:169–173.
Nomi JS, Bolt TS, Ezie CEC, Uddin LQ, Heller AS. 2017a. Moment-to-moment BOLD signal variability reflects regional changes in neural flexibility across the lifespan. *J Neurosci*. 37:5539–5548.
Nomi JS, Vij SG, Dajani DR, Steimke R, Damaraju E, Rachakonda S, Calhoun VD, Uddin LQ. 2017b. Chronnectomic patterns and neural flexibility underlie executive function. *Neuroimage*. 147:861–871.
Nooner KB, Colcombe SJ, Tobe RH, Mennes M, Benedict MM, Moreno AL, Panek LJ, Brown S, Zavitz ST, Li Q, et al. 2012. The NKI-Rockland sample: a model for accelerating the pace of discovery science in psychiatry. Front. *Neuroscience*. 6:152.
Park DC, Polk TA, Mikels JA, Taylor SF, Marshuetz C. 2001. Cerebral aging: integration of brain and behavioral models of cognitive function. *Dialogues Clin Neurosci*. 3:151–165.
Power JD, Plitt M, Laumann TO, Martin A. 2017. Sources and implications of whole-brain fMRI signals in humans. *Neuroimage*. 146:609–625.
Putcha D, Ross RS, Cronin-Golomb A, Janes AC, Stern CE. 2016. Salience and default mode network coupling predicts cognition in aging and Parkinson's disease. *J Int Neuropsychol Soc*. 22:205–215.
Qin J, Chen S-G, Hu D, Zeng L-L, Fan Y-M, Chen X-P, Shen H. 2015. Predicting individual brain maturity using dynamic functional connectivity. *Front Hum Neurosci*. 9:418.
Rabany L, Brocke S, Calhoun VD, Pittman B, Corbera S, Wexler BE, Bell MD, Pelphrey K, Pearlson GD, Assaf M. 2019. Dynamic functional connectivity in schizophrenia and autism spectrum disorder: convergence, divergence and classification. *Neuroimage Clin*. 24:101966.
Ramanan S, Narayanan J, D'Souza TP, Malik KS, Ratnavalli E. 2015. Total output and switching in ategory fluency successfully iscriminates Alzheimer's disease from mild cognitive impairment, but not from frontotemporal dementia. *Dementia & neuropsychologia*. 9:251–257.
Ridderinkhof KR, Span MM, van der Molen MW. 2002. Perseverative behavior and adaptive control in older adults: performance monitoring, rule induction, and set shifting. *Brain Cogn*. 49:382–401.
Ryali S, Supekar K, Chen T, Kochalka J, Cai W, Nicholas J, Padmanabhan A, Menon V. 2016. Temporal dynamics and developmental maturation of salience, default and central-executive network interactions revealed by Variational Bayes hidden Markov Modeling. *PLoS Comput Biol*. 12:e1005138.
Samanez-Larkin GR, Knutson B. 2015. Decision making in the ageing brain: changes in affective and motivational circuits. *Nat Rev Neurosci*. 16:278–289.
Scahill RI, Frost C, Jenkins R, Whitwell JL, Rossor MN, Fox NC. 2003. A longitudinal study of brain volume changes in normal aging using serial registered magnetic resonance imaging. *Arch Neurol*. 60:989.
Schaefer A, Kong R, Gordon EM, Laumann TO, Zuo X-N, Holmes AJ, Eickhoff SB, Yeo BTT. 2018. Local-global Parcellation of the human cerebral cortex from intrinsic functional connectivity MRI. *Cereb Cortex*. 28:3095–3114.
Schäfer T, Schwarz MA. 2019. The meaningfulness of effect sizes in psychological research: differences between sub-disciplines and the impact of potential biases. *Front Psychol*. 10:813.

Schölvinck ML, Maier A, Ye FQ, Duyn JH, Leopold DA. 2010. Neural basis of global resting-state fMRI activity. *Proc Natl Acad Sci U S A*. 107:10238–10243.

Seeley WW, Menon V, Schatzberg AF, Keller J, Glover GH, Kenna H, Reiss AL, Greicius MD. 2007. Dissociable intrinsic connectivity networks for salience processing and executive control. *J Neurosci*. 27:2349–2356.

Spreng RN, Nathan Spreng R, Lockrow AW, DuPre E, Setton R, Spreng KAP, Turner GR. 2018. Semanticized autobiographical memory and the default – executive coupling hypothesis of aging. *Neuropsychologia*. 110:37–43.

Spreng RN, Schacter DL. 2012. Default network modulation and large-scale network interactivity in healthy young and old adults. *Cereb Cortex*. 22:2610–2621.

Spreng RN, Turner GR. 2019. The shifting architecture of cognition and brain function in older adulthood. *Perspect Psychol Sci*. 14:523–542.

Stroop JR, Ridley Stroop J. 1992. Studies of interference in serial verbal reactions. *J Exp Psychol Gen*. 121:15–23.

Touroutoglou A, Zhang J, Andreano JM, Dickerson BC, Barrett LF. 2018. Dissociable effects of aging on salience subnetwork connectivity mediate age-related changes in executive function and affect. *Front Aging Neurosci*. 10:410.

Turner GR, Nathan Spreng R. 2015. Prefrontal engagement and reduced default network suppression co-occur and are dynamically coupled in older adults: the default–executive coupling hypothesis of aging. *J Cogn Neurosci*. 27:2462–2476.

Uddin LQ. 2020a. Bring the noise: Reconceptualizing spontaneous neural activity. *Trends Cogn Sci*. 24:734–746.

Uddin LQ. 2020b. Brain mechanisms supporting flexible cognition and behavior in adolescents with autism spectrum disorder. *Biol Psychiatry*. 89:172–183.

Uddin LQ. 2021. Cognitive and behavioural flexibility: neural mechanisms and clinical considerations. *Nat Rev Neurosci*. 22:167–179.

Uddin LQ, Supekar KS, Ryali S, Menon V. 2011. Dynamic reconfiguration of structural and functional connectivity across Core neurocognitive brain networks with development. *J Neurosci*. 31:18578–18589.

Uddin LQ, Yeo BTT, Spreng RN. 2019. Towards a universal taxonomy of macro-scale functional human brain networks. *Brain Topogr*. 32:926–942.

Wang L, Su L, Shen H, Hu D. 2012. Decoding lifespan changes of the human brain using resting-state functional connectivity MRI. *PLoS One*. 7:e44530.

Wasylyshyn C, Verhaeghen P, Sliwinski MJ. 2011. Aging and task switching: a meta-analysis. *Psychol Aging*. 26:15–20.

Xia Y, Chen Q, Shi L, Li M, Gong W, Chen H, Qiu J. 2019. Tracking the dynamic functional connectivity structure of the human brain across the adult lifespan. *Hum Brain Mapp*. 40:717–728.

Yan C-G, Wang X-D, Zuo X-N, Zang Y-F. 2016. DPABI: data processing & analysis for (resting-state) brain imaging. *Neuroinformatics*. 14:339–351.

Yeniad N, Malda M, Mesman J, van IJzendoorn MH, Pieper S. 2013. Shifting ability predicts math and reading performance in children: a meta-analytical study. *Learn Individ Differ*. 23:1–9.

Yochim B, Baldo J, Nelson A, Delis DC. 2007. D-KEFS Trail making test performance in patients with lateral prefrontal cortex lesions. *J Int Neuropsychol Soc*. 13:704–709.

Zelazo PD, Anderson JE, Richler J, Wallner-Allen K, Beaumont JL, Conway KP, Gershon R, Weintraub S. 2014. NIH toolbox cognition battery (CB): validation of executive function measures in adults. *J Int Neuropsychol Soc*. 20:620–629.

Zhang J, Andreano JM, Dickerson BC, Touroutoglou A, Barrett LF. 2020a. Stronger functional connectivity in the default mode and salience networks is associated with youthful memory in Superaging. *Cereb Cortex*. 30:72–84.

Zhang L, Zuo X-N, Ng KK, Chong JSX, Shim HY, Ong MQW, Loke YM, Choo BL, Chong EJY, Wong ZX, et al. 2020b. Distinct BOLD variability changes in the default mode and salience networks in Alzheimer's disease spectrum and associations with cognitive decline. *Nature Scientific Reports*. 10:6457.

CHAPTER 3:

Evoked and Intrinsic Brain Network Dynamics in Children with Autism Spectrum Disorder

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Evoked and intrinsic brain network dynamics in children with autism spectrum disorder

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ABSTRACT

Objective: Brain dynamics underlie flexible cognition and behavior, yet little is known regarding this relationship in autism spectrum disorder (ASD). We examined time-varying changes in functional co-activation patterns (CAPs) across rest and task-evoked brain states to characterize differences between children with ASD and typically developing (TD) children and identify relationships with severity of social behaviors and restricted and repetitive behaviors.

Method: 17 children with ASD and 27 TD children ages 7–12 completed a resting-state fMRI scan and four runs of a non-cued attention switching task. Metrics indexing brain dynamics were generated from dynamic CAPs computed across three major large-scale brain networks: midcingulo-insular (M-CIN), medial frontoparietal (M-FPN), and lateral frontoparietal (L-FPN).

Results: Five time-varying CAPs representing dynamic co-activations among network nodes were identified across rest and task fMRI datasets. Significant Diagnosis × Condition interactions were observed for the dwell time of CAP 3, representing co-activation between nodes of the M-CIN and L-FPN, and the frequency of CAP 1, representing co-activation between nodes of the L-FPN. A significant brain-behavior association between dwell time of CAP 5, representing co-activation between nodes of the M-FPN, and social abilities was also observed across both groups of children.

Conclusion: Analysis of brain co-activation patterns reveals altered dynamics among three core networks in children with ASD, particularly evident during later stages of an attention task. Dimensional analyses demonstrating relationships between M-FPN dwell time and social abilities suggest that metrics of brain dynamics may index individual differences in social cognition and behavior.

1. Introduction

Autism spectrum disorder (ASD) is a prevalent neurodevelopmental condition characterized by deficits in social communication and restricted and repetitive behaviors (American Psychiatric Association, 2013) (RRBs), and associated with atypical brain connectivity (Chen et al., 2017, 2018; Di Martino et al., 2011; Fishman et al., 2018; Keown et al., 2013; Müller and Fishman, 2018; Supekar et al., 2013; Mash et al., 2019). Despite decades of neuroimaging research exploring brain connectivity in ASD, a clear picture linking specific patterns of atypical connectivity to cognitive and behavioral profiles in ASD has yet to emerge (Kana et al., 2014; Falahpour et al., 2016; Rane et al., 2015; Vissers et al., 2012). To date, brain functional connectivity (FC) and co-activation patterns among brain regions as measured with fMRI has primarily been studied using "static" measures (Falahpour et al., 2016; White and Calhoun, 2019). Static FC methods average the entire time series across an fMRI scan, missing the opportunity to characterize moment-to-moment changes in coupling between brain regions (Allen et al., 2014; Calhoun et al., 2014). Recent FC research has used time-varying dynamic approaches that examine how brain function may

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change over time (Chang and Glover, 2010) thereby better capturing flexible aspects of brain systems (Allen et al., 2014).

Various methods exist to study dynamic, time-varying changes in the brain, including dynamic functional connectivity (dFC) and time-varying co-activation pattern analysis (CAP) (see Uddin & Karlsgodt (Uddin and Karlsgodt, 2018) and Uddin (Uddin, 2020) for a review). Dynamic FC most commonly capitalizes on the 'sliding window' approach (Chang and Glover, 2010). Despite its increasing use (Lurie et al., 2020), a limitation of this approach is the examination of FC over a fixed window length over which connectivity may fluctuate (Lurie et al., 2020; Preti et al., 2017). Rather than relying on sliding windows, CAP methods identify critical co-activating patterns that recur over time (Liu and Duyn, 2013). CAP methods seek to identify co-activation patterns by averaging time points with similar spatial distributions of brain activity by using a *k*-means clustering algorithm applied either at the whole-brain or region-of-interest (ROI) level (Liu et al., 2018).

Application of time-varying dynamic analyses to resting-state and task-based fMRI has revealed *brain states,* or recurring patterns of activity or connectivity (Allen et al., 2014; Liu et al., 2018), that can be quantified using metrics such as dwell time, frequency of occurrence, and number of transitions between states. Emerging evidence suggests that flexible resting-state dynamics underlies behavioral adaptation, enhancing the ability of the brain to dynamically reconfigure (Allen et al., 2014; Bassett et al., 2011; Jia et al., 2014). Similarly, the degree of brain network reconfiguration during a cognitive task has been demonstrated to relate to cognitive flexibility, or the ability to selectively switch between mental processes and respond behaviourally (Braun et al., 2015; Dajani and Uddin, 2015). Additional work comparing rest and task-based fMRI data has led to discoveries of network commonalities between the two, but also task-specific network changes (Bolt et al., 2017; Cole et al., 2014).

Multiple studies indicate that dynamic brain states may be important for uncovering novel insights into various psychiatric disorders, including ASD (Buckley et al., 2015; Uddin et al., 2015; Barttfeld et al., 2012). State-specific changes across resting and task fMRI paradigms may provide a more precise characterization of brain connectivity abnormalities in ASD, yet little research to date has concurrently examined both task and resting-state fMRI dynamics in ASD (Uddin et al., 2015). For example, Barttfeld et al. (Barttfeld et al., 2012) found that changes in the pattern of functional connectivity between individuals with ASD and neurotypical individuals were state-dependent (interoceptive and exteroceptive states). Additionally, the classification of static FC using a support vector machine algorithm based on the difference between states outperformed classification using connectivity of a single state (Barttfeld et al., 2012). The few resting-state fMRI studies of brain dynamics have found atypicalities in individuals with ASD using whole brain dFC. You et al. (You et al., 2013) found children with ASD had transitions between unconstrained resting states to sustained attention states characterized by widespread functional connectivity among frontal and parietal regions in addition to atypical modulation of distant connectivity during sustained attention relative to rest. Hypervariant dynamic connections have been identified in youth with ASD (Chen et al., 2017, 2018; Mash et al., 2019; Falahpour et al., 2016), with associations to symptom severity in the domains of both RRBs and social functioning (Chen et al., 2017, 2018; He et al., 2018). Decreased state transitions and longer dwell times have also been reported in children with ASD (de Lacy et al., 2017; Yao et al., 2016; Rashid et al., 2018). Crucially, higher levels of ASD symptoms are associated with longer dwell times and fewer transitions in globally disconnected states (Rashid et al., 2018; Watanabe and Rees, 2017). These results suggest that infrequent brain state switching and hypervariant dynamic connections might underlie the behavioral difficulties seen in ASD (Falahpour et al., 2016; Harlalka et al., 2019).

Previous work has focused on whole-brain time-varying changes, yet recent work has highlighted the importance of understanding transient patterns within specific large-scale brain networks (Ciric et al., 2017). Specific brain areas have been identified in subserving flexible behavior, including the midcingulo-insular network (M-CIN or salience network), which mediates switches between the lateral frontoparietal network (L-FPN or central executive network) and the medial frontoparietal network (M-FPN, or default mode network) (Uddin et al., 2015, 2019). In ASD, it has been demonstrated that weak modulation of brain states among these networks is associated with the severity of RRBs (Uddin et al., 2015). Using time-varying approaches, atypical dynamic interactions among these regions have additionally been related to social deficits (He et al., 2018). Further work has demonstrated that decreased switching between brain states among the M-FPN and L-FPN occurs within ASD populations and may be related to behavioral inflexibility (de Lacy et al., 2017). Despite emerging evidence that M-FPN, M-CIN, and L-FPN regions are critically involved in ASD pathology, no studies have directly compared task-related (evoked) and resting-state (intrinsic) time-varying relationships in ASD among these three core neurocognitive networks.

Here we examine co-activation patterns among six key nodes of the M-CIN, M-FPN, and L-FPN in children with and without ASD during both task and resting states for the first time. We hypothesized that children with ASD and typically developing (TD)/neurotypical children would exhibit differences in dynamic brain state metrics such as frequency of occurrence and dwell time across rest and task conditions. We further expected to find relationships between metrics of brain dynamics and parent-report measures of RRBs and social behaviors.

2. Methods

2.1. Participants

Participant enrollment included 35 children with ASD and 36 TD children recruited from the University of Miami and the University of Miami Center for Autism and Related Disabilities (CARD, http://www.umcard.org/). Exclusionary criteria included 1) less than 10 min of resting-state fMRI data 2) less than 4 usable task-fMRI runs 3) incidental findings. Subjects were additionally excluded if they had > 1 mm mean framewise displacement (FD) or failed a visual Quality Control inspection indicating that they had one or more visually identifiable artifacts including but not limited to: excessive motion, ringing, blurring, ghosting, wrapping, signal loss, and head coverage. This resulted in a final sample of 17 children with ASD (M = 9.95, SD = 1.51) and 27 TD children (M = 9.79, SD = 1.88) that did not differ significantly in gender, age, full scale IQ, and mean FD (p's > 0.05) (Table 1) (Power et al., 2014).

All participants underwent an initial phone screening followed by 1) neuropsychological assessment at the University of Miami Autism Spectrum Assessment Clinic (ASAC, http://www.umasac.org/) within CARD, and a 2) mock MRI scanner training followed by functional and structural brain imaging and completion of questionnaire forms. ASD participants were also administered the Autism Diagnostic Observation Schedule, Second Edition (ADOS-2) (Lord et al., 2012) by research-reliable examiners at the University of Miami ASAC. All participants were MRI compatible, able to perform the task, had a full-scale IQ > 65 as measured by the Wechsler Abbreviated Scale of Intelligence-Second Edition (WASI-II) (Wechsler, 2011), and were right-handed. Inclusion criteria for ASD participants included a previous diagnosis of ASD based on the DSM-5 criteria (American Psychiatric Association, 2013) by a community neurologist, psychologist, or other medical/mental health professional and meeting the cut-off for autism or autism spectrum on the ADOS-2, Module 3. See Table 1 for participant information. This study was approved by the Institutional Review Board at the University of Miami and conducted in compliance with the Declaration of Helsinki. All participants provided written informed consent and received financial compensation for their participation.

Table 1
Participant Demographics.

N = 44	Diagnostic Group		
	TD (n = 27)	ASD (n = 17)	
	Mean (*SD*)	Mean (*SD*)	p value
Sex	18 M/9F	14 M/3F	0.343
Age	9.79 (1.88)	9.95 (1.56)	0.489
range	[7.08–12.92]	[8.17–12.67]	–
Race [a]	0, 1, 1, 18, 4, 3	0, 1, 1, 13, 1, 1	0.843
Ethnicity, *Hispanic/Latino*	17	11	0.041
FSIQ [b]	107.88 (10.77)	106.9 (16.48)	0.836
range	[90–133]	[74–132]	–
Motion [c]			
Rest FD	0.146 (0.118)	0.148 (0.086)	0.958
Task 1 FD	0.145 (0.096)	0.110 (0.039)	0.160
Task 2 FD	0.151 (0.122)	0.143 (0.076)	0.813
Task 3 FD	0.195 (0.091)	0.227 (0.176)	0.420
Task 4 FD	0.197 (0.124)	0.221 (0.195)	0.618
SRS-2, *T score*	45.08 (4.57)	69.24 (12.34)	< 0.001
RBS-R, *T score*	2.148 (2.957)	15.941 (11.882)	< 0.001
ADOS-2			
Social Affect	–	8.56 (3.35)	–
Restricted and Repetitive Behaviors	–	2.00 (1.10)	–
Comparison Score	–	6.31 (1.54)	–

Note: FSIQ: Full Scale Intelligence Quotient; SRS-2: Social Responsiveness Scale, Second Edition; RBS-R: Repetitive Behaviors Scale-Revised; ADOS-2: Autism Diagnostic Observation Schedule-Second Edition

1. Numbers for each of the following racial categories presented in the following order: African American, Asian, Biracial, Caucasian, Other, Not Reported.
2. FSIQ: WASI-II full-scale IQ, 4 participants did not have WASI-II because WISC-V was administered within a year and had IQ > 65.
3. Power framewise displacement for raw rs-fMRI data calculated in dpabi.

2.2. Neuropsychological measures and assessments

The first session included a visit to the ASAC, where the WASI-II, a standardized measure of intelligence that provides three measures of IQ: Verbal, Performance, and Full (Wechsler, 2011), and ADOS-2, a standardized measure of communication, social interaction, play, and RRBs (Lord et al., 2012), were administered. These assessments were administered by licensed clinical psychologists who had previously achieved research-reliability on the ADOS-2.

Parents or caregivers completed the SRS-2 (Constantino and Gruber, 2012) and RBS-R (Lam and Aman, 2007), used to assess social abilities and RRBs continuously and quantitatively. The SRS-2 is a 65-item parent report measure that yields a total T-score indicating overall social ability. The RBS-R is a 44-item parent report measure that yields a total T-score indicating overall repetitive behaviors. The SRS-2 and RBS-R total raw scores were converted to age equivalent T-scores, with higher scores indicating more severe impairment (Constantino and Gruber, 2012; Lam and Aman, 2007).

2.3. fMRI data acquisition parameters

Children initially participated in a mock scan to adjust to the scanning environment and to practice the fMRI task. MRI data were acquired using a 3 T GE scanner with a 32-channel head coil. Functional images were collected using a gradient echo sequence (TR/TE/flip angle/FOV = 2 s/30 ms/75°/220 mm; orientation: 42 axial slices angled along the AC-PC; slice thickness: 3.4 mm no inter-slice skip, interleaved acquisition order and anterior-posterior encoding). At the beginning of the scanning session, participants completed a 10-min resting-state run consisting of 295 volumes. They were instructed to lie still with their eyes closed while remaining awake. The resting-state run was followed by four 122- vol task runs. The first 5 volumes of each run were discarded to account for gradient stabilization.

2.4. Non-cued attention switching task

Participants completed four runs of a task examining the ability to shift attention between stimulus dimensions (Casey et al., 2004; Britton et al., 2010). While in the scanner, participants viewed a display with three stimuli presented on a black background. One of the three objects differed from the other two in either shape (S), (e.g., square or circle) or color (C), (e.g., gray or white) (Fig. 1). Participants were instructed to identify the differing object by pressing a button corresponding to the unique object. They were not explicitly told how the objects differed (S or C). (See Supplement 1 for further details).

On each trial, three objects were presented for 1000 ms with a 1500 ms interstimulus interval (ISI). One object had a unique attribute, either shape (square or circle) or color (gray or white). Participants indicated the location of the unique object via a button press. The stimuli were presented in a blocked design with 12 trials per shape/color and 24 trials per mixed block. Each run lasted 4 min and 16 s (see Dirks et al. (Dirks et al., 2020) for further details).

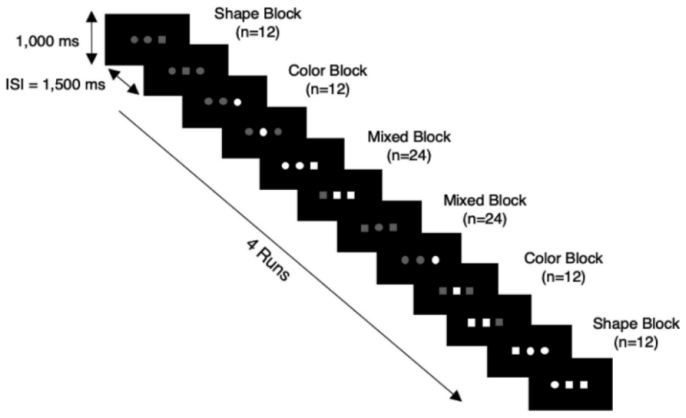

Fig. 1. Task Paradigm.

3. Data analysis

3.1. Behavioral data

Total accuracy was computed across all trials in each block and reported as the proportion of correct trials relative to the total number of trials the subject completed. Reaction time (RT) was calculated for each subject for correct trials only and computed as a total mean RT across all correct trials in each block (see supplementary Table S1). Accuracy and RT were analyzed using a 2 Diagnosis (ASD, TD) × 4 Condition (task run 1, task run 2, task run 3, task run 4) mixed-model ANOVA.

3.2. fMRI data preprocessing and region-of-interest (ROI) selection

The resting-state run and four task runs were preprocessed separately using the Data Processing and Analysis for Brain Imaging (DPABI) version 3.1 toolbox (http://rfmri.org/dpabi) (Yan et al., 2016). The following steps were completed in the same order for all task and rest datasets: despiking (AFNI's 3dDespike), slice timing correction (Parker and Razlighi, 2019), realignment, brain extraction, segmentation, normalization to a standard SPM EPI template (3 × 3 × 3 mm), and smoothing (FWHM = 6 mm). Despiking identifies voxelwise TR outliers > 2.5 standard deviations of the time series and replaces them with an adjusted value based on the mathematical formula: s' = c1 + (c2-c1)*tanh((s-c1/(c2-c1)) where c1 = 2.5, c2 = 4, s = original TR value, s' = replaced TR value. Despiking was chosen over other censoring methods to preserve temporal continuity in the rest and task data.

Six ROIs were selected, including the right fronto-insular cortex (rFIC) and anterior cingulate cortex (ACC) of the M-CIN; right dorsolateral prefrontal cortex (rDLPFC) and right posterior parietal cortex (rPPC) of the L-FPN; and the ventromedial prefrontal cortex (VMPFC) and posterior cingulate cortex (PCC) of the M-FPN. Coordinates delineating these ROIs in a previous study were used (Table S2) (Uddin et al., 2011).

3.3. Independent component analysis (ICA) denoising

Each subject's rest and task fMRI datasets were individually denoised by hand-classifying ICA components after running FSL's Melodic ICA algorithm with automatic dimensionality estimation. Components identified as noise (e.g. those containing artifacts such as white matter, cerebrospinal fluid, head motion, or proportionally large amounts of high-frequency information) were regressed out of the data prior to subsequent post-processing using the fsl_regfilt command (Griffanti et al., 2017; Jenkinson et al., 2012).

3.4. Post-ICA processing and analysis

After ICA denoising, the average time series were extracted from 6-mm radius spheres of six ROIs in key nodes of the M-CIN, M-FPN, and L-FPN. Time courses were then linearly detrended, low pass filtered (0.01–0.1 Hz), and subjected to regression of the Friston 24 head motion parameters (6 motion parameters of each volume, the preceding volume, and the 12 corresponding squared items) (Friston et al., 1996), white matter, and CSF, as calculated in the DBAPI toolbox (Yan et al., 2016).

3.5. Co-activation pattern (CAP) analysis

Time series extracted from the six ROIs during task and resting-state runs were converted to z statistics and then concatenated into a single group matrix (787 TR × 44 subjects) following previous studies (Hutchison and Morton, 2015; Denkova et al., 2019). The concatenated matrix was subjected to *k*-means clustering. Testing values of k = 2–20, the optimal value of k = 5 was determined using the elbow criterion by applying a least-squares fit line to the cluster validity index, defined as the ratio of within-cluster to between-cluster differences (**Figure S1**) (Allen et al., 2014; Damaraju et al., 2014). A CAP analysis was conducted using *k*-means clustering (squared Euclidean distance) using the optimal k of 5 on the group concatenated time series of the 6 ROIs across all subjects (Liu et al., 2013). CAP metrics were then calculated separately for each of the five conditions (rest run, task run 1, task run 2, task run 3, task run 4) and for all task runs combined (task all = task runs 1–4). The CAP metrics computed included a) dwell time (DT), calculated as the average number of TRs that a participant stayed in a given brain state in each condition b) frequency of occurrence of brain states, calculated as a percent over time that the brain state occurred throughout the duration of each condition, and c) the number of transitions, calculated as the number of switches between brain states.

3.6. Statistical analysis

DT and frequency of occurrences were subjected to a 2 Diagnosis (ASD, TD) × 5 Condition (rest run, task run 1, task run 2, task run 3, task run 4) mixed model ANOVA. DT and frequency of occurrences were additionally subjected to a 2 Diagnosis (ASD, TD) × 2 Condition (rest run, task all) mixed model ANOVA. Post-hoc two-tailed *t*-tests were conducted to identify differences in the means for each of the runs. Number of transitions during rest were subjected to a *t*-test, and task run transitions were analyzed using a 2 Diagnosis (ASD, TD) × 4 Condition (task run 1, task run 2, task run 3, task run 4) mixed model ANOVA. (See Supplement 1 for details regarding analyses of confounding variables).

3.7. Brain-behavior analysis

The relationship between brain state metrics and social and repetitive behaviors were assessed by calculating Pearson correlations between the CAP metrics (DT, frequency of occurrences, and transitions) and SRS-2 and the RBS-R T-scores in dimensional analyses across all subjects. We additionally calculated partial Pearson correlations between DT and SRS-2 while controlling for age.

4. Results

4.1. Time-varying resting-state and task fMRI

CAP analyses revealed five brain states that dynamically occurred during rest and task runs (Fig. 2). CAP 1 was characterized by co-activation among the nodes of the L-FPN. CAP 2 was characterized by co-activation among the nodes of the M-CIN. CAP 3 was characterized by co-activation among the nodes of the M-CIN and the nodes of the L-FPN. CAP 4 was characterized by co-activation among the nodes of the M-FPN, the L-FPN, and M-CIN. CAP 5 was characterized by co-activation among the nodes of the M-FPN.

4.2. Behavioral

For RT, a mixed model ANOVA revealed there was a significant linear effect of Condition ($F(1,34) = 13.580$, $p = 0.001$). Pairwise comparisons between runs showed that the RTs for task run 1 were significantly higher (slower) than both task run 3 and 4 (p's < 0.05). There were no significant differences between RTs of task runs 1 and 2, 2 and 3, and 2 and 4 (p's > 0.05). There were no significant interactions for RT, and RT did not significantly differ by diagnostic group (p's > 0.05) (Fig. 3**A**).

Mean accuracy was greater than 90% for each run in both ASD and TD groups (See supplementary Table S1). A mixed model ANOVA revealed that there were no significant main effects or interactions for accuracy (p's > 0.05) (Fig. 3**B**).

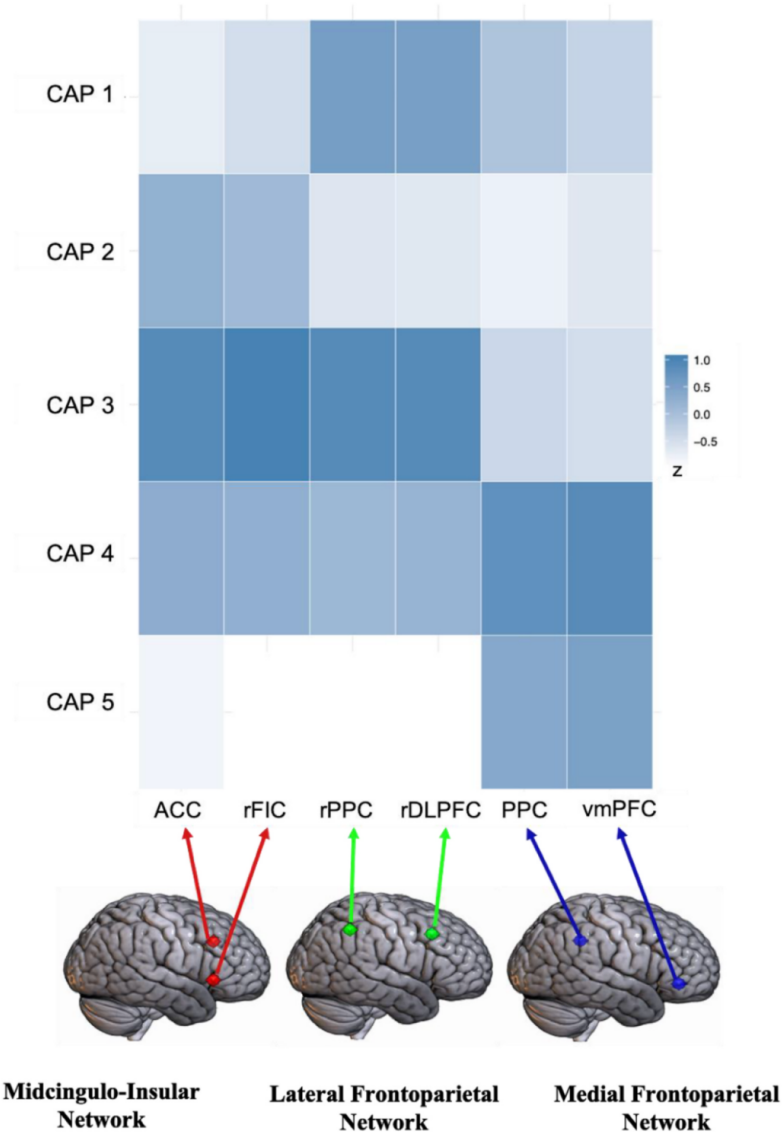

Fig. 2. Time series from 6 regions of interest (ROIs) were extracted across rest and task runs for both children with ASD and TD children, and z-scored and concatenated into a single matrix. The matrix was subjected to *k*-means clustering and CAP analysis using a *k* of 5. The intensity of colors in the CAP matrix indicate the z-scored activation value of the ROIs within each centroid.

4.3. CAP frequency of occurrence

A mixed model ANOVA revealed a significant linear interaction for the frequency of occurrence of CAP 1, the state with co-activation among nodes of the L-FPN, [$F(1,42) = 6.512$, $p = 0.014$], (Fig. 3**C**). Thus, the occurrences of CAP 1 were similar between groups during rest, and children with ASD initially had fewer occurrences of CAP 1 during the first two task runs but then showed more occurrences of CAP 1 in the last two task runs compared with TD children. Post-hoc *t*-tests were conducted on each run, revealing a significant difference between diagnostic groups within task run 4 ($p = 0.021$). No other run comparisons were significant (p's > 0.05), (**Figure S3**). A post-hoc

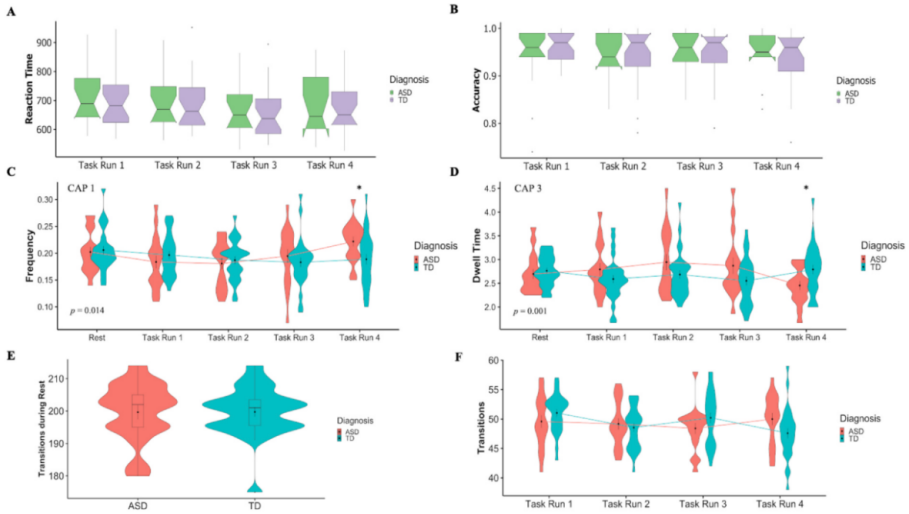

Fig. 3. Behavioral data and CAP frequency, dwell time, and transitions during rest and task states. A) Box plot of RT in children with ASD and TD children. There were no significant differences between groups in RT (p's $>$ 0.05). B) Box plot of Accuracy in children with ASD and TD children. There were no significant differences between groups in accuracy (p's $>$ 0.05). C) Frequency of occurrence of CAP 1, characterized by co-activation of L-FPN nodes, was greater in Task Run 4 for children with ASD than TD children. D) Dwell time of CAP 3, characterized by co-activation of M-CIN and L-FPN nodes, was shorter in Task Run 4 for children with ASD than TD children. E) Transitions between CAPs during rest were not significantly different between groups. F) Transitions between CAPs during task performance were not significantly different between groups when high motion subjects were removed. Results from mixed model ANOVA are shown inside graphs with significant interactions (C and D). * = p $<$ 0.05 from post-hoc t-test.

regression model comparing frequency of occurrences between ASD and TD for task run 4 while controlling for head motion during task run 4 was significant (p = 0.035). Thus, head motion did not influence the group differences observed in task run 4. A post-hoc mixed model ANOVA was conducted excluding four high motion subjects, and a significant linear interaction was still observed, [F(1,38) = 6.526, p = 0.015].

There was a significant cubic main effect of Condition for the frequency of occurrences of CAP 3, [F(1, 42) = 6.25, p = 0.016], however there was no significant main effect of Diagnosis [F(1, 42) = 0.001, p = 0.976]. There was a significant quadratic main effect of Condition for the frequency of occurrences of CAP 5, [F(1, 42) = 6.346, p = 0.016], however there was no significant main effect of Diagnosis [F(1, 42) = 0.324, p = 0.572]. There were no significant main effects or interactions for CAP 2 and CAP 4 (p's $>$ 0.05).

Additionally, there were no significant main effects or interactions for the 2 Diagnosis (ASD, TD) × 2 Condition (rest run, task all) mixed model ANOVAs for CAPs 1–5 (p's $>$ 0.05).

4.4. CAP dwell time

A mixed model ANOVA revealed a significant quadratic main effect of Condition for DT of CAP 1, [F(1,42) = 22.316, p $<$ 0.001], however there was no significant main effect of Diagnosis [F(1, 42) = 0.574, p = 0.453]. There was a significant quadratic main effect of Condition for the DT of CAP 2, [F(1, 42) = 11.368, p = 0.002], however there was no significant main effect of Diagnosis [F(1, 42) = 0.875, p = 0.355]. There was a significant quadratic interaction for DT of CAP 3, where the M-CIN is coupled with the L-FPN [F(1, 42) = 12.785, p = 0.001] (Fig. 3**D**). These results demonstrate that the DT of CAP 3 was similar between groups during rest, and children with ASD initially spend more time in CAP 3 for task runs 1–3, then spend less time in CAP 3 for task run 4 compared with TD children. Post-hoc t-tests were conducted on each condition to compare diagnostic groups and revealed a significant difference in task run 4 (p = 0.034) (**Figure S4**). No other condition comparison was significant (p's $>$ 0.05). A post-hoc regression model comparing DT between ASD and TD for task run 4 while controlling for head motion was significant (p = 0.029). A post-hoc mixed model ANOVA was conducted excluding four high motion subjects, and a significant quadratic interaction was still observed [F(1,38) = 8.625, p = 0.006]. There were no significant main effects or interactions for CAP 4 and CAP 5 (p's $>$ 0.05).

2 Diagnosis (ASD, TD) × 2 Condition (rest run, task all) mixed model ANOVAs for CAPs 1–5 revealed main effects of Condition for CAP 1, [F(1, 42) = 22.366, p = $<$ 0.001], CAP 2, [F(1, 42) = 29.003, p = $<$ 0.001], and CAP 4, [F(1, 42) = 7.745, p = 0.008] and CAP 5, [F(1, 42) = 9.387, p = 0.004]. CAP 3 did not exhibit a significant main effect (p = 0.389). There were no main effects of Diagnosis for any of the CAPs (p's $>$ 0.05).

4.5. CAP transitions

A mixed model ANOVA revealed a significant cubic interaction for the number of transitions [F(1, 42) = 4.124, p = 0.049], indicating TD children have more transitions in task run 1, but have fewer in task run 2, then more in task run 3 and again fewer in task run 4 compared to children with ASD (Fig. 3**E**). However, there was no significant main effect of Diagnosis [F(1, 42) = 0.008, p = 0.930]. A post-hoc mixed model ANOVA using a low motion sample (N = 40) revealed the interaction was no longer significant [F(1, 38) = 3.360, p = 0.075]. We conducted t-tests comparing ASD and TD groups on the resting-state run (Fig. 3**F**), on each task run separately, and on the task runs combined, and found no significant differences between the diagnostic groups (p's $>$ 0.05) (Table S6).

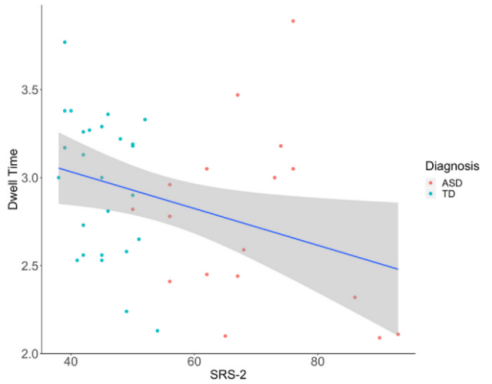

Fig. 4. Pearson correlation between resting-state CAP 5, the M-FPN CAP, and symptom severity indexed by the Social Responsiveness Scale (SRS-2) (Constantino and Gruber, 2012).

4.6. Brain-behavior results

Pearson correlations between SRS-2 and RBS-R total T-scores with each CAP metric revealed a significant correlation between SRS-2 and DT during CAP 5 in the rest condition ($r = -0.337$, $p = 0.027$) (uncorrected) (Fig. 4). No other significant correlations were observed between SRS-2, RBS-R, and any other CAP metrics for CAPSs 1–4 (p's > 0.05). Partial Pearson correlations between SRS-2 and DT while controlling for age revealed that the correlation between SRS-2 and DT during CAP 5 in the rest condition was still significant ($r = -0.341$, $p = 0.027$). We additionally found a significant correlation between SRS-2 and DT during CAP 3 in the task 4 condition ($r = -0.472$, $p = 0.002$) (uncorrected). No other correlations were significant (p's > 0.05).

5. Discussion

Here, we investigate brain dynamics among three neurocognitive networks ubiquitously present in the functional neuroimaging literature (Uddin, 2015). The M-CIN, comprising the anterior insula and anterior cingulate cortices, is thought to enable dynamic switching between the M-FPN (comprising medial prefrontal and posterior cingulate cortices and involved in internally oriented cognition) and the L-FPN (comprising lateral prefrontal and posterior parietal cortices and involved in goal-directed behaviors) (Goulden et al., 2014; Fox et al., 2006). A large literature supports the role of the M-CIN as a mediator of incoming stimuli, guiding appropriate behavioral responses (Uddin, 2015; Goulden et al., 2014; Menon and Uddin, 2010; Uddin and Menon, 2009). Specific regions of the M-CIN such as the ACC have also been shown to have varying modulatory interactions with brain regions during rest and task conditions (Di et al., 2020). While atypical patterns of brain activation and connectivity of these networks have previously been documented in ASD (Uddin et al., 2013), very few studies have examined network configurations as they change between rest and task states (Uddin et al., 2015). Characterizing dynamic changes in the brain lends insight into these alterations, but most studies to date have focused on whole-brain dynamics using sliding window dynamic functional connectivity approaches that have limitations including the use of an arbitrary window length (Allen et al., 2014; Uddin, 2020). Inflexibility among these networks has been shown to underlie core symptoms of ASD (Uddin et al., 2015), yet no previous studies have examined time-varying patterns of co-activation among these networks during intrinsic and evoked states in children with the disorder. Here we used CAP, a method that relies on fewer model assumptions than the sliding window approach, for the first time to characterize brain dynamics among the M-CIN, L-FPN, and M-FPN during a resting-state scan and four runs of an attention task in children with ASD and TD children.

Using CAP, we found evidence for five recurring brain states involving dynamic patterns among M-CIN, M-FPN, and L-FPN nodes during task and resting states. CAP 1 and CAP 2 states exhibited co-activation patterns within L-FPN and within M-CIN, respectively. CAP 3 was characterized by a co-activation among both the M-CIN and L-FPN, a state commonly associated with cognitive task performance (Corbetta and Shulman, 2002) and sometimes referred to as "task-positive" networks (Di and Biswal, n.d.). Together, these results suggest the M-CIN and L-FPN may function independently or simultaneously as needed in the service of task demands or during resting conditions. CAP 4 was characterized by co-activation among all three networks, a brain state which is consistent with evidence from prior studies (Marshall et al., 2020). CAP 5 was a state in which strong M-FPN node co-activation was observed. M-FPN, typically referred to as the "task-negative" network, recently has been revealed to play a role in specific task conditions (Krieger-Redwood et al., 2016; Mars et al., 2012; Spreng et al., 2014; Vatansever et al., 2015) potentially during minimally demanding cognitive tasks (Vatansever et al., 2015). Taken together, these five CAPs display patterns that are consistent with the role of the M-CIN in mediating both the L-FPN and M-FPN, and suggests dynamic interactions among the networks during both task and resting-states (Goulden et al., 2014).

For CAPs 1 and 3, distinct group differences were identified in dynamic metrics of frequency of occurrences and dwell time. Interestingly, in the last two task runs children with ASD exhibited more frequent occurrences of CAP 1 and spent less time in CAP 3 during task run 4 compared to TD children. Greater frequency of occurrences of the L-FPN and less dwell time of simultaneous M-CIN and L-FPN co-activation during the last task runs in children with ASD suggests they rely more on the L-FPN when needing to exert greater effort to reach the same behavioral outcomes as children with TD. Previous work has shown that high functioning individuals with ASD may perform a task at an above-average level, as shown here, but may require more detailed-focused processing (Happé and Frith, 2006). This type of behavior has been previously associated with overly stable brain dynamics in adults with ASD (Watanabe and Rees, 2017). The lack of behavioral differences found in this study suggests that children with ASD performed at a high level across all four task runs, and this behavior is supported by altered changes in dynamic fluctuations across the networks from the early to the later phases of the task.

The observed disruption in the coordination of the L-FPN and M-CIN nodes, evident in the last task runs, is consistent with emerging evidence using dynamic methods of disruptions in between-network connectivity in children with ASD (de Lacy et al., 2017). Similarly, coordination among all three neurocognitive networks has been previously shown to dynamically occur less frequently during intrinsic states in children with ASD compared to TD children, indicating a reduced co-activation of the M-CIN with nodes of the M-FPN and L-FPN (Marshall et al., 2020). Although our finding is across evoked states, our results are consistent with the prior study suggesting the nodes of the M-CIN have reduced coordination with the nodes of the L-FPN, primarily during the last task runs. A disruption in the coordination of networks may underlie cognitive and behavioral inflexibility seen in children with ASD (Uddin et al., 2015; Barttfeld et al., 2012). Additionally, dynamics assessed during evoked states may reveal unique network re-configurations under varying task demands, and differential employment of cognitive effort in ASD (Cheng et al., 2018). Dynamic analyses can track differential brain responses in ASD across changing task demands over time. The current findings imply that CAP metrics computed across multiple task runs can be more revealing of neural profiles in autism than differences between task and rest contexts, which have been the focus of previous similar works (Uddin et al.,

2015). However, further studies are needed to support this interpretation. There were no group differences in transitions observed, primarily after removing subjects with excessive motion. This is contradictory to a growing dynamic functional connectivity literature suggesting that transitions between states are altered in ASD (Uddin, 2020; de Lacy et al., 2017; Watanabe and Rees, 2017). Our results may be influenced by our limited sample size and the relatively short duration of our task and rest scans; future studies including larger sample sizes and longer scan times are needed to further explore this issue. Additionally, our lack of significant findings for transitions may be attributed to our analysis of three neurocognitive networks rather than whole-brain analyses, as conducted in previous studies (Uddin, 2020; de Lacy et al., 2017; Watanabe and Rees, 2017).

The only significant relationship between the brain dynamic metrics and behavior was identified in the M-FPN. Greater dwell time in the M-FPN was associated with better social abilities as indexed by the SRS-2. This is in line with previous research linking the M-FPN with social ability in ASD (He et al., 2018; Padmanabhan et al., 2017) and in the general population (Mars et al., 2012; Li et al., 2014). The dynamic analyses presented here provide new insight into the relationship between the M-FPN and social behaviors. Our findings indicate that children who exhibit greater M-FPN engagement during resting states are those who are higher functioning in the social domain. This is in line with a large literature implicating the M-FPN in thinking about others (Uddin et al., 2007).

During task performance, we observed no behavioral differences in reaction time and accuracy between children with ASD and TD children. In both groups, accuracy remained elevated across the four task runs (Accuracy $>$ 90%), indicating there was not an underlying behavioral change across the four task runs to account for the brain dynamic changes we observed. As previously reported, high accuracy and a lack of behavioral differences between groups indicate that this task was relatively easy for both children with ASD and TD children to complete (Dirks et al., 2020). Nevertheless, children with ASD recruited brain regions involved in executive control to a greater extent than TD children by task run 4, indicating that they exerted greater cognitive effort to reach the same level of performance as TD children. This suggests that children with ASD may neurally compensate to reach the same level of behavioral performance as TD children across the duration of a task (Livingston and Happé, 2017).

5.1. Limitations

There are a few limitations important to note in the present study. Our sample size was limited as we maintained strict requirements including a visual Quality Control inspection, a full 10 min of resting-state fMRI, and completion of all four task runs. While these requirements increased power related to our study aims, they reduced our sample size, as several children were excluded due to our strict criteria. Future work with larger sample sizes is needed to confirm and extend the results presented here. Lastly, other large-scale networks including the dorsal attention network (DAN)/dorsal frontoparietal network (D-FPN) have also been shown to interact with the three networks investigated here, depending on context (Dixon et al., 2018). Future studies should further expand on the current work by investigating the D-FPN and its dynamic relationship to the L-FPN, M-CIN and M-FPN in children with ASD.

5.2. Conclusions

This study investigated brain dynamic metrics concurrently during rest and an attention task in children with ASD and TD children. Group differences between children with ASD and TD children were evident in brain states consisting of the L-FPN and M-CIN specifically during the fourth task run, suggesting atypical between-network coordination in children with ASD during prolonged periods of task engagement. Atypical between-network coordination may underlie neural compensation in children with ASD, enabling comparable behavioral performance as TD children. Finally, greater M-FPN dwell time was associated with stronger social abilities, indicating that the dynamics of this network may be important in our understanding of social dysfunction in both ASD and the general population.

CRediT authorship contribution statement

Lauren Kupis: Formal analysis, Writing - original draft, Writing - review & editing, Visualization. **Celia Romero:** Data curation, Writing - review & editing. **Bryce Dirks:** Data curation, Writing - review & editing. **Stephanie Hoang:** Visualization, Writing - review & editing. **Meaghan V. Parladé:** Resources, Supervision, Writing - review & editing. **Amy L. Beaumont:** Resources, Supervision, Writing - review & editing. **Sandra M. Cardona:** Resources, Supervision, Writing - review & editing. **Michael Alessandri:** Resources, Supervision, Writing - review & editing. **Catie Chang:** Methodology, Supervision, Writing - review & editing. **Jason S. Nomi:** Data curation, Supervision, Writing - original draft, Writing - review & editing. **Lucina Q. Uddin:** Conceptualization, Supervision, Writing - original draft, Writing - review & editing, Funding acquisition.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Presentation information

This study was presented as an abstract at the University of Miami Graduate and Postgraduate Research Symposium; March 5, 2020; Coral Gables, Florida and presented virtually at the Organization for Human Brain Mapping; June 24–July 2, 2020.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.nicl.2020.102396.

References

Allen, E.A., Damaraju, E., Plis, S.M., Erhardt, E.B., Eichele, T., Calhoun, V.D., 2014. Tracking whole-brain connectivity dynamics in the resting state. Cereb. Cortex. 24 (3), 663–676.

American Psychiatric Association, 2013. Diagnostic and Statistical Manual of Mental Disorders (DSM-5®). American Psychiatric Pub.

Barttfeld, P., Wicker, B., Cukier, S., et al., 2012. State-dependent changes of connectivity patterns and functional brain network topology in autism spectrum disorder. Neuropsychologia. 50 (14), 3653–3662.

Bassett, D.S., Wymbs, N.F., Porter, M.A., Mucha, P.J., Carlson, J.M., Grafton, S.T., 2011. Dynamic reconfiguration of human brain networks during learning. Proc. Natl. Acad. Sci. U S A. 108 (18), 7641–7646.

Bolt, T., Nomi, J.S., Rubinov, M., Uddin, L.Q., 2017. Correspondence between evoked and intrinsic functional brain network configurations. Hum. Brain Mapp. 38 (4), 1992–2007.

Braun, U., Schäfer, A., Walter, H., et al., 2015. Dynamic reconfiguration of frontal brain networks during executive cognition in humans. Proc. Natl. Acad. Sci. U S A. 112 (37), 11678–11683.

Britton, J.C., Rauch, S.L., Rosso, I.M., et al., 2010. Cognitive inflexibility and frontal-cortical activation in pediatric obsessive-compulsive disorder. J. Am. Acad. Child Adolesc. Psychiatry 49 (9), 944–953. https://doi.org/10.1016/j.jaac.2010.05.006.
Buckley, A.W., Scott, R., Tyler, A., et al., 2015. State-dependent differences in functional connectivity in young children with autism spectrum disorder. EBioMedicine. 2 (12), 1905–1915.
Calhoun, V.D., Miller, R., Pearlson, G., Adalı, T., 2014. The chronnectome: time-varying connectivity networks as the next frontier in fMRI data discovery. Neuron. 84 (2), 262–274.
Casey, B.J., Davidson, M.C., Hara, Y., et al., 2004. Early development of subcortical regions involved in non-cued attention switching. Devel. Sci. 7 (5), 534–542. https://doi.org/10.1111/j.1467-7687.2004.00377.x.
Chang, C., Glover, G.H., 2010. Time–frequency dynamics of resting-state brain connectivity measured with fMRI. Neuroimage. 50 (1), 81–98.
Chen, H., Nomi, J.S., Uddin, L.Q., Duan, X., Chen, H., 2017. Intrinsic functional connectivity variance and state-specific under-connectivity in autism. Hum. Brain Mapp. 38 (11), 5740–5755.
Chen, H., Wang, J., Uddin, L.Q., et al., 2018. Aberrant functional connectivity of neural circuits associated with social and sensorimotor deficits in young children with autism spectrum disorder. Autism Res. 11 (12), 1643–1652.
Cheng, L., Zhu, Y., Sun, J., et al., 2018. Principal states of dynamic functional connectivity reveal the link between resting-state and task-state brain: an fMRI study. Int. J. Neural Syst. 28 (07), 1850002. https://doi.org/10.1142/s0129065718500028.
Ciric, R., Nomi, J.S., Uddin, L.Q., Satpute, A.B., 2017. Contextual connectivity: a framework for understanding the intrinsic dynamic architecture of large-scale functional brain networks. Sci. Rep. 7 (1), 6537.
Cole, M.W., Bassett, D.S., Power, J.D., Braver, T.S., Petersen, S.E., 2014. Intrinsic and task-evoked network architectures of the human brain. Neuron 83 (1), 238–251.
Constantino, J.N., Gruber, C.P., 2012. Social Responsiveness Scale Second Edition (SRS-2): Manual.
Corbetta, M., Shulman, G.L., 2002. Control of goal-directed and stimulus-driven attention in the brain. Nat. Rev. Neurosci. 3 (3), 201–215.
Dajani, D.R., Uddin, L.Q., 2015. Demystifying cognitive flexibility: Implications for clinical and developmental neuroscience. Trends Neurosci. 38 (9), 571–578.
Damaraju, E., Allen, E.A., Belger, A., et al., 2014. Dynamic functional connectivity analysis reveals transient states of dysconnectivity in schizophrenia. Neuroimage Clin. 5, 298–308.
de Lacy, N., Doherty, D., King, B.H., Rachakonda, S., Calhoun, V.D., 2017. Disruption to control network function correlates with altered dynamic connectivity in the wider autism spectrum. Neuroimage Clin. 15, 513–524.
Denkova, E., Nomi, J.S., Uddin, L.Q., 2019. Dynamic brain network configurations during rest and an attention task with frequent occurrence of mind wandering. Hum. Brain Mapp. https://doi.org/10.1002/hbm.24721.
Di, X., Biswal, B.B., n.d. Modulatory interactions between the default mode network and task positive networks in resting-state. doi: 10.7287/peerj.preprints.124.
Di Martino, A., Kelly, C., Grzadzinski, R., et al., 2011. Aberrant striatal functional connectivity in children with autism. Biol. Psychiatry. 69 (9), 847–856.
Di, X., Zhang, H., Biswal, B.B., 2020. Anterior cingulate cortex differently modulates frontoparietal functional connectivity between resting-state and working memory tasks. Hum. Brain Mapp. 41 (7), 1797–1805.
Dirks, B., Romero, C., Voorhies, W., Kupis, L., Nomi, J.S., Dajani, D.R., Odriozola, P., Burrows, C.A., Beaumont, A.L., Cardona, S.M., Parlade, M.V., Alessandri, M., Britton, J.C., Uddin, L.Q., 2020. Neural responses to a putative set-shifting task in children with autism spectrum disorder. Autism Res.
Dixon, M.L., De La Vega, A., Mills, C., et al., 2018. Heterogeneity within the frontoparietal control network and its relationship to the default and dorsal attention networks. Proc. Natl. Acad. Sci. U S A. 115 (7), E1598–E1607.
Falahpour, M., Thompson, W.K., Abbott, A.E., et al., 2016. Underconnected, but not broken? Dynamic functional connectivity MRI shows underconnectivity in Autism is linked to increased intra-individual variability across time. Brain Connect. 6 (5), 403–414.
Fishman, I., Linke, A.C., Hau, J., Carper, R.A., Müller, R.-A., 2018. Atypical functional connectivity of amygdala related to reduced symptom severity in children with Autism. J. Am. Acad. Child Adolesc. Psychiatry. 57 (10), 764–774.e3.
Fox, M.D., Corbetta, M., Snyder, A.Z., Vincent, J.L., Raichle, M.E., 2006. Spontaneous neuronal activity distinguishes human dorsal and ventral attention systems. Proc. Natl. Acad. Sci. U S A. 103 (26), 10046–10051.
Friston, K.J., Williams, S., Howard, R., Frackowiak, R.S.J., Turner, R., 1996. Movement-related effects in fMRI time-series. Magn. Reson. Med. 35 (3), 346–355.
Goulden, N., Khusnulina, A., Davis, N.J., et al., 2014. The salience network is responsible for switching between the default mode network and the central executive network: replication from DCM. Neuroimage. 99, 180–190.
Griffanti, L., Douaud, G., Bijsterbosch, J., et al., 2017. Hand classification of fMRI ICA noise components. NeuroImage. 154, 188–205. https://doi.org/10.1016/j.neuroimage.2016.12.036.
Happé, F., Frith, U., 2006. The weak coherence account: detail-focused cognitive style in autism spectrum disorders. J. Autism Dev. Disord. 36 (1), 5–25. https://doi.org/10.1007/s10803-005-0039-0.
Harlalka, V., Bapi, R.S., Vinod, P.K., Roy, D., 2019. Atypical flexibility in dynamic functional connectivity quantifies the severity in autism spectrum disorder. Front. Hum. Neurosci. 13, 6.
He, C., Chen, Y., Jian, T., et al., 2018. Dynamic functional connectivity analysis reveals decreased variability of the default-mode network in developing autistic brain. Autism Res. 11 (11), 1479–1493.
Hutchison, R.M., Morton, J.B., 2015. Tracking the brain's functional coupling dynamics over development. J. Neurosci. 35 (17), 6849–6859.
Jenkinson, M., Beckmann, C.F., Behrens, T.E.J., Woolrich, M.W., Smith, S.M., 2012. FSL. NeuroImage. 62 (2), 782–790. https://doi.org/10.1016/j.neuroimage.2011.09.015.
Jia, H., Hu, X., Deshpande, G., 2014. Behavioral relevance of the dynamics of the functional brain connectome. Brain Connect. 4 (9), 741–759.
Kana, R.K., Uddin, L.Q., Kenet, T., Chugani, D., Müller, R.-A., 2014. Brain connectivity in autism. Front. Hum. Neurosci. 8, 349.
Keown, C.L., Shih, P., Nair, A., Peterson, N., Mulvey, M.E., Müller, R.-A., 2013. Local functional overconnectivity in posterior brain regions is associated with symptom severity in autism spectrum disorders. Cell Rep. 5 (3), 567–572.
Krieger-Redwood, K., Jefferies, E., Karapanagiotidis, T., et al., 2016. Down but not out in posterior cingulate cortex: deactivation yet functional coupling with prefrontal cortex during demanding semantic cognition. NeuroImage. 141, 366–377. https://doi.org/10.1016/j.neuroimage.2016.07.060.
Lam, K.S., Aman, M.G., 2007. The repetitive behavior scale-revised: independent validation in individuals with autism spectrum disorders. J. Autism Dev. Disord. 37 (5), 855–866.
Li, W., Mai, X., Liu, C., 2014. The default mode network and social understanding of others: what do brain connectivity studies tell us. Front. Hum. Neurosci. 8. https://doi.org/10.3389/fnhum.2014.00074.
Liu, X., Chang, C., Duyn, J.H., 2013. Decomposition of spontaneous brain activity into distinct fMRI co-activation patterns. Front. Syst. Neurosci. 7. https://doi.org/10.3389/fnsys.2013.00101.
Liu, X., Duyn, J.H., 2013. Time-varying functional network information extracted from brief instances of spontaneous brain activity. Proc. Natl. Acad. Sci. U S A. 110 (11), 4392–4397.
Liu, X., Zhang, N., Chang, C., Duyn, J.H., 2018. Co-activation patterns in resting-state fMRI signals. Neuroimage. 180 (Pt B), 485–494.
Livingston, L.A., Happé, F., 2017. Conceptualising compensation in neurodevelopmental disorders: reflections from autism spectrum disorder. Neurosci. Biobehav. Rev. 80, 729–742. https://doi.org/10.1016/j.neubiorev.2017.06.005.
Lord, C., Rutter, M., DiLavore, P., Risi, S., Gotham, K., Bishop, S., 2012. Autism diagnostic observation schedule–2nd edition (ADOS-2). Western Psychological Corporation, Los Angeles, CA.
Lurie, D.J., Kessler, D., Bassett, D.S., et al., 2020. Questions and controversies in the study of time-varying functional connectivity in resting fMRI. Netw. Neurosci. 4 (1), 30–69.
Mars, R.B., Neubert, F.-X., Noonan, M.P., Sallet, J., Toni, I., Rushworth, M.F.S., 2012. On the relationship between the "default mode network" and the "social brain". Front. Hum. Neurosci. 6. https://doi.org/10.3389/fnhum.2012.00189.
Marshall, E., Nomi, J.S., Dirks, B., Romero, C., Kupis, L., Chang, C., Uddin, L.Q., 2020. **Co-activation pattern analysis reveals altered salience network dynamics in children with autism spectrum disorder Nomi, J. S., Dirks, B., Romero, C., Kupis, L., Chang, C., & Uddin, L. Q.**. Network Neurosci.
Mash, L.E., Linke, A.C., Olson, L.A., Fishman, I., Liu, T.T., Müller, R.-A., 2019. Transient states of network connectivity are atypical in autism: a dynamic functional connectivity study. Hum. Brain Mapp. 40 (8), 2377–2389.
Menon, V., Uddin, L.Q., 2010. Saliency, switching, attention and control: a network model of insula function. Brain Struct. Funct. 214 (5–6), 655–667.
Müller, R.-A., Fishman, I., 2018. Brain connectivity and neuroimaging of social networks in Autism. Trends Cogn. Sci. 22 (12), 1103–1116.
Padmanabhan, A., Lynch, C.J., Schaer, M., Menon, V., 2017. The default mode network in Autism. Biol. Psychiatry: Cognitive Neurosci. Neuroimag. 2 (6), 476–486. https://doi.org/10.1016/j.bpsc.2017.04.004.
Parker, D.B., Razlighi, Q.R., 2019. The benefit of slice timing correction in common fMRI preprocessing pipelines. Front. Neurosci. 13, 821.
Power, J.D., Mitra, A., Laumann, T.O., Snyder, A.Z., Schlaggar, B.L., Petersen, S.E., 2014. Methods to detect, characterize, and remove motion artifact in resting state fMRI. Neuroimage. 84, 320–341.
Preti, M.G., Bolton, T.A., Van De Ville, D., 2017. The dynamic functional connectome: state-of-the-art and perspectives. Neuroimage. 160, 41–54.
Rane, P., Cochran, D., Hodge, S.M., Haselgrove, C., Kennedy, D.N., Frazier, J.A., 2015. Connectivity in Autism: a review of MRI connectivity studies. Harv. Rev. Psychiatry. 23 (4), 223–244.
Rashid, B., Blanken, L.M.E., Muetzel, R.L., et al., 2018. Connectivity dynamics in typical development and its relationship to autistic traits and autism spectrum disorder. Hum. Brain Mapp. 39 (8), 3127–3142.
Spreng, R.N., DuPre, E., Selarka, D., et al., 2014. Goal-congruent default network activity facilitates cognitive control. J. Neurosci. 34 (42), 14108–14114.
Supekar, K., Uddin, L.Q., Khouzam, A., et al., 2013. Brain hyperconnectivity in children with autism and its links to social deficits. Cell Rep. 5 (3), 738–747.
Uddin, L.Q., 2015. Salience processing and insular cortical function and dysfunction. Nat. Rev. Neurosci. 16 (1), 55–61.
Uddin, L.Q., 2020. Brain mechanisms supporting flexible cognition and behavior in adolescents with autism spectrum disorder. Biol. Psychiatry. https://doi.org/10.1016/j.biopsych.2020.05.010.
Uddin, L.Q., Karlsgodt, K.H., 2018. Future directions for examination of brain networks in neurodevelopmental disorders. J. Clin. Child Adolesc. Psychol. 47 (3), 483–497.
Uddin, L.Q., Iacoboni, M., Lange, C., Keenan, J.P., 2007. The self and social cognition: the role of cortical midline structures and mirror neurons. Trends Cognitive Sci. 11 (4), 153–157. https://doi.org/10.1016/j.tics.2007.01.001.
Uddin, L.Q., Menon, V., 2009. The anterior insula in autism: under-connected and under-examined. Neurosci. Biobehav. Rev. 33 (8), 1198–1203.
Uddin, L.Q., Supekar, K.S., Ryali, S., Menon, V., 2011. Dynamic reconfiguration of structural and functional connectivity across core neurocognitive brain networks with development. J. Neurosci. 31 (50), 18578–18589. https://doi.org/10.1523/jneurosci.4465-11.2011.
Uddin, L.Q., Supekar, K., Lynch, C.J., et al., 2013. Salience network-based classification

and prediction of symptom severity in children with Autism. JAMA Psychiatry. 70 (8), 869. https://doi.org/10.1001/jamapsychiatry.2013.104.
Uddin, L.Q., Supekar, K., Lynch, C.J., et al., 2015. Brain state differentiation and behavioral inflexibility in Autism. Cereb. Cortex. 25 (12), 4740–4747.
Uddin, L.Q., Yeo, B.T.T., Spreng, R.N., 2019. Towards a universal taxonomy of macroscale functional human brain networks. Brain Topogr. 32 (6), 926–942.
Vatansever, D., Menon, D.K., Manktelow, A.E., Sahakian, B.J., Stamatakis, E.A., 2015. Default mode dynamics for global functional integration. J. Neurosci. 35 (46), 15254–15262. https://doi.org/10.1523/jneurosci.2135-15.2015.
Vissers, M.E., Cohen, X.M., Geurts, H.M., 2012. Brain connectivity and high functioning autism: a promising path of research that needs refined models, methodological convergence, and stronger behavioral links. Neurosci. Biobehav. Rev. 36 (1), 604–625.
Watanabe, T., Rees, G., 2017. Brain network dynamics in high-functioning individuals with autism. Nat. Commun. 8, 16048.
Wechsler, D., 2011. Wechsler Abbreviated Scale of Intelligence-Second Edition. NCS Pearson, San Antonio, TX.
White, T., Calhoun, V.D., 2019. Dissecting static and dynamic functional connectivity: example from the autism spectrum. J. Exp. Neurosci. 13.
Yan, C.-G., Wang, X.-D., Zuo, X.-N., Zang, Y.-F., 2016. DPABI: Data Processing & Analysis for (Resting-State) Brain Imaging. Neuroinformatics. 14 (3), 339–351.
Yao, Z., Hu, B., Xie, Y., et al., 2016. Resting-state time-varying analysis reveals aberrant variations of functional connectivity in Autism. Front. Hum. Neurosci. 10, 463.
You, X., Norr, M., Murphy, E., et al., 2013. Atypical modulation of distant functional connectivity by cognitive state in children with Autism Spectrum Disorders. Front. Hum. Neurosci. 7. https://doi.org/10.3389/fnhum.2013.00482.

CHAPTER 4:

Brain Dynamics in Toddlers with and without Autism Spectrum Disorder

ABSTRACT

Autism spectrum disorder (ASD) affects one in 36 children. Early diagnosis is critical for optimizing outcomes, yet children are not typically diagnosed until 4 years of age. In concert with early behavioral signs, early neural markers could identify toddlers at risk of developing ASD to aid earlier diagnosis and targeted interventions. Neuroimaging studies have primarily examined structural brain alterations in toddlers at high risk of developing ASD. While innovative dynamic functional magnetic resonance imaging (fMRI) methods reveal candidate brain networks of dysfunction in older children with ASD (7-12 years of age), little work has been done to examine brain network dynamics in toddlers with ASD. The goal of this project is to identify early functional brain biomarkers of ASD and their relationships with early flexible behaviors (e.g., repetitive and adaptive behaviors). Using data from 48 ASD and 27 non-ASD toddlers, we examined brain network dynamics in the whole brain and among the salience network (SN), default mode (DMN), and central executive networks (CEN). Early brain network dynamics were similar across all toddlers, however, ASD toddlers exhibited altered brain dynamics in a state consisting of SN, DMN, and CEN co-activation. Across both diagnostic groups, there were relationships between early brain dynamics and real-world measures of cognitive flexibility. At the whole brain level, greater dwell times and frequencies of states with core neural networks and visual and subcortical regions were associated with greater flexible

behaviors. While investigating interactions among the core neural networks (SN, DMN, and CEN), a greater dwell time of a state with DMN co-activation was associated with poorer RRBs and adaptive behaviors; conversely, greater frequency of a state with a greater SN and CEN co-activation and DMN de-activation was associated with better RRB and adaptive behavior outcomes. Overall, this is one of the first studies to investigate brain network dynamics in typical and atypical toddlers.

INTRODUCTION

Autism Spectrum Disorder (ASD) is a complex neurodevelopmental condition characterized by social communication deficits, and restricted and repetitive behavior, interests, or activities (American Psychiatric Association 2015). The prevalence rate of ASD in the United States is rising, with one in 36 children diagnosed each year (Maenner et al. 2023), and this rate continues to rise. Despite the increasing prevalence, the median age of ASD diagnosis remains at four years, leading to a delay in treatment initiation during crucial periods of brain development (Tierney and Nelson 2009b). Early diagnosis and intervention significantly improve cognitive, language, adaptive behavior, and overall quality of life for individuals with ASD (Elder et al. 2017; Richler et al. 2010b).

Currently, ASD diagnosis relies primarily on behavioral symptoms, including social deficits and restricted and repetitive behaviors (RRBs) (American Psychiatric Association 2015). RRBs present as restricted interests, insistence on sameness, repetitive speech (e.g., echolalia), and difficulties with behavioral transitions. RRBs interfere with learning, social development, daily activities, and family functioning (American Psychiatric Association 2015; Richler et al.

2010b), causing impairments in development and increased stress for caretakers (Bishop et al. 2007). Moreover, RRB severity is linked to later developing comorbid psychiatric conditions such as anxiety and depression ("Relations among Restricted and Repetitive Behaviors, Anxiety and Sensory Features in Children with Autism Spectrum Disorders" 2014), highlighting the need for early identification and targeted intervention strategies. Early behavioral signs of ASD have been suggested (Barbaro and Dissanayake 2009) but these signs are typically observed in the social domain (Ozonoff et al. 2010) and are not always reliable enough to establish diagnosis (Pierce et al. 2019; Jarquin et al. 2011). Therefore, it is imperative to identify early biomarkers, particularly those based on brain functioning, to facilitate early intervention and improve outcomes for individuals with ASD.

Neuroimaging techniques, such as magnetic resonance imaging (MRI), provide valuable insights into the neurobiology of ASD and potential brain biomarkers underlying overt behavioral symptoms in ASD. In particular, functional MRI (fMRI) methods, including resting-state functional MRI (rsfMRI), have been used to reveal alterations in brain regions associated with children and adults with ASD compared with neurotypical peers (Lucina Q. Uddin, Supekar, and Menon 2013; Neufeld et al. 2017; K. Supekar et al. 2013). RsfMRI captures the intrinsic functional architecture of the brain based on the spontaneous low frequency fluctuations of the BOLD signal (Biswal et al. 1995; Lee, Smyser, and Shimony 2013). RsfMRI and sleep fMRI paradigms have revolutionized clinical research (Yang, Dong, and Lei 2021; Pierce 2011) by providing a way to study populations that would not otherwise be able to be studied under typical task conditions or without anesthesia. RsfMRI studies also reveal intrinsic connectivity

patterns that correlate with behavior and cognition in individuals with and without ASD ("Indices of Repetitive Behaviour Are Correlated with Patterns of Intrinsic Functional Connectivity in Youth with Autism Spectrum Disorder" 2018; Easson, Fatima, and McIntosh 2019; K. Supekar et al. 2013). The promise of rsfMRI and sleep-based paradigms provide novel ways to investigate early brain development and understand the neural development underlying autism.

Emerging evidence from sleep fMRI studies of toddlers suggests that alterations in brain connectivity may serve as early markers for ASD. Structural connectivity, assessed through diffusion tensor imaging and MRI scans, has revealed aberrant white matter tract microstructure and network inefficiencies in high-risk infants as young as 6 weeks to 6 months, which predict later language development and autism symptoms (Wolff et al. 2012; Elison et al. 2014). Furthermore, functional connectivity studies using rsfMRI have identified potential risk markers in infants with a family history of ASD as young as 6 months of age (Hazlett et al. 2017; I. Molnar-Szakacs, Kupis, and Uddin 2021). Early brain connectivity differences in autism also reveal promising early biomarkers underlying emerging behavior. However, most studies have focused on social communication skills (Hazlett et al. 2017; I. Molnar-Szakacs, Kupis, and Uddin 2021; Lombardo et al. 2015). These findings provide valuable promise of fMRI as a biomarker for later developing ASD and behaviors. However, previous studies have primarily focused on static functional connectivity or structural MRI biomarkers, neglecting the dynamic nature of brain activity in ASD toddlers and the core behavioral deficit of RRBs.

Recent advances in dynamic functional connectivity analysis have demonstrated the temporal variability of brain networks, offering a more nuanced understanding of brain-behavior relationships (Hutchison et al. 2013; Braun et al. 2015). Brain dynamics capture moment-by-moment changes in brain network configurations related to changes in cognition or behavior (Braun et al. 2015; Gu et al. 2020; "Brain Mechanisms Supporting Flexible Cognition and Behavior in Adolescents With Autism Spectrum Disorder" 2021). Dynamic methods such as dynamic functional connectivity (DFC; e.g., sliding window) (Chang and Glover 2010) and co-activation pattern analysis (CAP) (X. Liu and Duyn 2013) are popularly used to quantify brain dynamics (Lurie et al. 2020). Brain connectivity dynamics also help explain differences in clinical populations and behavior (Kupis, Romero, Dirks, Hoang, Parladé, et al. 2020; Kupis, Goodman, Kircher, et al. 2021; Damaraju et al. 2014; Nomi et al. 2017; Matthew Hutchison and Bruce Morton 2015). Previous work has shown brain connectivity dynamics based on sliding window and CAP analyses may help explain cognitive and behavioral inflexibility associated with RRBs across the lifespan (Kupis, Goodman, Kornfeld, et al. 2021) and in 7-12 year old children with ASD (Kupis, Romero, Dirks, Hoang, Parladé, et al. 2020; Kupis, Goodman, Kircher, et al. 2021; Marshall et al. 2020b). Further, children with ASD show atypical brain dynamic patterns during both task performance and resting-states (Kupis, Romero, Dirks, Hoang, Parladé, et al. 2020; Marshall et al. 2020b). Despite the promise of dynamic fMRI methods, there is little work done utilizing resting-state dynamics as biomarkers for early signs of ASD and RRBs in toddlers.

Brain network dynamic methods have also revealed promising biomarkers underlying autism and later developing behavior difficulties. For instance, previous research has demonstrated associations between the severity of RRBs and brain connectivity dynamics among three core neural networks in older children with ASD (“Brain Mechanisms Supporting Flexible Cognition and Behavior in Adolescents With Autism Spectrum Disorder” 2021), notably the salience (SN), default mode (DMN), and central executive networks (CEN). These networks, originally proposed within the influential "triple network model," have been extensively cited in the literature and consistently linked to psychiatric conditions such as autism (Vinod Menon 2011). Previous studies have indicated associations between the severity of RRBs and connectivity among these large-scale brain networks in older children with ASD (Traynor and Hall 2015b; Lucina Q. Uddin et al. 2015b, 2013b). Network dynamics and time-varying interactions of these three large-scale networks are additionally associated with cognitive and behavioral flexibility (Marshall et al. 2020b; Kupis, Romero, Dirks, Hoang, Parladé, et al. 2020; Lucina Q. Uddin et al. 2015b). Therefore, it is critical to investigate the time-varying interactions of the SN, CEN, and DMN in toddlers prior to the diagnosis of ASD and their relationship with early-life RRBs.

Our proposed research aims to utilize novel brain dynamic approaches to identify early biomarkers associated with ASD and RRB outcomes independent of diagnosis. Building upon previous work, which has elucidated brain connectivity dynamics in older children with ASD (“Brain Mechanisms Supporting Flexible Cognition and Behavior in Adolescents With Autism Spectrum Disorder” 2021) using both CAP and dFC methods, we seek to extend these findings

to toddlers, a critical developmental period characterized by rapid brain maturation and plasticity (Gilmore, Santelli, and Gao 2018). By investigating the dynamic interactions among large-scale brain networks, including the SN, CEN, and DMN, we aim to uncover neural substrates underlying RRBs and cognitive inflexibility in toddlers with and without ASD. Both CAPs and dFC methods will be utilized in the current study since both methods are popularly used in children and adult studies of ASD, and both methods differ methodologically and therefore may reveal different aspects of brain dynamics. This study will not only advance our understanding of the neurobiological underpinnings of ASD but also inform the development of targeted interventions aimed at mitigating RRBs and improving long-term outcomes for affected individuals.

METHODS

Participants

Toddlers were recruited through community referral and a population-based screening method in collaboration with pediatricians via the Get SET Early Approach (Pierce et al. 2021). All toddlers participated in clinical assessments, including the Autism Diagnostic Observation Schedule (ADOS) (Lord et al. 2000), Mullen Scales of Early Learning (Mullen 1995) and Vineland Adaptive Behavior Scales (Sparrow 2005). Toddlers who received their initial diagnostic and clinical evaluations at <36 months were invited to return for repeat evaluations until they reached 48 months. Clinical scores at the most recent visit were used to determine the diagnosis group (Table 1). Clinical testing occurred at the University of California, San Diego Autism Center of Excellence.

Clinical scores and fMRI scans were collected from 75 toddlers (48 with ASD, 27 Non-ASD; 52 male, 23 female, 14–55 months old). Resting-state fMRI data were collected from all 75 subjects during natural sleep and most scans were completed before the final diagnosis (<33 mos; mean = 26.56 mos). Participants were considered non-ASD if their diagnosis at the outcome visit was non-ASD and their Mullen Early Learning Composite score fell within 2 standard deviations of the mean score (i.e., > 70). This allowed for the examination of brain dynamics along a continuum of RRB and cognitive abilities from below to above average in non-ASD children as supported by RDOC (Cuthbert 2022) and previously done using this sample (Xiao et al. 2023). Further details of the methods can be found in our previous study (Xiao et al. 2023).

Table 1

	ASD (N = 48) M (SD)	Non-ASD (N = 27) M (SD)	*p*-value
Age at scan	29 mos (16-55 mos)	25.52 (14-46 mos)	.12
Sex	37 M 11 F	15 M 12 F	.05
Ethnicity	22 Hispanic or Latino	8 Hispanic or Latino	.17
Race	30 White; 5 Asian; 1 Black; 5 more than 1 race; 7 N/A	22 White; 1 American Indian; 1 Black; 2 More than 1 race; 1 unknown	.17
ADOS-RRB	4.83 (2.59)	1.67 (2.61)	.22

Vineland Adaptive Behavior	82.9 (12.47)	97.04 (12.33)	.29
Mullen	68.44 (26.29)	97.15 (25.56)	.03
Mean FD	.08 (.05)	.09 (.05)	.24

Sleep fMRI

Toddler scans were conducted during natural sleep, a method that has been previously used to study both toddlers with and without ASD (Pierce 2011). To ensure optimal conditions for sleep fMRI, parents were instructed to eliminate naps from their child's routine on the day of the scan and to keep the child awake at home until arriving at the scanning facility, approximately 1 hour past their usual bedtime. Toddlers were placed on the scanner bed approximately 20 minutes after the onset of sleep to standardize sleep stages during scanning. Previous studies have demonstrated that successful sleep fMRI acquisition predominantly occurs during non-REM stage 3 sleep (Mitra et al. 2017), thereby promoting uniformity in sleep state among scans that were successfully obtained. Studies have also explored brain dynamics during sleep and reveal that brain dynamics and brain states can be characterized during sleep (Rué-Queralt et al. 2021; "Connectivity Dynamics from Wakefulness to Sleep" 2020; Stevner et al. 2019) and stage 3 sleep state dynamics is distinguishable from head motion ("Connectivity Dynamics from Wakefulness to Sleep" 2020).

Behavior Measures

ADOS RRB

All participants underwent the ADOS, a semi-structured observational tool used by trained clinicians to score autism symptoms in two domains: social affective and restrictive, repetitive, behavior (RRB) (Lord et al. 2012). The RRB subdomain scores unstructured instances of restricted interests and repetitive, stereotyped behaviors. We hypothesized that early brain dynamics would correlate with ADOS RRBs across all participants.

Vineland Adaptive Behavior

The Vineland Adaptive Behavior Scales (VABS) is a parent-report measure of adaptive behavioral skills in children (Sparrow, Cicchetti, and Balla 2012). The VABS assesses adaptive behavior in domains of communication, daily living skills, and socialization. The subdomains are combined to form a composite score of adaptive functioning. The adaptive behavior composite score in the VABS has been found to negatively correlate with RRBs such that lower adaptive functioning is associated with more severe RRBs (Cuccaro et al. 2003). The VABS composite score will be used as a dimensional index of behavioral flexibility.

fMRI Data Acquisition

All fMRI data were acquired using a 3 T GE scanner at the University of California, San Diego Center for Functional MRI. Functional images were obtained using a multi-echo echo planar imaging protocol with four echo times (TEs) of 15 ms, 28 ms, 42 ms, and 56 ms, a repetition time (TR) of 2,500 ms, a flip angle of 78°, a matrix size of 64 × 64, a slice thickness of 4 mm, and a field of view of 256 mm, covering 34 slices. Additionally, structural images were acquired using a T1-weighted 3D magnetization-prepared rapid gradient-echo sequence with a field of view of 256 mm, TE of 3.172 ms, TR of 8.142 ms, and a flip angle of 12°.

Imaging Data Preprocessing

Functional data underwent preprocessing using the multi-echo independent component analysis pipeline 'meica.py' implemented in AFNI and Python. Prior to preprocessing, the first four volumes of each run were discarded to ensure steady-state magnetization and ME-ICA denoising was completed. To denoise the data, principal and independent component analyses were used to separate BOLD and non-BOLD signals. Only BOLD-like components were retained after denoising and the time series of the four TEs were combined into a single time series. Preprocessing steps included motion correction, followed by slice timing correction and normalization to an age-matched toddler template (e.g., 2-year-old template) (Shi et al. 2011) as a majority of participants were around this age (mean age: 2.21 years), and smoothing. Head motion was assessed using framewise displacement (FD), with minimal motion observed in sleeping toddlers (mean FD <0.3 mm). No significant differences in head motion were observed between ASD and non-ASD groups.

Brain Dynamics

Post-ICA fMRI processing

The resting state data from all 75 toddlers were subjected to a high model order ICA by using the Group ICA of fMRI Toolbox (GIFT) (https://trendscenter.org/software/gift/). A model order of 30 independent components (ICs) was used as it is recommended in toddlers based on previous work (Ma, Wu, and Shi 2020). To ensure the stability of this estimation, the ICA algorithm was repeated 20 times using ICASSO (http://www.cis.hut.fi/projects/ica/icasso). The 30 components were visually inspected and classified as noise or non noise. The components

related to movement or white matter were removed from the analysis. The remaining 27 components were grouped into 7 functional networks based on a parcellation (Schaefer et al. 2018) (**Figure 1**). Then the SN, DMN, and CEN components were examined independently as part of a triple network approach as these networks are commonly indicated to be altered in ASD. A 284 volume × 27 ICs matrix containing the time series for each subject was post-processed using Matlab code from the GIFT toolbox. Post-processing included covariate regression of white matter, CSF, and Friston 24 head motion parameters, linear detrending, despiking (using AFNI's 3D despike), and bandpass filtering (0.01–0.1 Hz) (Allen et al., 2014). Global signal regression was not used in this study after the evaluation of the resulting matrices with and without global signal and based on methods in previous studies in this population (Marshall et al. 2020b).

Co-Activation Pattern Analysis

The individual matrices of the non-noise components were concatenated into a single group matrix comprising all subjects (21,300 TR (284 volume × 75 subjects) × 27 non-noise ICs) and subjected to k-means clustering. Various k values (2–20) were tested, and the optimal value of $k = 5$ was determined using the elbow criterion, which applies a least-squares fit line to the cluster validity index (ratio of within-cluster to between-cluster differences). Utilizing squared Euclidean distance to cluster the patterns, a Co-activation Pattern (CAP) analysis was conducted with the optimal value of $k = 5$ on the group matrix. ASD and non ASD participants were combined into one group for the CAP analysis based on previous findings indicating no group differences between CAPs when groups are separated (Kupis et al., 2020; Marshall et al., 2020).

Dynamic metrics were then calculated for each subject. The CAP metric of Dwell Time (DT) was calculated as the average number of TRs that a participant continuously remained in a given brain state. DT measures the average number of unchanged TRs between the state and the subsequent TR. Additionally, the frequency of occurrence of brain states was calculated as a percentage over time that the states were observed. Lastly, the number of transitions, representing switches between brain states, was calculated. DT, frequency of occurrence, and the number of transitions were computed for each participant and compared between groups, and further associated with cognitive flexibility measures (RRB, Vineland).

Sliding Window Dynamic Functional Network Connectivity

Dynamic functional network connectivity (dFNC) between all non-noise independent components (ICs) was computed using the GIFT dynamic-functional network connectivity (d-FNC) toolbox. This toolbox employs a sliding-window analysis to compute dynamic functional connectivity, where correlations between ICs are calculated within time-specified windows across the rsfMRI scan. Given our data sets repetition time (TR) of 2.5 seconds, we opted for sliding windows spanning 30 volumes. This resulted in 284 windows per subject. As part of calculating the window and correlation matrices, the d-FNC toolbox applies a 3-sigma Gaussian curve to smooth transitions between windows are the resulting windowed correlation matrices were regularized using the graphical LASSO method (Varoquaux et al. 2010) to minimize within-window noise. The graphical LASSO method estimates functional connectivity by applying L1 regularization to the inverse covariance matrix, improving model performance and promoting sparsity (Allen et al. 2014). All the dynamic functional networks across all subjects

were used to estimate the FC states using a k-means clustering analysis. This was repeated 100 times to cluster the dynamic FC windows. A Euclidean distance was used to group similar FC matrices of the different windows and elbow criterion was used to estimate the number of clusters, which was 5 states. The Pearson correlation coefficient was used for the clustering analysis.

Statistics

Group Differences

For both the CAP and dFNC analyses, between group differences (ASD vs. non ASD) were assessed using multiple regression controlling for age and head motion for each of the brain dynamic metrics (DT, frequency, and transitions for CAPS, and DT for DFC).

Brain-Behavior Relationships

For the CAP analysis, the relationship between the dynamic metrics (DT, frequency, and transitions) and behavior (RRBs, and Vineland Adaptive Scale) was assessed using multiple regressions controlling for age and head motion. For the DFC analyses, the relationship between DT and behavior (RRBs, and Vineland Adaptive scale) was assessed using multiple regressions controlling for age and head motion. All analyses were completed using R studio (www.rstudio.com).

RESULTS

CAPS

ICA

Out of 30 ICs that were estimated, three were classified as noise. The remaining components were then grouped into brain networks including visual, somatomotor, salience, dorsal attention, central executive, default mode, and subcortical regions (Figure 1A).

Figure 1

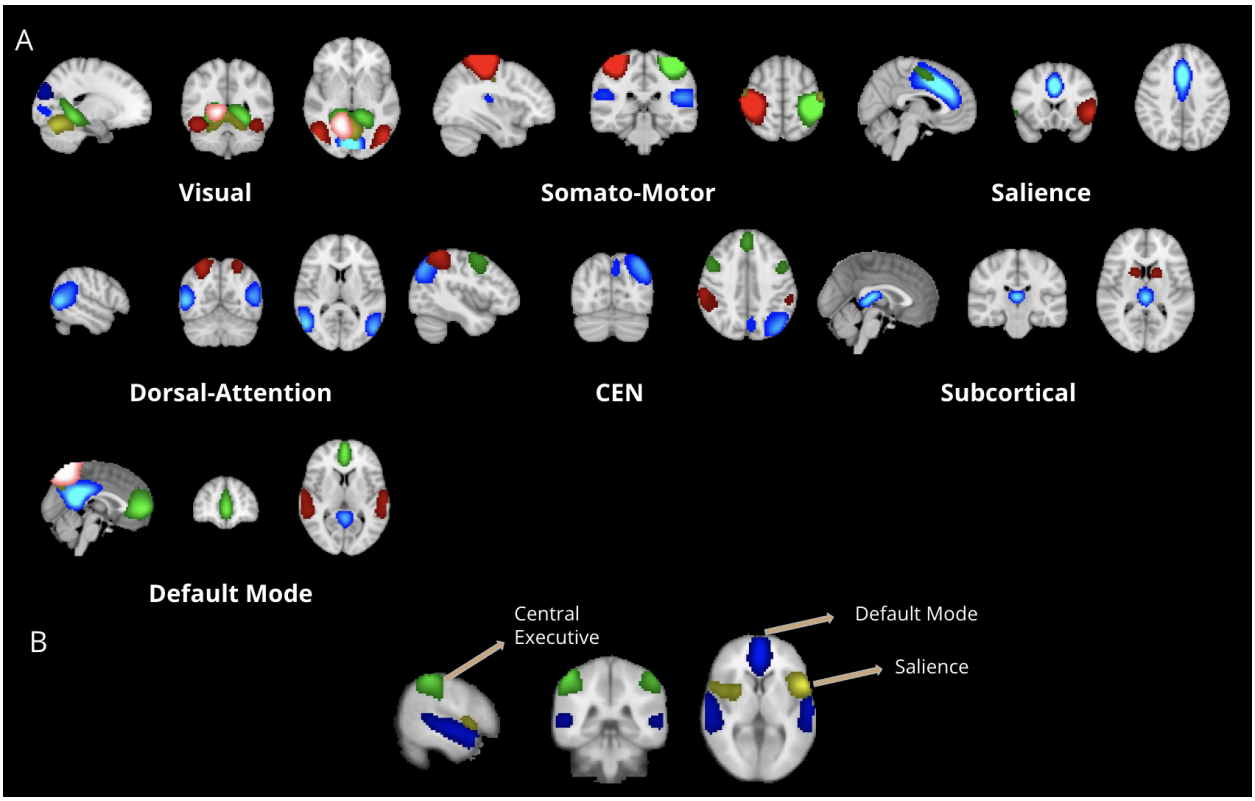

Figure 1. (A) Organization of ICA components into functional networks. (B) Triple network analysis of the salience, default mode, and central executive networks. CEN, central executive network.

Figure 2

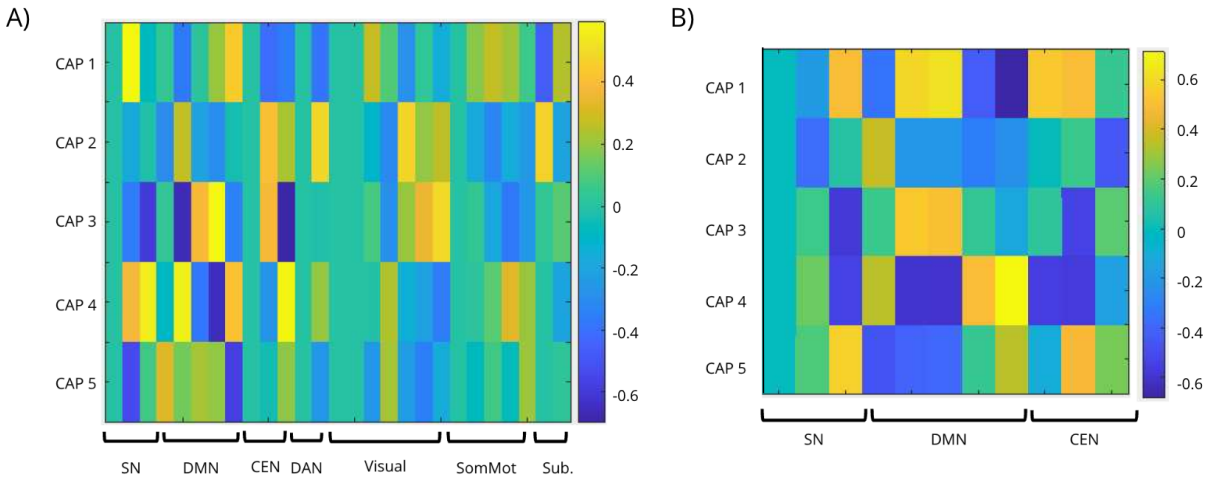

Figure 2. Whole brain and triple network co-activation patterns (CAPs)/Brain states. SN, salience network; DMN, default mode network; CEN, central executive network; DAN, dorsal attention network; SomMot, somato motor network; Sub, subcortical.

Whole Brain

There was an optimal k of 5 clusters across all toddlers. A) **CAP 1** was characterized by greater SN and DMN co-activation. **CAP 2** was characterized by greater CEN, DAN, DMN, visual, and subcortical network co-activation. **CAP 3** was characterized by greater DMN, CEN; and visual co-activation, and SN, DMN, and CEN de-activation. **CAP 4** was characterized by greater SN, DMN, and CEN co-activation. **CAP 5** was characterized by greater DMN activation; and SN and DMN de-activation. There was a significant group difference in CAP 4 dwell time *(p* = .04) when controlling for age and head motion. There were no other significant group differences in the whole-brain CAPs (*p*'s > .05) (Figure 3; Table 2).

Figure 3

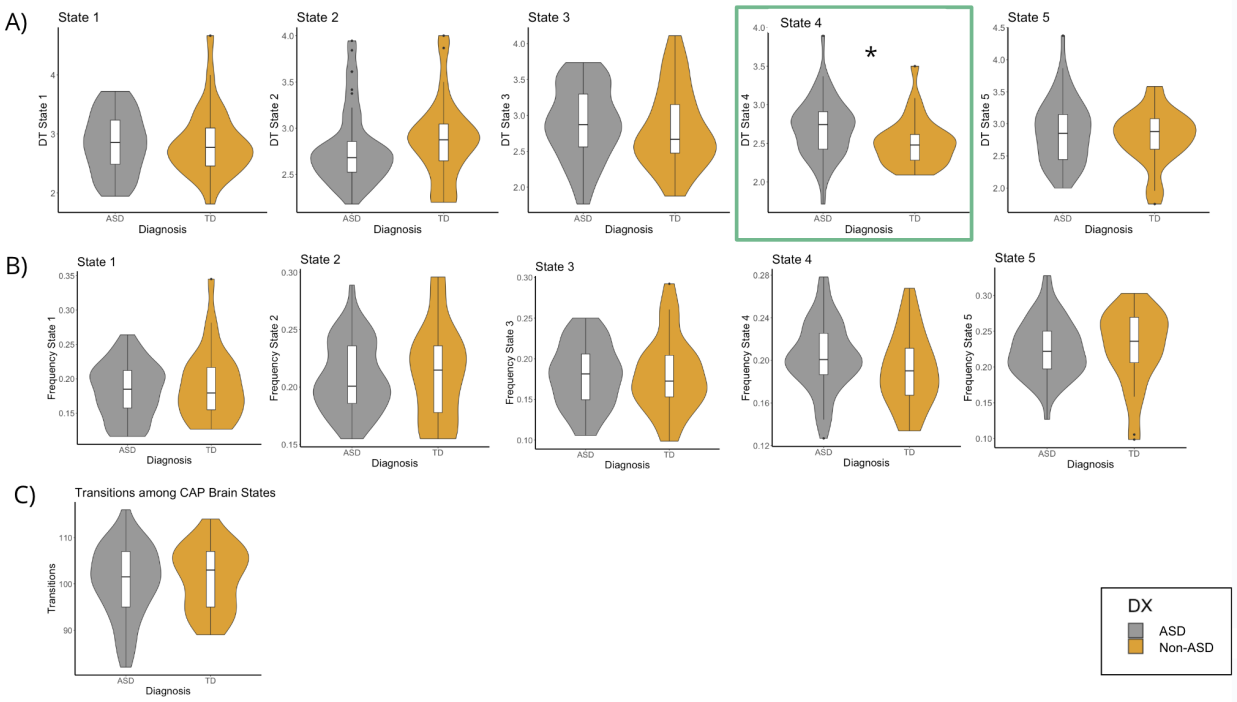

Figure 3. Between Group Differences. There was a significant group difference in the dwell time (DT) of state 4 between ASD and non-ASD groups such that ASD groups dwelled longer in this state. State 4 consisted of greater SN, CEN, and DMN co-activation.

Table 2

	Dynamic Metric	ASD (N = 48) M (SD)	Non ASD (N = 27) M (SD)	*p*-value
CAP 1	DT	2.55 (.38)	2.86 (.53)	.86
	Frequency	.19 (.04)	.19 (.04)	.80
	DT	2.52 (.45)	2.88 (.40)	.32

CAP 2	Frequency	.17 (.04)	.21 (.03)	.35
CAP 3	DT	2.62 (.45)	2.82 (.53)	.36
	Frequency	.24 (.03)	.18 (.04)	.74
CAP 4	DT	2.52 (.51)	2.52 (.37)	.04*
	Frequency	.20 (.04)	.19 (.03)	.20
CAP 5	DT	2.33 (.34)	2.80 (.45)	.61
	Frequency	.20 (.03)	.23 (.05)	.78

Triple-Network Approach

Next, a triple network analysis of the salience, default mode, and central executive network dynamics across 5 brain states was examined. **CAP 1** was characterized by greater SN, DMN, and CEN co-activation and DMN de-activation. **CAP 2** had no strong co-activations. **CAP 3** consisted of a greater DMN co-activation, and SN and CEN de-activation. **CAP 4** consisted of greater DMN co-activation, and SN, DMN, and CEN de-activation. **CAP 5** consisted of a greater SN and CEN co-activation, and DMN de-activation (**Figure 2B**). There were no significant group differences between the groups in the ROI CAP analyses (*p*'s > .05) (Table 3).

Table 3

	Dynamic Metric	ASD (N = 48) M (SD)	Non-ASD (N = 27) M (SD)	*p*-value
CAP 1	DT	2.93 (.50)	2.79 (.51)	.19
	Frequency	.19 (.04)	.19 (.04)	.99
CAP 2	DT	2.38 (.36)	2.32 (.35)	.62
	Frequency	.20 (.04)	.19 (.04)	.31
CAP 3	DT	2.78 (.45)	2.66 (.45)	.32
	Frequency	.23 (.04)	.22 (.04)	.68
CAP 4	DT	3.00 (.60)	2.90 (.62)	.36
	Frequency	.19 (.04)	.19 (.04)	.74
CAP 5	DT	2.75 (.42)	2.79 (.39)	.41
	Frequency	.20 (.04)	.21 (.04)	.07

CAPS Brain Behavior

A dimensional analysis across both diagnostic groups was conducted to evaluate relationships between brain dynamics (dwell time, frequency, transitions) and RRBs and adaptive behaviors.

Whole Brain

First, the model was conducted using dynamics across the whole brain CAPs. A dimensional brain-behavior regression model revealed a significant relationship between the frequency of CAP 5 and ADOS RRB ($p = .038$) when controlling for age and head motion. There was also a significant relationship between the frequency of CAP 2 and the Vineland Adaptive Behavior measure ($p = .048$). There were no other significant brain-behavior relationships in the whole brain CAP dynamics with behavioral measures ($p>.05$).

Triple Network Analysis

Next, the regression model was conducted using the triple network CAP dynamics (SN, DMN, CEN) and their relationship with RRBs and adaptive behavior. Brain-behavior relationships were observed between the DT of CAP 1 and the ADOS RRB measure ($p = .034$), and the frequency of CAP 5 and the ADOS RRB measure ($p = .024$). Brain-behavior relationships were also observed between the DT of CAP 1 and the Vineland Adaptive Behavior measure ($p = .046$), and the frequency of CAP 5 and the Vineland Adaptive Behavior measure ($p < .001$). All significant results are summarized in Table 4 and Figure 4.

Table 4. Summary of the regression analysis of CAP Brain States dynamic metrics and behavior measures. To simplify the presentation, we present only the variables of interest and not the covariates, and only significant findings.

	b	*SE*	*t*	*p*
CAP State 4 DT and Diagnosis				
	-.18	.09	-2.09	.041
CAP State 5 Frequency and RRB				
	-.004	.002	-2.11	.038
CAP State 2 Frequency and Vineland Adaptive Behavior				
	.001	.0003	2.01	.048
ROI CAP State 1 DT and RRB				
	.05	.02	2.17	.034
ROI CAP State 5 Frequency and RRB				
	-.004	.002	-2.30	.024
ROI CAP State 1 DT and Vineland Adaptive Behavior				
	-.01	.005	-2.03	.046
ROI CAP State 5 Frequency and Vineland Adaptive Behavior				
	.001	.0005	3.66	.0005

* Uncorrected p values < .05
** Uncorrected p values < .01
*** Uncorrected p values < .001
ROI, region of interest

Figure 4

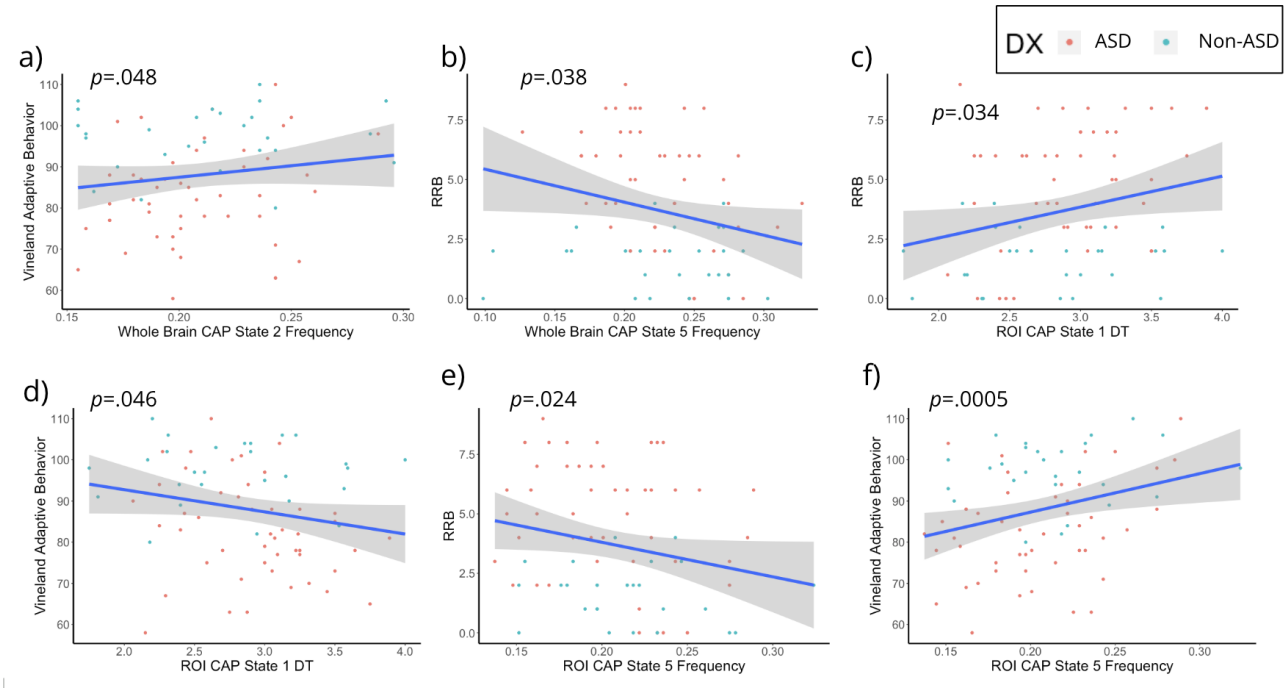

Figure 4. A) Greater frequency of the whole brain state consisting of greater CEN, DAN, DMN, visual, and subcortical network co-activation was associated with greater adaptive behaviors. Conversely, in B), greater frequency of the whole brain state consisting of greater DMN activation; and SN and DMN de-activation was associated with poorer restricted repetitive behaviors (RRBs). In the region of interest (ROI) or triple network states, C), greater dwell time of a state consisting of greater SN, DMN, and CEN co-activation and DMN de-activation was associated with poorer RRBs and D) poorer adaptive behaviors. Next, the greater frequency of a state consisting of greater SN and CEN co-activation, and DMN de-activation was associated with E) fewer RRBs, and F) greater adaptive behaviors.

Figure 5

Dynamic Functional Connectivity (dFC)

ICA

The same ICA process for DFC was conducted for DFC using the GIFT toolbox and as described above.

DFC States

Following the elbow criterion, the whole sample showed 5 different states (see Figure 6). State 1 (14% of the windows) was characterized by strong connectivity between visual and subcortical networks and slightly elevated connectivity with the salience network. State 2 (22% of the windows) was characterized by strong connectivity between the subcortical, salience, and a few regions in the visual networks. State 3 (9% of the windows), was characterized by strong connectivity amongst the visual, somato motor, subcortical, DMN, and salience networks. State 4 (34% of the windows) was characterized by strong connectivity within the subcortical network, and weak connectivity between the DAN and DMN. State 5 (22% of the windows), was characterized by strong connectivity within visual and subcortical networks, and weak connectivity between the DAN and DMN.

Figure 5

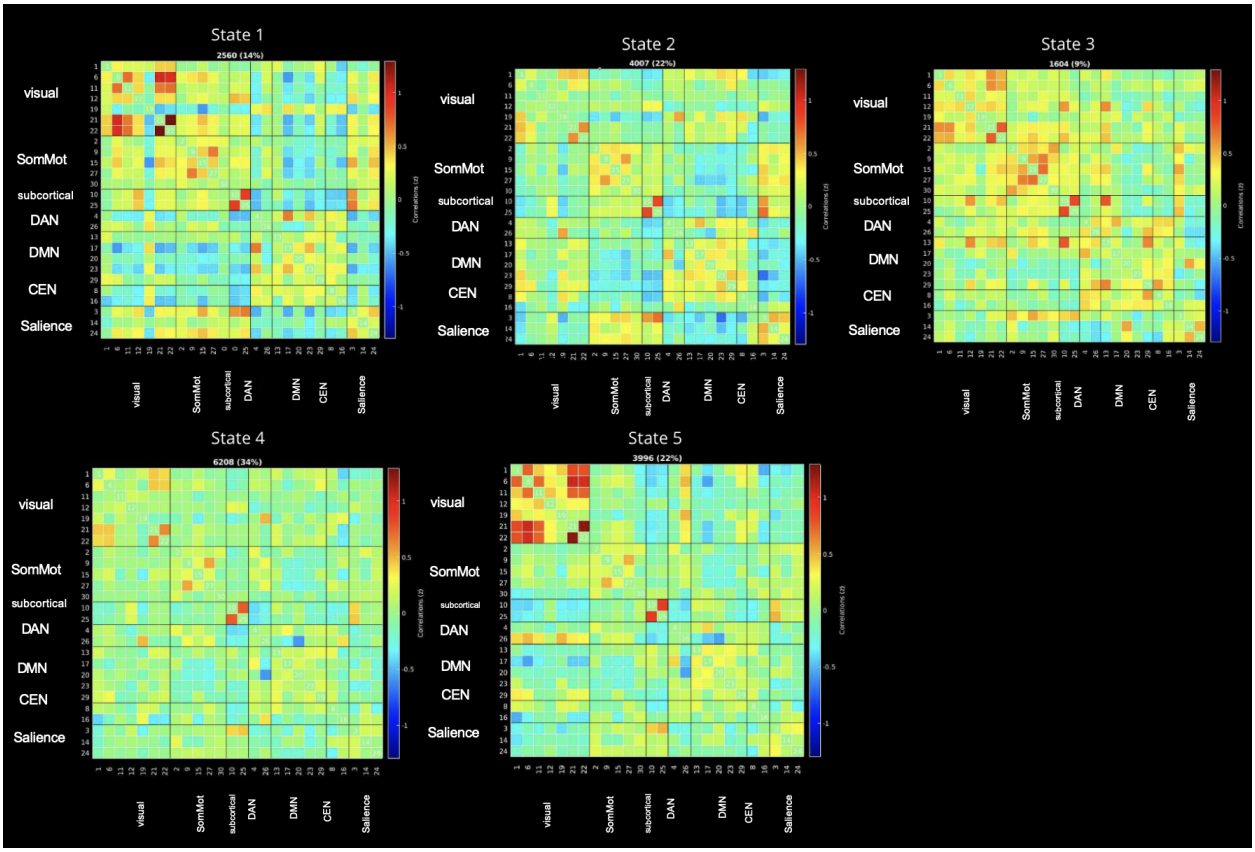

Figure Legend: Dynamic functional network connectivity (dFNC) matrices for all subjects and 5 states. The value in each cell in the FC is the Pearson correlation coefficient between two brain regions. The color bar represents the strength of the FC between two nodes (warm color, positive FC; cool color, negative FC). SomMot, somatomotor; DAN, dorsal attention network; DMN, default mode network; CEN, central executive network.

Group

There were no significant group differences in the dynamic brain state dwell times when controlling for age and head motion (*p*'s > .05).

Table 5

	ASD (N = 48) M (SD)	TD (N = 27) M (range)	*p*-value
CAP 1 DT	16.65 (24.89)	20.39 (25.46)	.76
CAP 2 DT	18.15 (18.08)	22.35 (18.31)	.24
CAP 3 DT	12.07 (30.79)	15.72 (31.74)	.83
CAP 4 DT	28.60 (37.62)	38.85 (37.58)	.77
CAP 5 DT	35.77 (50.20)	10.40 (51.74)	.08

Brain Behavior

A dimensional multiple regression analysis between the dynamic FC dwell times and behavior measures while controlling for age and head motion revealed significant relationships between State 4 DT and Vineland adaptive behavior (p = .049), and State 5 DT and ADOS RRB (p =.034). These results are after the removal of outliers. There were no other significant brain-behavior relationships (p's > .05).

Table 6. Summary of the regression analysis of DFC dynamic metrics and behavior measures. To simplify the presentation, we present only the variables of interest and not the covariates, and significant findings.

	b	*SE*	*t*	*p*

DFC State 4 DT and Vineland Adaptive Behavior				
	.56	.27	2.00	.049*
DFC State 5 DT and RRB				
	2.27	1.05	2.17	.034*

* Uncorrected p values < .05

Figure 6

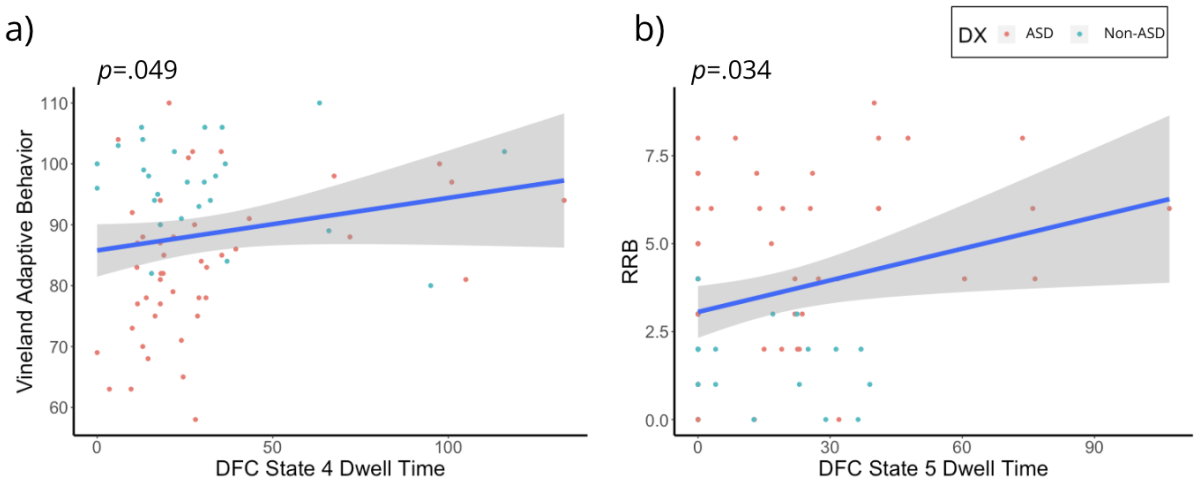

Figure 6. A) Greater dwell time of a state consisting of strong connectivity with the subcortical, somato motor, and visual network nodes, and negative connectivity between the DAN and DMN was associated with greater adaptive behaviors. B) Greater dwell time of a state consisting of strong connectivity between visual and subcortical networks, and weak connectivity between the DAN and DMN was associated with poorer restricted and repetitive behaviors (RRBs).

DISCUSSION

The current study sought to identify early brain biomarkers for autism spectrum disorder (ASD) in toddlers prior to their diagnosis of ASD and identify brain-behavior relationships in toddlers who went on to receive an ASD and non-ASD diagnosis. The current study used a data-driven, ICA approach, and investigated brain dynamics using two methods: co-activation pattern analysis (CAP) and dynamic functional connectivity (DFC). While investigating both ASD and non-ASD toddlers, we observed that toddlers who went on to receive an ASD diagnosis dwelled longer in a brain state characterized by salience network (SN), central executive network (CEN), and default mode network (DMN) co-activation compared to non-ASD toddlers. Additionally, across both diagnostic groups, early brain dynamics in states associated with the core neural networks (DMN, SN, CEN) were associated with restricted and repetitive behaviors (RRBs) and behavioral flexibility.

First we found that ASD toddlers dwelled longer in a state consisting of co-activations of SN, CEN, and DMN (state 4) compared with non-ASD toddlers. Many psychiatric conditions including ASD are often characterized by dysfunction among these three neural networks (SN, CEN, DMN) (Jones et al. 2023; B. Menon 2019; Vinod Menon 2011). This finding is supported by the triple network theory that proposes the interactions among the SN, CEN, and DMN are important for mental health and underlies the dysfunction observed in ASD (Vinod Menon 2011). Separately the networks hold important roles for cognition. The CEN is thought to be involved in cognitive decisions and tasks (e.g., executive function) and associated with flexible cognition (Marek and Dosenbach 2018); The DMN is thought to be involved in internal processes and social cognition such as theory of mind (Raichle 2015); The SN, is thought to

monitor salient information internally or in the environment and coordinate switching between the CEN and DMN for either task based (CEN) or internally focused (DMN) states (V. Menon 2015b; Chand et al. 2017). The triple network model suggests that the interactions among the SN, CEN, and DMN are critical to overall cognitive function. However, in ASD and many psychiatric conditions, the switching between these neural networks underlying cognition appears to be dysfunctional (B. Menon 2019). Previous work has found under and over connectivity of these three neural networks in ASD (K. Wang, Li, and Niu 2021; L. Wang et al. 2024; Hull et al. 2016; Lucina Q. Uddin, Supekar, and Menon 2013). Dysfunction within and among these three networks may result in network isolation especially during task states, limiting dynamic interactions between networks important for flexible cognition and behaviors (Cole et al. 2014). Altered connectivity within these networks have also been linked with and predictive of behavioral difficulties such as social communication deficits, RRBs, and adaptive behavior skills (McKinnon et al. 2019; Kaustubh Supekar et al. 2021; Plitt et al. 2015; Abbott et al. 2016). Dysfunction of these neural networks appear to underlie many of the symptoms observed in ASD and may underlie our finding of there being a greater dwell time of a state consisting of co-activation of all three neural networks in ASD toddlers. Additionally, dynamic connectivity patterns in brain states and among the SN, CEN, and DMN are altered in ASD individuals compared with non-ASD individuals (Marshall et al. 2020b; Kupis, Romero, Dirks, Hoang, Parladé, et al. 2020; Kupis, Goodman, Kircher, et al. 2021; Yue et al. 2022). ASD individuals have been found to dwell longer and transition less between states (Yao et al. 2016; Watanabe and Rees 2017; de Lacy et al. 2017; “Brain Mechanisms Supporting Flexible Cognition and

Behavior in Adolescents With Autism Spectrum Disorder" 2021), and spend more time in states with altered patterns such as abnormal patterns of increased DMN activation (Harlalka et al. 2019; Yue et al. 2022), as similarly found in this study. There is also some evidence of early functional connectivity dysfunction within these networks in high risk or later developing autistic infants and toddlers (McKinnon et al. 2019; Tsang et al. 2024). As these three neural networks are found to develop across the third trimester of gestation and the first postnatal month (Scheinost et al. 2022), it makes it even more important to further investigate in autism at these early stages.

Across all participants, we found states with primarily CEN and SN co-activation and DMN de-activation were associated with greater adaptive behaviors and fewer RRBs; conversely states with greater DMN co-activation were associated with poorer RRBs and adaptive behaviors. These findings are consistent with previous studies and relationships between brain and behavior findings (Sridharan, Levitin, and Menon 2008). Both the SN and CEN are often associated with cognitive processes (Sridharan, Levitin, and Menon 2008; Vinod Menon and D'Esposito 2022; Aron 2007; Dreher and Berman 2002; Bressler and Menon 2010; Shaw et al. 2021) and have been associated with RRBs and adaptive behavior in previous studies with and without ASD participants (Lucina Q. Uddin et al. 2013b; Kupis, Romero, Dirks, Hoang, Parladé, et al. 2020; Kupis, Goodman, Kornfeld, et al. 2021; He et al. 2018; Harlalka et al. 2019; P. Lin et al. 2016; "Brain Mechanisms Supporting Flexible Cognition and Behavior in Adolescents With Autism Spectrum Disorder" 2021). Additionally, atypical brain network dynamics have been shown to underlie ASD and contribute to individuals' cognitive and behavioral inflexibility

(Lucina Q. Uddin et al. 2015b; Marshall et al. 2020b; "Brain Mechanisms Supporting Flexible Cognition and Behavior in Adolescents With Autism Spectrum Disorder" 2021). Proper functioning and switching between these three neural networks are broadly associated with healthy cognition (Chand et al. 2017). Dysfunction of the switching process, or disengagement of the CEN, SN, or DMN may contribute to cognitive and behavioral difficulties, and psychiatric conditions (Lucina Q. Uddin 2015; Kaustubh Supekar et al. 2019). In healthy populations, CEN and SN interactions in particular support positive cognitive control, behavioral and cognitive flexibility, and adaptive behaviors (Kupis, Goodman, Kornfeld, et al. 2021; Krönke et al. 2020; Cole et al. 2014). Greater DMN interactions with the SN typically support self referential processes (Gusnard et al. 2001), and when present during task states is usually associated with poorer task performance (Hutchison et al. 2013; V. Menon 2015a). Our finding of the increased state including DMN co-activation with the SN and CEN in toddlers who were later diagnosed with ASD compared with non-ASD toddlers may be an early sign of network dysfunction as observed in older children and individuals with ASD. Although brain network dynamics are not greatly studied in infants and toddlers at this time, there is promising work showing early brain alterations are predictive of later ASD diagnosis and cognitive and behavior functions (I. Molnar-Szakacs, Kupis, and Uddin 2021; Wolff, Jacob, and Elison 2018; Girault and Piven 2020). Overall, our findings support the SN, CEN, and DMN as promising brain biomarkers for individuals at risk of ASD during young development and later emerging behavior difficulties associated with RRBs and behavioral flexibility.

In the dFNC method used (a sliding window approach), we found greater dwell time in a state consisting of weak overall connectivity and strong subcortical connectivity was associated with greater adaptive behavior; conversely greater dwell time in a state consisting of greater visual and subcortical connectivity was associated with poorer RRBs across all participants. In both cases, we found a weak (negative) connectivity between the DAN and DMN. Our findings are similar to findings in the literature using dFNC. For example, other studies have found DFC states predictive of symptom severity were mostly those associated with visual and sensorimotor regions (Harlalka et al. 2019; McKinnon et al. 2019). One of the earliest studies of FC and RRBs in toddlers at risk of ASD found abnormal brain network dynamics with the DMN, DAN and CEN were associated with RRBs and rigid behaviors (McKinnon et al. 2019). They found further evidence to support the hypothesis that inverse relationships between the DAN and DMN underlie healthy and adaptive behaviors. Here, we found weak FC between the DAN and DMN was associated with both positive and negative flexible behaviors. Our slightly different findings may be due to having a larger age range in our toddler sample, whereas the previous study separated toddlers by 12 mos and 24 mos of age. RRBs have also been previously linked to visual skills in ASD before and greater visual sensitivity has been correlated with more severe RRBs (W. Lin et al. 2023; Schulz and Stevenson 2020). Further studies have found RRBs may even be a response to sensory abnormalities (Gabriels et al. 2008) which supports the theory that early sensory abnormalities may be associated with long term dysfunction in higher order behaviors. Our finding of a longer dwell time in a visual/subcortical FC state associated with poorer RRBs further supports the idea of sensory abnormalities linked with later RRBs. One

study with some of the youngest infants at-risk for ASD (1.5 months and 9 months) found evidence of abnormal thalamus connectivity in at-risk infants, suggesting early subcortical, visual, and sensory abnormalities may cascade into later ASD symptoms and higher order issues (Wagner et al. 2023). Overall, our dFNC findings further support the ideas of early sensory connections underlie the development of later emerging RRBs and adaptive behaviors.

In this study, we utilized two methods to investigate early brain dynamics in toddlers with and without ASD. The dynamic functional network connectivity (dFNC; e.g., sliding window) (Chang and Glover 2010) and co-activation pattern analysis (CAP) (X. Liu and Duyn 2013) methods were used and are both popularly used to quantify brain dynamics (Lurie et al. 2020). Both DFC and CAP methods are similar in that they account for brain network changes across multiple time points throughout the scan, rather than averaging across the entire duration of the scan. DFNC, however, uses sliding windows, or 30-60 second windows that break up the time series. From there an average is taken from each window, thus arbitrarily collapsing the data into time. CAPs on the other hand, does not collapse the data into arbitrary window lengths and instead relies on all of the data and a data-driven approach to cluster the data into the brain states. These methodological differences, further depicted here, may account for the differences observed in the results between these two methods. In the whole-brain CAP analyses, we found most of the brain states contained a variety of co-activated brain regions including the SN, DMN, and CEN. Only CAP brain state 5 had weaker co-activations of brain regions. These findings are similar to other CAP studies (Marshall et al. 2020b; Kupis, Romero, Dirks, Hoang, Parladé, et al. 2020; Kupis, Goodman, Kornfeld, et al. 2021). The dFNC states had similar connectivity

patterns including strong FC of the core neural networks as in the CAP states but had more states with a stronger visual network FC. Additionally, the dFNC results had states of widespread connectivity and low strength in connectivity. Interestingly, studies using dFNC methods typically report that ASD groups tend to spend more time in FC states characterized by weak or absent connectivity compared with healthy controls (Rabany et al. 2019; de Lacy et al. 2017; Rashid et al. 2018; Chen et al. 2017). Although we did not find a significant difference in dFNC states in ASD versus non-ASD groups, overall we found similar states to previous studies. In both methods, we did find brain-behavior relationships across diagnostic groups. Although we found differences in the dFNC and CAPs methods, both methods may be used to reveal interesting and nuanced insights into early brain development and brain-behavior relationships.

Limitations of this study include a limited sample size. Collecting sleep MRI scans poses a challenge as it is difficult to recruit families for night-time scans and there are difficulties with children remaining asleep during the scan duration. Additionally, there were too few neurotypical toddlers to create a diagnostic group based on that criteria. Therefore, the non-ASD group was based on the standardized scores to allow for more toddlers within the neurotypical range and to include more toddlers of varying levels as suggested by RDOC criteria. Additionally, sleep dynamics and dynamics in toddlers in general are a new field and more work is needed for this research. Additionally, further analyses need to be done with the dFNC method including a triple network approach, and investigating the results using more sliding window lengths (e.g., 30 sec, 60 sec, 45 sec) and a comparison of global signal regression on the dFNC results.

REFERENCES

Abbott, Angela E., Aarti Nair, Christopher L. Keown, Michael Datko, Afrooz Jahedi, Inna Fishman, and Ralph-Axel Müller. 2016. "Patterns of Atypical Functional Connectivity and Behavioral Links in Autism Differ Between Default, Salience, and Executive Networks." *Cerebral Cortex* 26 (10): 4034–45.

Abramowitz, Jonathan S., and Ryan J. Jacoby. 2015. "Obsessive-Compulsive and Related Disorders: A Critical Review of the New Diagnostic Class." *Annual Review of Clinical Psychology* 11 (January):165–86.

Alcauter, Sarael, Weili Lin, J. Keith Smith, Sarah J. Short, Barbara D. Goldman, J. Steven Reznick, John H. Gilmore, and Wei Gao. 2014. "Development of Thalamocortical Connectivity during Infancy and Its Cognitive Correlations." *The Journal of Neuroscience: The Official Journal of the Society for Neuroscience* 34 (27): 9067–75.

Allen, Elena A., Eswar Damaraju, Sergey M. Plis, Erik B. Erhardt, Tom Eichele, and Vince D. Calhoun. 2014. "Tracking Whole-Brain Connectivity Dynamics in the Resting State." *Cerebral Cortex* 24 (3): 663–76.

American Psychiatric Association. 2015. *DSM-5 Classification*. American Psychiatric Publishing.

———. 2021. *Diagnostic and Statistical Manual of Mental Disorders (DSM-5)*. American Psychiatric Publishing.

Anderson, Kristy A., Paul T. Shattuck, Benjamin P. Cooper, Anne M. Roux, and Mary Wagner. 2014. "Prevalence and Correlates of Postsecondary Residential Status among Young Adults with an Autism Spectrum Disorder." *Autism: The International Journal of Research and Practice* 18 (5): 562–70.

Aqdassi, Louisa, Saeid Sadeghi, Hamid Reza Pouretemad, and Jalil Fathabadi. 2021. "A Family-Based Telerehabilitation Program for Improving Gross Motor Skills in Children With High Functioning Autism Spectrum Disorder." *Journal of Modern Rehabilitation*, November. https://doi.org/10.18502/jmr.v15i3.7738.

Aron, Adam R. 2007. "The Neural Basis of Inhibition in Cognitive Control." *The Neuroscientist: A Review Journal Bringing Neurobiology, Neurology and Psychiatry* 13 (3): 214–28.

Barbaro, Josephine, and Cheryl Dissanayake. 2009. "Autism Spectrum Disorders in Infancy and Toddlerhood: A Review of the Evidence on Early Signs, Early Identification Tools, and Early Diagnosis." *Journal of Developmental and Behavioral Pediatrics: JDBP* 30 (5): 447–59.

Bishop, Somer L., Jennifer Richler, Albert C. Cain, and Catherine Lord. 2007. "Predictors of Perceived Negative Impact in Mothers of Children with Autism Spectrum Disorder." *American Journal of Mental Retardation: AJMR* 112 (6): 450–61.

Biswal, B., F. Z. Yetkin, V. M. Haughton, and J. S. Hyde. 1995. "Functional Connectivity in the Motor Cortex of Resting Human Brain Using Echo-Planar MRI." *Magnetic Resonance in Medicine: Official Journal of the Society of Magnetic Resonance in Medicine / Society of Magnetic Resonance in Medicine* 34 (4): 537–41.

"Brain Mechanisms Supporting Flexible Cognition and Behavior in Adolescents With Autism Spectrum Disorder." 2021. *Biological Psychiatry* 89 (2): 172–83.
Braun, Urs, Axel Schäfer, Henrik Walter, Susanne Erk, Nina Romanczuk-Seiferth, Leila Haddad, Janina I. Schweiger, et al. 2015. "Dynamic Reconfiguration of Frontal Brain Networks during Executive Cognition in Humans." *Proceedings of the National Academy of Sciences* 112 (37): 11678–83.
Bray, S., A. E. Arnold, R. M. Levy, and G. Iaria. 2015. "Spatial and Temporal Functional Connectivity Changes between Resting and Attentive States." *Human Brain Mapping* 36 (2). https://doi.org/10.1002/hbm.22646.
Bressler, Steven L., and Vinod Menon. 2010. "Large-Scale Brain Networks in Cognition: Emerging Methods and Principles." *Trends in Cognitive Sciences* 14 (6): 277–90.
Cao, Jianghui, Guangzhi Liu, Xuekun Li, Zheng Yue, Jipeng Ren, Wei Zhu, and Baolin Wu. 2021. "Dynamic Functional Connectivity Changes in the Triple Networks and Its Association with Cognitive Impairment in Hemodialysis Patients." *Brain and Behavior* 11 (8). https://doi.org/10.1002/brb3.2314.
Chand, Ganesh B., Junjie Wu, Ihab Hajjar, and Deqiang Qiu. 2017. "Interactions of the Salience Network and Its Subsystems with the Default-Mode and the Central-Executive Networks in Normal Aging and Mild Cognitive Impairment." *Brain Connectivity* 7 (7): 401–12.
Chang, Catie, and Gary H. Glover. 2010. "Time-Frequency Dynamics of Resting-State Brain Connectivity Measured with fMRI." *NeuroImage* 50 (1): 81–98.
Chen, Heng, Jason S. Nomi, Lucina Q. Uddin, Xujun Duan, and Huafu Chen. 2017. "Intrinsic Functional Connectivity Variance and State-Specific under-Connectivity in Autism." *Human Brain Mapping* 38 (11): 5740–55.
"Cognitive Flexibility as the Shifting of Brain Network Flows by Flexible Neural Representations." 2024. *Current Opinion in Behavioral Sciences* 57 (June):101384.
Cohen, J. R. 2018. "The Behavioral and Cognitive Relevance of Time-Varying, Dynamic Changes in Functional Connectivity." *NeuroImage* 180 (Pt B). https://doi.org/10.1016/j.neuroimage.2017.09.036.
Cole, Michael W., Danielle S. Bassett, Jonathan D. Power, Todd S. Braver, and Steven E. Petersen. 2014. "Intrinsic and Task-Evoked Network Architectures of the Human Brain." *Neuron* 83 (1): 238–51.
"Connectivity Dynamics from Wakefulness to Sleep." 2020. *NeuroImage* 220 (October):117047.
Courchesne, E., H. J. Chisum, J. Townsend, A. Cowles, J. Covington, B. Egaas, M. Harwood, S. Hinds, and G. A. Press. 2000. "Normal Brain Development and Aging: Quantitative Analysis at in Vivo MR Imaging in Healthy Volunteers." *Radiology* 216 (3): 672–82.
Courchesne, Eric, Vahid H. Gazestani, and Nathan E. Lewis. 2020. "Prenatal Origins of ASD: The When, What, and How of ASD Development." *Trends in Neurosciences* 43 (5): 326–42.
Craig, F., F. Margari, A. R. Legrottaglie, R. Palumbi, C. de Giambattista, and L. Margari. 2016. "A Review of Executive Function Deficits in Autism Spectrum Disorder and Attention-Deficit/hyperactivity Disorder." *Neuropsychiatric Disease and Treatment* 12:1191–1202.
Cuccaro, Michael L., Yujan Shao, Janet Grubber, Michael Slifer, Chantelle M. Wolpert,

Shannon L. Donnelly, Ruth K. Abramson, et al. 2003. “Factor Analysis of Restricted and Repetitive Behaviors in Autism Using the Autism Diagnostic Interview-R.” *Child Psychiatry and Human Development* 34 (1): 3–17.
Cuthbert, Bruce N. 2022. “Research Domain Criteria (RDoC): Progress and Potential.” *Current Directions in Psychological Science* 31 (2): 107.
Damaraju, E., E. A. Allen, A. Belger, J. M. Ford, S. McEwen, D. H. Mathalon, B. A. Mueller, et al. 2014. “Dynamic Functional Connectivity Analysis Reveals Transient States of Dysconnectivity in Schizophrenia.” *NeuroImage. Clinical* 5 (July):298–308.
Dichter, Gabriel S. 2012. “Functional Magnetic Resonance Imaging of Autism Spectrum Disorders.” *Dialogues in Clinical Neuroscience* 14 (3): 319–51.
Dreher, Jean-Claude, and Karen Faith Berman. 2002. “Fractionating the Neural Substrate of Cognitive Control Processes.” *Proceedings of the National Academy of Sciences of the United States of America* 99 (22): 14595–600.
Easson, Amanda K., Zainab Fatima, and Anthony R. McIntosh. 2019. “Functional Connectivity-Based Subtypes of Individuals with and without Autism Spectrum Disorder.” *Network Neuroscience* 3 (2): 344.
Edward Roberts, R., Elaine J. Anderson, and Masud Husain. 2013. “White Matter Microstructure and Cognitive Function.” *The Neuroscientist: A Review Journal Bringing Neurobiology, Neurology and Psychiatry* 19 (1): 8.
Elder, Jennifer Harrison, Consuelo Maun Kreider, Susan N. Brasher, and Margaret Ansell. 2017. “Clinical Impact of Early Diagnosis of Autism on the Prognosis and Parent-Child Relationships.” *Psychology Research and Behavior Management* 10 (August):283–92.
Elison, Jed T., Jason J. Wolff, J. Steven Reznick, Kelly N. Botteron, Annette M. Estes, Hongbin Gu, Heather C. Hazlett, et al. 2014. “Repetitive Behavior in 12-Month-Olds Later Classified with Autism Spectrum Disorder.” *Journal of the American Academy of Child and Adolescent Psychiatry* 53 (11): 1216–24.
Emerson, Robert W., Chloe Adams, Tomoyuki Nishino, Heather Cody Hazlett, Jason J. Wolff, Lonnie Zwaigenbaum, John N. Constantino, et al. 2017. “Functional Neuroimaging of High-Risk 6-Month-Old Infants Predicts a Diagnosis of Autism at 24 Months of Age.” *Science Translational Medicine* 9 (393). https://doi.org/10.1126/scitranslmed.aag2882.
Emerson, Robert W., Wei Gao, and Weili Lin. 2016. “Longitudinal Study of the Emerging Functional Connectivity Asymmetry of Primary Language Regions during Infancy.” *The Journal of Neuroscience: The Official Journal of the Society for Neuroscience* 36 (42): 10883–92.
Eyler, Lisa T., Karen Pierce, and Eric Courchesne. 2012. “A Failure of Left Temporal Cortex to Specialize for Language Is an Early Emerging and Fundamental Property of Autism.” *Brain: A Journal of Neurology* 135 (Pt 3): 949–60.
Farley, Megan A., William M. McMahon, Eric Fombonne, William R. Jenson, Judith Miller, Michael Gardner, Heidi Block, et al. 2009. “Twenty-Year Outcome for Individuals with Autism and Average or near-Average Cognitive Abilities.” *Autism Research: Official Journal of the International Society for Autism Research* 2 (2): 109–18.
Friston, K. J., C. D. Frith, P. F. Liddle, and R. S. Frackowiak. 1993. “Functional Connectivity:

The Principal-Component Analysis of Large (PET) Data Sets." *Journal of Cerebral Blood Flow and Metabolism: Official Journal of the International Society of Cerebral Blood Flow and Metabolism* 13 (1): 5–14.

Gabriels, Robin L., John A. Agnew, Lucy Jane Miller, Jane Gralla, Zhaoxing Pan, Edward Goldson, James C. Ledbetter, Juliet P. Dinkins, and Elizabeth Hooks. 2008. "Is There a Relationship between Restricted, Repetitive, Stereotyped Behaviors and Interests and Abnormal Sensory Response in Children with Autism Spectrum Disorders?" *Research in Autism Spectrum Disorders* 2 (4): 660–70.

Gao, Wei, Amanda Elton, Hongtu Zhu, Sarael Alcauter, J. Keith Smith, John H. Gilmore, and Weili Lin. 2014. "Intersubject Variability of and Genetic Effects on the Brain's Functional Connectivity during Infancy." *The Journal of Neuroscience: The Official Journal of the Society for Neuroscience* 34 (34): 11288–96.

Gardiner, E., and G. Iarocci. 2017. "Everyday Executive Function Predicts Adaptive and Internalizing Behavior among Children with and without Autism Spectrum Disorder." *Autism Research: Official Journal of the International Society for Autism Research*. https://doi.org/10.1002/aur.1877.

Gilmore, John H., Rebecca Knickmeyer Santelli, and Wei Gao. 2018. "Imaging Structural and Functional Brain Development in Early Childhood." *Nature Reviews. Neuroscience* 19 (3): 123.

Gilotty, L., L. Kenworthy, L. Sirian, D. O. Black, and A. E. Wagner. 2002. "Adaptive Skills and Executive Function in Autism Spectrum Disorders." *Child Neuropsychology: A Journal on Normal and Abnormal Development in Childhood and Adolescence* 8 (4): 241–48.

Gioia, G. A., P. K. Isquith, L. Kenworthy, and R. M. Barton. 2002. "Profiles of Everyday Executive Function in Acquired and Developmental Disorders." *Child Neuropsychology: A Journal on Normal and Abnormal Development in Childhood and Adolescence* 8 (2): 121–37.

Girault, Jessica B., and Joseph Piven. 2020. "The Neurodevelopment of Autism from Infancy Through Toddlerhood." *Neuroimaging Clinics of North America* 30 (1): 97–114.

Goodman, Zachary T., Sierra A. Bainter, Salome Kornfeld, Catie Chang, Jason S. Nomi, and Lucina Q. Uddin. 2021. "Whole-Brain Functional Dynamics Track Depressive Symptom Severity." *Cerebral Cortex* 31 (11): 4867.

Grayson, David S., and Damien A. Fair. 2017. "Development of Large-Scale Functional Networks from Birth to Adulthood: A Guide to the Neuroimaging Literature." *NeuroImage* 160 (October):15–31.

Gu, Hong, Kurt P. Schulz, Jin Fan, and Yihong Yang. 2020. "Temporal Dynamics of Functional Brain States Underlie Cognitive Performance." *Cerebral Cortex* 31 (4): 2125–38.

Gusnard, D. A., E. Akbudak, G. L. Shulman, and M. E. Raichle. 2001. "Medial Prefrontal Cortex and Self-Referential Mental Activity: Relation to a Default Mode of Brain Function." *Proceedings of the National Academy of Sciences of the United States of America* 98 (7): 4259–64.

Guthrie, Whitney, Kate Wallis, Amanda Bennett, Elizabeth Brooks, Jesse Dudley, Marsha Gerdes, Juhi Pandey, Susan E. Levy, Robert T. Schultz, and Judith S. Miller. 2019.

"Accuracy of Autism Screening in a Large Pediatric Network." *Pediatrics* 144 (4). https://doi.org/10.1542/peds.2018-3963.

Harlalka, Vatika, Raju S. Bapi, P. K. Vinod, and Dipanjan Roy. 2019. "Atypical Flexibility in Dynamic Functional Connectivity Quantifies the Severity in Autism Spectrum Disorder." *Frontiers in Human Neuroscience* 13 (February):6.

Harrop, Clare, Helen McConachie, Richard Emsley, Kathy Leadbitter, Jonathan Green, and PACT Consortium. 2014. "Restricted and Repetitive Behaviors in Autism Spectrum Disorders and Typical Development: Cross-Sectional and Longitudinal Comparisons." *Journal of Autism and Developmental Disorders* 44 (5): 1207–19.

Hazlett, Heather Cody, Hongbin Gu, Robert C. McKinstry, Dennis W. W. Shaw, Kelly N. Botteron, Stephen R. Dager, Martin Styner, et al. 2012. "Brain Volume Findings in 6-Month-Old Infants at High Familial Risk for Autism." *The American Journal of Psychiatry* 169 (6): 601–8.

Hazlett, Heather Cody, Hongbin Gu, Brent C. Munsell, Sun Hyung Kim, Martin Styner, Jason J. Wolff, Jed T. Elison, et al. 2017. "Early Brain Development in Infants at High Risk for Autism Spectrum Disorder." *Nature* 542 (7641): 348–51.

He, Changchun, Yanchi Chen, Taorong Jian, Heng Chen, Xiaonan Guo, Jia Wang, Lijie Wu, Huafu Chen, and Xujun Duan. 2018. "Dynamic Functional Connectivity Analysis Reveals Decreased Variability of the Default-Mode Network in Developing Autistic Brain." *Autism Research: Official Journal of the International Society for Autism Research* 11 (11): 1479–93.

Hernandez, Leanna M., Jeffrey D. Rudie, Shulamite A. Green, Susan Bookheimer, and Mirella Dapretto. 2015. "Neural Signatures of Autism Spectrum Disorders: Insights into Brain Network Dynamics." *Neuropsychopharmacology: Official Publication of the American College of Neuropsychopharmacology* 40 (1): 171–89.

Hull, Jocelyn V., Lisa B. Dokovna, Zachary J. Jacokes, Carinna M. Torgerson, Andrei Irimia, and John Darrell Van Horn. 2016. "Resting-State Functional Connectivity in Autism Spectrum Disorders: A Review." *Frontiers in Psychiatry / Frontiers Research Foundation* 7:205.

Hutchison, R. M., T. Womelsdorf, E. A. Allen, P. A. Bandettini, V. D. Calhoun, M. Corbetta, S. Della Penna, et al. 2013. "Dynamic Functional Connectivity: Promise, Issues, and Interpretations." *NeuroImage* 80 (October). https://doi.org/10.1016/j.neuroimage.2013.05.079.

"Indices of Repetitive Behaviour Are Correlated with Patterns of Intrinsic Functional Connectivity in Youth with Autism Spectrum Disorder." 2018. *Brain Research* 1685 (April):79–90.

Insel, Thomas R., and Philip S. Wang. 2010. "Rethinking Mental Illness." *JAMA: The Journal of the American Medical Association* 303 (19): 1970–71.

Jarquin, Vanessa G., Lisa D. Wiggins, Laura A. Schieve, and Kim Van Naarden-Braun. 2011. "Racial Disparities in Community Identification of Autism Spectrum Disorders over Time; Metropolitan Atlanta, Georgia, 2000-2006." *Journal of Developmental and Behavioral Pediatrics: JDBP* 32 (3): 179–87.

Jiujias, Marina, Elizabeth Kelley, and Layla Hall. 2017. “Restricted, Repetitive Behaviors in Autism Spectrum Disorder and Obsessive--Compulsive Disorder: A Comparative Review.” *Child Psychiatry and Human Development* 48 (6): 944–59.

Jones, Jonathan S., Alicja Monaghan, Amelia Leyland-Craggs, CALM Team, and Duncan E. Astle. 2023. “Testing the Triple Network Model of Psychopathology in a Transdiagnostic Neurodevelopmental Cohort.” *NeuroImage. Clinical* 40 (November):103539.

Kenworthy, Lauren, Laura Gutermuth Anthony, Daniel Q. Naiman, Lynn Cannon, Meagan C. Wills, Caroline Luong-Tran, Monica Adler Werner, et al. 2014. “Randomized Controlled Effectiveness Trial of Executive Function Intervention for Children on the Autism Spectrum.” *Journal of Child Psychology and Psychiatry, and Allied Disciplines* 55 (4): 374–83.

Keunen, Kristin, Serena J. Counsell, and Manon J. N. L. Benders. 2017. “The Emergence of Functional Architecture during Early Brain Development.” *NeuroImage* 160 (October):2–14.

Krönke, Klaus-Martin, Max Wolff, Yiquan Shi, Anja Kräplin, Michael N. Smolka, Gerhard Bühringer, and Thomas Goschke. 2020. “Functional Connectivity in a Triple-Network Saliency Model Is Associated with Real-Life Self-Control.” *Neuropsychologia* 149 (December):107667.

Kupis, Lauren, Zachary T. Goodman, Leigha Kircher, Celia Romero, Bryce Dirks, Catie Chang, Jason S. Nomi, and Lucina Q. Uddin. 2021. “Altered Patterns of Brain Dynamics Linked with Body Mass Index in Youth with Autism.” *Autism Research: Official Journal of the International Society for Autism Research* 14 (5): 873–86.

Kupis, Lauren, Zachary T. Goodman, Salome Kornfeld, Stephanie Hoang, Celia Romero, Bryce Dirks, Joseph Dehoney, et al. 2021. “Brain Dynamics Underlying Cognitive Flexibility Across the Lifespan.” *Cerebral Cortex* 31 (11): 5263–74.

Kupis, Lauren, Celia Romero, Bryce Dirks, Stephanie Hoang, Meaghan V. Parlade, Amy L. Beaumont, Sandra M. Cardona, et al. 2020. “Evoked and Intrinsic Brain Network Dynamics in Children with Autism Spectrum Disorder.” *Neuroimage: Clinical*.

Kupis, Lauren, Celia Romero, Bryce Dirks, Stephanie Hoang, Meaghan V. Parladé, Amy L. Beaumont, Sandra M. Cardona, et al. 2020. “Evoked and Intrinsic Brain Network Dynamics in Children with Autism Spectrum Disorder.” *NeuroImage. Clinical* 28 (August):102396.

Lacy, N. de, D. Doherty, B. H. King, S. Rachakonda, and V. D. Calhoun. 2017. “Disruption to Control Network Function Correlates with Altered Dynamic Connectivity in the Wider Autism Spectrum.” *NeuroImage. Clinical* 15 (May):513–24.

Lage, Claudia, Eleanor S. Smith, and Rebecca P. Lawson. 2024. “A Meta-Analysis of Cognitive Flexibility in Autism Spectrum Disorder.” *Neuroscience and Biobehavioral Reviews* 157 (February):105511.

Lau, Way K. W., Mei-Kei Leung, and Benson W. M. Lau. 2019. “Resting-State Abnormalities in Autism Spectrum Disorders: A Meta-Analysis.” *Scientific Reports* 9 (1): 1–8.

Lee, M. H., C. D. Smyser, and J. S. Shimony. 2013. “Resting-State fMRI: A Review of Methods and Clinical Applications.” *AJNR. American Journal of Neuroradiology* 34 (10): 1866–72.

Lewis, Mark, and Soo-Jeong Kim. 2009. “The Pathophysiology of Restricted Repetitive

Behavior." *Journal of Neurodevelopmental Disorders* 1 (2): 114–32.
Lin, Pan, Yong Yang, Jorge Jovicich, Nicola De Pisapia, Xiang Wang, Chun S. Zuo, and James Jonathan Levitt. 2016. "Static and Dynamic Posterior Cingulate Cortex Nodal Topology of Default Mode Network Predicts Attention Task Performance." *Brain Imaging and Behavior* 10 (1): 212–25.
Lin, Wanyun, Yiong Huak Chan, Jennifer S. H. Kiing, Tammy S. H. Lim, Shang Chee Chong, Ying Qi Kang, Ramkumar Aishworiya, Kalyani Vijayakumar Mulay, and Mae Yue Tan. 2023. "Restricted and Repetitive Behaviors and Association with Cognition and Adaptive Functioning in Children with Autism Spectrum Disorder in Singapore." *Frontiers in Psychiatry / Frontiers Research Foundation* 14 (November):1249071.
Liu, Janelle, Tawny Tsang, Lisa Jackson, Carolyn Ponting, Shafali S. Jeste, Susan Y. Bookheimer, and Mirella Dapretto. 2019. "Altered Lateralization of Dorsal Language Tracts in 6-Week-Old Infants at Risk for Autism." *Developmental Science* 22 (3): e12768.
Liu, Wen-Ching, Judy F. Flax, Kevin G. Guise, Vishad Sukul, and April A. Benasich. 2008. "Functional Connectivity of the Sensorimotor Area in Naturally Sleeping Infants." *Brain Research* 1223 (August):42–49.
Liu, Xiao, Catie Chang, and Jeff H. Duyn. 2013. "Decomposition of Spontaneous Brain Activity into Distinct fMRI Co-Activation Patterns." *Frontiers in Systems Neuroscience*. https://doi.org/10.3389/fnsys.2013.00101.
Liu, Xiao, and Jeff H. Duyn. 2013. "Time-Varying Functional Network Information Extracted from Brief Instances of Spontaneous Brain Activity." *Proceedings of the National Academy of Sciences of the United States of America* 110 (11): 4392–97.
Liu, Xiao, Nanyin Zhang, Catie Chang, and Jeff H. Duyn. 2018. "Co-Activation Patterns in Resting-State fMRI Signals." *NeuroImage* 180 (Pt B): 485.
Lombardo, Michael V., Karen Pierce, Lisa Eyler, Cindy Carter Barnes, Clelia Ahrens-Barbeau, Stephanie Solso, Kathleen Campbell, and Eric Courchesne. 2015. "Different Functional Neural Substrates for Good and Poor Language Outcome in Autism." *Neuron* 86 (2): 567.
Lopez, B. R., A. J. Lincoln, S. Ozonoff, and Z. Lai. 2005. "Examining the Relationship between Executive Functions and Restricted, Repetitive Symptoms of Autistic Disorder." *Journal of Autism and Developmental Disorders* 35 (4): 445–60.
Lord, C., S. Risi, L. Lambrecht, E. H. Cook Jr, B. L. Leventhal, P. C. DiLavore, A. Pickles, and M. Rutter. 2000. "The Autism Diagnostic Observation Schedule-Generic: A Standard Measure of Social and Communication Deficits Associated with the Spectrum of Autism." *Journal of Autism and Developmental Disorders* 30 (3): 205–23.
Lord, C., M. Rutter, P. DiLavore, S. Risi, K. Gotham, and S. Bishop. 2012. "Autism Diagnostic Observation Schedule--2nd Edition (ADOS-2)." *Los Angeles, CA: Western Psychological Corporation*.
Lurie, Daniel J., Daniel Kessler, Danielle S. Bassett, Richard F. Betzel, Michael Breakspear, Shella Kheilholz, Aaron Kucyi, et al. 2020. "Questions and Controversies in the Study of Time-Varying Functional Connectivity in Resting fMRI." *Network Neuroscience* 4 (1): 30.
Maenner, Matthew J., Zachary Warren, Ashley Robinson Williams, Esther Amoakohene, Amanda V. Bakian, Deborah A. Bilder, Maureen S. Durkin, et al. 2023. "Prevalence and

Characteristics of Autism Spectrum Disorder Among Children Aged 8 Years - Autism and Developmental Disabilities Monitoring Network, 11 Sites, United States, 2020." *Morbidity and Mortality Weekly Report. Surveillance Summaries* 72 (2): 1–14.

Marek, Scott, and Nico U. F. Dosenbach. 2018. "The Frontoparietal Network: Function, Electrophysiology, and Importance of Individual Precision Mapping." *Dialogues in Clinical Neuroscience* 20 (2): 133–40.

Marshall, Emily, Jason S. Nomi, Bryce Dirks, Celia Romero, Lauren Kupis, Catie Chang, and Lucina Q. Uddin. 2020a. "Co-Activation Pattern Analysis Reveals Altered Salience Network Dynamics in Children with Autism Spectrum Disorder." *Network Neuroscience*, August, 1–16.

———. 2020b. "Coactivation Pattern Analysis Reveals Altered Salience Network Dynamics in Children with Autism Spectrum Disorder." *Network Neuroscience* 4 (4): 1219.

Masi, Anne, Marilena M. DeMayo, Nicholas Glozier, and Adam J. Guastella. 2017. "An Overview of Autism Spectrum Disorder, Heterogeneity and Treatment Options." *Neuroscience Bulletin* 33 (2): 183–93.

Mason, David, Helen McConachie, Deborah Garland, Alex Petrou, Jacqui Rodgers, and Jeremy R. Parr. 2018. "Predictors of Quality of Life for Autistic Adults." *Autism Research: Official Journal of the International Society for Autism Research* 11 (8): 1138–47.

Matthew Hutchison, R., and J. Bruce Morton. 2015. "Tracking the Brain's Functional Coupling Dynamics over Development." *The Journal of Neuroscience: The Official Journal of the Society for Neuroscience* 35 (17): 6849–59.

Ma, Xueling, Xiushuang Wu, and Yuan Shi. 2020. "Changes of Dynamic Functional Connectivity Associated With Maturity in Late Preterm Infants." *Frontiers in Pediatrics* 8 (July):412.

McKinnon, Claire J., Adam T. Eggebrecht, Alexandre Todorov, Jason J. Wolff, Jed T. Elison, Chloe M. Adams, Abraham Z. Snyder, et al. 2019. "Restricted and Repetitive Behavior and Brain Functional Connectivity in Infants at Risk for Developing Autism Spectrum Disorder." *Biological Psychiatry. Cognitive Neuroscience and Neuroimaging* 4 (1): 50–61.

Menon, Bindu. 2019. "Towards a New Model of Understanding - The Triple Network, Psychopathology and the Structure of the Mind." *Medical Hypotheses* 133 (December):109385.

Menon, V. 2015a. "Large-Scale Functional Brain Organization." In *Brain Mapping*, edited by Arthur W. Toga, 449–59. Waltham: Academic Press.

———. 2015b. "Salience Network." In *Brain Mapping*, edited by Arthur W. Toga, 597–611. Waltham: Academic Press.

Menon, Vinod. 2011. "Large-Scale Brain Networks and Psychopathology: A Unifying Triple Network Model." *Trends in Cognitive Sciences* 15 (10): 483–506.

Menon, Vinod, and Mark D'Esposito. 2022. "The Role of PFC Networks in Cognitive Control and Executive Function." *Neuropsychopharmacology: Official Publication of the American College of Neuropsychopharmacology* 47 (1): 90–103.

Menon, Vinod, and Lucina Q. Uddin. 2010. "Saliency, Switching, Attention and Control: A Network Model of Insula Function." *Brain Structure & Function* 214 (5-6): 655–67.

Mitra, Anish, Abraham Z. Snyder, Enzo Tagliazucchi, Helmut Laufs, Jed Elison, Robert W. Emerson, Mark D. Shen, et al. 2017. "Resting-State fMRI in Sleeping Infants More Closely Resembles Adult Sleep than Adult Wakefulness." *PloS One* 12 (11). https://doi.org/10.1371/journal.pone.0188122.

Molnar-Szakacs, I., L. Kupis, and L. Q. Uddin. 2021. "Neuroimaging Markers of Risk and Pathways to Resilience in Autism Spectrum Disorder." *Biological Psychiatry. Cognitive Neuroscience and Neuroimaging* 6 (2). https://doi.org/10.1016/j.bpsc.2020.06.017.

Molnar-Szakacs, Istvan, Lauren Kupis, and Lucina Q. Uddin. 2020. "Neuroimaging Markers of Risk and Pathways to Resilience in Autism Spectrum Disorders." *Biological Psychiatry: Cognitive Neuroscience and Neuroimaging*. https://www.sciencedirect.com/science/article/pii/S2451902220301725?casa_token=QEwpCirLY24AAAAA:9HdTjKpQybtvdSVpMfFKQhzNMWJNPKet2EklNxfChipK82DaY7YDh9NwM69O7gfKFg4NVF1_xP14.

Muhle, Rebecca A., Hannah E. Reed, Katharine A. Stratigos, and Jeremy Veenstra-VanderWeele. 2018. "The Emerging Clinical Neuroscience of Autism Spectrum Disorder: A Review." *JAMA Psychiatry* 75 (5): 514–23.

Mullen, Eileen M. 1995. *Mullen Scales of Early Learning Manual*.

Neufeld, J., R. Kuja-Halkola, K. Mevel, É. Cauvet, P. Fransson, and S. Bölte. 2017. "Alterations in Resting State Connectivity along the Autism Trait Continuum: A Twin Study." *Molecular Psychiatry* 23 (7): 1659–65.

Nomi, Jason S., and Lucina Q. Uddin. 2015. "Developmental Changes in Large-Scale Network Connectivity in Autism." *NeuroImage. Clinical* 7 (March):732–41.

Nomi, Jason S., Shruti Gopal Vij, Dina R. Dajani, Rosa Steimke, Eswar Damaraju, Srinivas Rachakonda, Vince D. Calhoun, and Lucina Q. Uddin. 2017. "Chronnectomic Patterns and Neural Flexibility Underlie Executive Function." *NeuroImage* 147 (February):861–71.

Okoye, Chiugo, Chidi M. Obialo-Ibeawuchi, Omobolanle A. Obajeun, Sarosh Sarwar, Christine Tawfik, Madeeha Subhan Waleed, Asad Ullah Wasim, Iman Mohamoud, Adebola Y. Afolayan, and Rheiner N. Mbaezue. 2023. "Early Diagnosis of Autism Spectrum Disorder: A Review and Analysis of the Risks and Benefits." *Cureus* 15 (8): e43226.

Ozonoff, Sally, Ana-Maria Iosif, Ms Fam Baguio, Ian C. Cook, Ms Monique Moore Hill, Ted Hutman, Sally J. Rogers, et al. 2010. "A Prospective Study of the Emergence of Early Behavioral Signs of Autism." *Journal of the American Academy of Child and Adolescent Psychiatry* 49 (3): 256.

Pierce, Karen. 2011. "Early Functional Brain Development in Autism and the Promise of Sleep fMRI." *Brain Research* 1380 (March):162–74.

Pierce, Karen, Vahid Gazestani, Elizabeth Bacon, Eric Courchesne, Amanda Cheng, Cynthia Carter Barnes, Srinivasa Nalabolu, et al. 2021. "Get SET Early to Identify and Treatment Refer Autism Spectrum Disorder at 1 Year and Discover Factors That Influence Early Diagnosis." *The Journal of Pediatrics* 236 (September):179–88.

Pierce, Karen, Vahid H. Gazestani, Elizabeth Bacon, Cynthia Carter Barnes, Debra Cha, Srinivasa Nalabolu, Linda Lopez, Adrienne Moore, Sunny Pence-Stophaeros, and Eric Courchesne. 2019. "Evaluation of the Diagnostic Stability of the Early Autism Spectrum

Disorder Phenotype in the General Population Starting at 12 Months." *JAMA Pediatrics* 173 (6): 578–87.
Plitt, Mark, Kelly Anne Barnes, Gregory L. Wallace, Lauren Kenworthy, and Alex Martin. 2015. "Resting-State Functional Connectivity Predicts Longitudinal Change in Autistic Traits and Adaptive Functioning in Autism." *Proceedings of the National Academy of Sciences of the United States of America* 112 (48): E6699–6706.
Pote, Inês, Siying Wang, Vaheshta Sethna, Anna Blasi, Eileen Daly, Maria Kuklisova-Murgasova, Sarah Lloyd-Fox, et al. 2019. "Familial Risk of Autism Alters Subcortical and Cerebellar Brain Anatomy in Infants and Predicts the Emergence of Repetitive Behaviors in Early Childhood." *Autism Research: Official Journal of the International Society for Autism Research* 12 (4): 614–27.
Power, Jonathan D., Kelly A. Barnes, Abraham Z. Snyder, Bradley L. Schlaggar, and Steven E. Petersen. 2012. "Spurious but Systematic Correlations in Functional Connectivity MRI Networks Arise from Subject Motion." *NeuroImage* 59 (3): 2142–54.
Preti, Maria Giulia, Thomas Aw Bolton, and Dimitri Van De Ville. 2017. "The Dynamic Functional Connectome: State-of-the-Art and Perspectives." *NeuroImage* 160 (October):41–54.
Rabany, Liron, Sophy Brocke, Vince D. Calhoun, Brian Pittman, Silvia Corbera, Bruce E. Wexler, Morris D. Bell, Kevin Pelphrey, Godfrey D. Pearlson, and Michal Assaf. 2019. "Dynamic Functional Connectivity in Schizophrenia and Autism Spectrum Disorder: Convergence, Divergence and Classification." *NeuroImage. Clinical* 24 (August):101966.
Raichle, Marcus E. 2015. "The Brain's Default Mode Network." *Annual Review of Neuroscience* 38 (July):433–47.
Rashid, Barnaly, Laura M. E. Blanken, Ryan L. Muetzel, Robyn Miller, Eswar Damaraju, Mohammad R. Arbabshirani, Erik B. Erhardt, et al. 2018. "Connectivity Dynamics in Typical Development and Its Relationship to Autistic Traits and Autism Spectrum Disorder." *Human Brain Mapping* 39 (8): 3127–42.
Redcay, Elizabeth, and Eric Courchesne. 2008. "Deviant Functional Magnetic Resonance Imaging Patterns of Brain Activity to Speech in 2-3-Year-Old Children with Autism Spectrum Disorder." *Biological Psychiatry* 64 (7): 589–98.
"Relations among Restricted and Repetitive Behaviors, Anxiety and Sensory Features in Children with Autism Spectrum Disorders." 2014. *Research in Autism Spectrum Disorders* 8 (2): 82–92.
"Reproducible Coactivation Patterns of Functional Brain Networks Reveal the Aberrant Dynamic State Transition in Schizophrenia." 2021. *NeuroImage* 237 (August):118193.
Richler, Jennifer, Marisela Huerta, Somer L. Bishop, and Catherine Lord. 2010a. "Developmental Trajectories of Restricted and Repetitive Behaviors and Interests in Children with Autism Spectrum Disorders." *Development and Psychopathology* 22 (1): 55.
———. 2010b. "Developmental Trajectories of Restricted and Repetitive Behaviors and Interests in Children with Autism Spectrum Disorders." *Development and Psychopathology* 22 (1): 55–69.
Rudie, Jeffrey David, and Mirella Dapretto. 2013. "Convergent Evidence of Brain

Overconnectivity in Children with Autism?" *Cell Reports* 5 (3): 565–66.
Rué-Queralt, Joan, Angus Stevner, Enzo Tagliazucchi, Helmut Laufs, Morten L. Kringelbach, Gustavo Deco, and Selen Atasoy. 2021. "Decoding Brain States on the Intrinsic Manifold of Human Brain Dynamics across Wakefulness and Sleep." *Communications Biology* 4 (1): 1–11.
Sadeghi, Saeid, and Hamid Reza Pouretemad. 2022. "Executive Functions Predict Restricted and Repetitive Behaviors in Toddlers under 36 Months Old with Autism Spectrum Disorder." *Infant Behavior & Development* 67 (May):101721.
Schaefer, Alexander, Ru Kong, Evan M. Gordon, Timothy O. Laumann, Xi-Nian Zuo, Avram J. Holmes, Simon B. Eickhoff, and B. T. Thomas Yeo. 2018. "Local-Global Parcellation of the Human Cerebral Cortex from Intrinsic Functional Connectivity MRI." *Cerebral Cortex* 28 (9): 3095–3114.
Scheinost, Dustin, Joseph Chang, Emma Brennan-Wydra, Cheryl Lacadie, R. Todd Constable, Katarzyna Chawarska, and Laura R. Ment. 2022. "Developmental Trajectories of the Default Mode, Executive Control, and Salience Networks from the Third Trimester through the Newborn Period." *bioRxiv*. https://doi.org/10.1101/2022.09.27.509687.
Schulz, Samantha E., and Ryan A. Stevenson. 2020. "Differentiating between Sensory Sensitivity and Sensory Reactivity in Relation to Restricted Interests and Repetitive Behaviours." *Autism: The International Journal of Research and Practice* 24 (1): 121–34.
Shaw, Saurabh Bhaskar, Margaret C. McKinnon, Jennifer Heisz, and Suzanna Becker. 2021. "Dynamic Task-Linked Switching between Brain Networks - A Tri-Network Perspective." *Brain and Cognition* 151 (July):105725.
Shen, Mark D., Christine W. Nordahl, Gregory S. Young, Sandra L. Wootton-Gorges, Aaron Lee, Sarah E. Liston, Kayla R. Harrington, Sally Ozonoff, and David G. Amaral. 2013. "Early Brain Enlargement and Elevated Extra-Axial Fluid in Infants Who Develop Autism Spectrum Disorder." *Brain: A Journal of Neurology* 136 (Pt 9): 2825–35.
Shi, Feng, Pew-Thian Yap, Guorong Wu, Hongjun Jia, John H. Gilmore, Weili Lin, and Dinggang Shen. 2011. "Infant Brain Atlases from Neonates to 1- and 2-Year-Olds." *PloS One* 6 (4): e18746.
Smitha, K. A., K. Akhil Raja, K. M. Arun, P. G. Rajesh, Bejoy Thomas, T. R. Kapilamoorthy, and Chandrasekharan Kesavadas. 2017. "Resting State fMRI: A Review on Methods in Resting State Connectivity Analysis and Resting State Networks." *The Neuroradiology Journal* 30 (4): 305–17.
Sparrow, Sara S. 2005. *Vineland II: Vineland Adaptative Behavior Scales : Survey Forms Manual : A Revision of Hte Vineland Social Maturity Scale by Edgar A. Doll*.
Sparrow, Sara S., Domenic Cicchetti, and David A. Balla. 2012. "Vineland Adaptive Behavior Scales, Second Edition." *PsycTESTS Dataset*. https://doi.org/10.1037/t15164-000.
Sridharan, Devarajan, Daniel J. Levitin, and Vinod Menon. 2008. "A Critical Role for the Right Fronto-Insular Cortex in Switching between Central-Executive and Default-Mode Networks." *Proceedings of the National Academy of Sciences of the United States of America* 105 (34): 12569–74.
Stevner, A. B. A., D. Vidaurre, J. Cabral, K. Rapuano, S. F. V. Nielsen, E. Tagliazucchi, H.

Laufs, et al. 2019. “Discovery of Key Whole-Brain Transitions and Dynamics during Human Wakefulness and Non-REM Sleep.” *Nature Communications* 10 (1): 1–14.
Supekar, Kaustubh, Weidong Cai, Rajeev Krishnadas, Lena Palaniyappan, and Vinod Menon. 2019. “Dysregulated Brain Dynamics in a Triple-Network Saliency Model of Schizophrenia and Its Relation to Psychosis.” *Biological Psychiatry* 85 (1): 60–69.
Supekar, Kaustubh, Srikanth Ryali, Percy Mistry, and Vinod Menon. 2021. “Aberrant Dynamics of Cognitive Control and Motor Circuits Predict Distinct Restricted and Repetitive Behaviors in Children with Autism.” *Nature Communications* 12 (1): 3537.
Supekar, K., L. Q. Uddin, A. Khouzam, J. Phillips, W. D. Gaillard, L. E. Kenworthy, B. E. Yerys, C. J. Vaidya, and V. Menon. 2013. “Brain Hyperconnectivity in Children with Autism and Its Links to Social Deficits.” *Cell Reports* 5 (3). https://doi.org/10.1016/j.celrep.2013.10.001.
Thelen, Esther. 1980. “Determinants of Amounts of Stereotyped Behavior in Normal Human Infants.” *Ethology and Sociobiology* 1 (2): 141–50.
Thomason, Moriah E., Maya T. Dassanayake, Stephen Shen, Yashwanth Katkuri, Mitchell Alexis, Amy L. Anderson, Lami Yeo, et al. 2013. “Cross-Hemispheric Functional Connectivity in the Human Fetal Brain.” *Science Translational Medicine* 5 (173): 173ra24.
Tierney, Adrienne L., and Charles A. Nelson 3rd. 2009a. “Brain Development and the Role of Experience in the Early Years.” *Zero to Three* 30 (2): 9–13.
———. 2009b. “Brain Development and the Role of Experience in the Early Years.” *Zero to Three* 30 (2): 9–13.
Traynor, Jenna M., and Geoffrey B. C. Hall. 2015a. “Structural and Functional Neuroimaging of Restricted and Repetitive Behavior in Autism Spectrum Disorder.” *Journal of Intellectual Disability-Diagnosis and Treatment* 3 (1): 21–34.
———. 2015b. “Structural and Functional Neuroimaging of Restricted and Repetitive Behavior in Autism Spectrum Disorder.” *Journal of Intellectual Disability - Diagnosis and Treatment* 3 (1): 21–34.
Troyb, Eva, Kelley Knoch, Lauren Herlihy, Michael C. Stevens, Chi-Ming Chen, Marianne Barton, Kimberli Treadwell, and Deborah Fein. 2016. “Restricted and Repetitive Behaviors as Predictors of Outcome in Autism Spectrum Disorders.” *Journal of Autism and Developmental Disorders* 46 (4): 1282.
Tsang, Tawny, Shulamite A. Green, Janelle Liu, Katherine Lawrence, Shafali Jeste, Susan Y. Bookheimer, and Mirella Dapretto. 2024. “Salience Network Connectivity Is Altered in 6-Week-Old Infants at Heightened Likelihood for Developing Autism.” *Communications Biology* 7 (1): 485.
Uddin, L. Q., and K. H. Karlsgodt. 2018. “Future Directions for Examination of Brain Networks in Neurodevelopmental Disorders.” *Journal of Clinical Child and Adolescent Psychology: The Official Journal for the Society of Clinical Child and Adolescent Psychology, American Psychological Association, Division 53* 47 (3). https://doi.org/10.1080/15374416.2018.1443461.
Uddin, L. Q., K. S. Supekar, S. Ryali, and V. Menon. 2011. “Dynamic Reconfiguration of Structural and Functional Connectivity Across Core Neurocognitive Brain Networks with

Development." *Journal of Neuroscience*. https://doi.org/10.1523/jneurosci.4465-11.2011.
Uddin, Lucina Q. 2015. "Salience Processing and Insular Cortical Function and Dysfunction." *Nature Reviews. Neuroscience* 16 (1): 55–61.
———. 2021. "Cognitive and Behavioural Flexibility: Neural Mechanisms and Clinical Considerations." *Nature Reviews. Neuroscience* 22 (3): 167–79.
Uddin, Lucina Q., Kaustubh Supekar, Charles J. Lynch, Katherine M. Cheng, Paola Odriozola, Maria E. Barth, Jennifer Phillips, Carl Feinstein, Daniel A. Abrams, and Vinod Menon. 2015a. "Brain State Differentiation and Behavioral Inflexibility in Autism." *Cerebral Cortex* 25 (12): 4740–47.
———. 2015b. "Brain State Differentiation and Behavioral Inflexibility in Autism." *Cerebral Cortex* 25 (12): 4740–47.
Uddin, Lucina Q., Kaustubh Supekar, Charles J. Lynch, Amirah Khouzam, Jennifer Phillips, Carl Feinstein, Srikanth Ryali, and Vinod Menon. 2013a. "Salience Network–Based Classification and Prediction of Symptom Severity in Children With Autism." *JAMA Psychiatry* 70 (8): 869–79.
———. 2013b. "Salience Network-Based Classification and Prediction of Symptom Severity in Children with Autism." *JAMA Psychiatry* 70 (8): 869–79.
Uddin, Lucina Q., Kaustubh Supekar, and Vinod Menon. 2013. "Reconceptualizing Functional Brain Connectivity in Autism from a Developmental Perspective." *Frontiers in Human Neuroscience* 7 (August):53721.
Uddin, Lucina Q., B. T. Thomas Yeo, and R. Nathan Spreng. 2019. "Towards a Universal Taxonomy of Macro-Scale Functional Human Brain Networks." *Brain Topography* 32 (6): 926–42.
Varoquaux, G., Alexandre Gramfort, J. Poline, and B. Thirion. 2010. "Brain Covariance Selection: Better Individual Functional Connectivity Models Using Population Prior." *Advances in Neural Information Processing Systems*, August, 2334–42.
Vivanti, Giacomo, Margot Prior, Katrina Williams, and Cheryl Dissanayake. 2014. "Predictors of Outcomes in Autism Early Intervention: Why Don't We Know More?" *Frontiers in Pediatrics* 2 (June):58.
Wagner, Lauren, Megan Banchik, Nana J. Okada, Nicole McDonald, Shafali S. Jeste, Susan Y. Bookheimer, Shulamite A. Green, and Mirella Dapretto. 2023. "Associations between Thalamocortical Functional Connectivity and Sensory over-Responsivity in Infants at High Likelihood for ASD." *Cerebral Cortex* 33 (12): 8075.
Wang, Kai, Ke Li, and Xiaoyu Niu. 2021. "Altered Functional Connectivity in a Triple-Network Model in Autism With Co-Occurring Attention Deficit Hyperactivity Disorder." *Frontiers in Psychiatry / Frontiers Research Foundation* 12 (December):736755.
Wang, Lei, Yue Qin, Shuhan Yang, Dayong Jin, Yinhu Zhu, Xin Li, Wei Li, Yarong Wang, and Chenwang Jin. 2024. "Posterior Default Mode Network Is Associated with the Social Performance in Male Children with Autism Spectrum Disorder: A Dynamic Causal Modeling Analysis Based on Triple-Network Model." *Human Brain Mapping* 45 (8): e26750.
Watanabe, Takamitsu, and Geraint Rees. 2017. "Brain Network Dynamics in High-Functioning

Individuals with Autism." *Nature Communications* 8 (July):16048.
Wolff, Jason J., Hongbin Gu, Guido Gerig, Jed T. Elison, Martin Styner, Sylvain Gouttard, Kelly N. Botteron, et al. 2012. "Differences in White Matter Fiber Tract Development Present from 6 to 24 Months in Infants with Autism." *The American Journal of Psychiatry* 169 (6): 589–600.
Wolff, Jason J., Suma Jacob, and Jed T. Elison. 2018. "The Journey to Autism: Insights from Neuroimaging Studies of Infants and Toddlers." *Development and Psychopathology* 30 (2): 479–95.
Xiao, Yaqiong, Teresa H. Wen, Lauren Kupis, Lisa T. Eyler, Vani Taluja, Jaden Troxel, Disha Goel, Michael V. Lombardo, Karen Pierce, and Eric Courchesne. 2023. "Atypical Functional Connectivity of Temporal Cortex with Precuneus and Visual Regions May Be an Early-Age Signature of ASD." *Molecular Autism* 14 (1): 1–11.
Xiao, Yaqiong, Teresa Wen, Lauren Kupis, Lisa Eyler, Disha Goel, Michael Lombardo, Karen Pierce, and Eric Courchesne. 2021. "Neural Responses to Affective Speech, Including Motherese, Map onto Clinical and Social Eye Tracking Profiles in ASD Toddlers." *Nature Human Behavior*.
Yang, Tingting, Xiaojuan Dong, and Xu Lei. 2021. "Hard to Initiate Sleep: A New Paradigm for Resting-State fMRI." *Cognitive Neurodynamics* 15 (5): 825–33.
Yao, Zhijun, Bin Hu, Yuanwei Xie, Fang Zheng, Guangyao Liu, Xuejiao Chen, and Weihao Zheng. 2016. "Resting-State Time-Varying Analysis Reveals Aberrant Variations of Functional Connectivity in Autism." *Frontiers in Human Neuroscience* 10 (September):463.
Yue, Xipeng, Ge Zhang, Xiaochen Li, Yu Shen, Wei Wei, Yan Bai, Yu Luo, et al. 2022. "Abnormal Dynamic Functional Network Connectivity in Adults with Autism Spectrum Disorder." *Clinical Neuroradiology* 32 (4): 1087–96.
Zhang, Han, Dinggang Shen, and Weili Lin. 2019. "Resting-State Functional MRI Studies on Infant Brains: A Decade of Gap-Filling Efforts." *NeuroImage* 185 (January):664–84.
Zhang, Han, Weiyan Yin, Weili Lin, and Dinggang Shen. 2017. "Early Brain Functional Segregation and Integration Predict Later Cognitive Performance." In *Connectomics in NeuroImaging*, 116–24. Springer International Publishing.

CHAPTER 5:

CONCLUSION

Cognitive flexibility or the ability to mentally shift in response to change is a crucial ability for lifelong success. FMRI and brain network dynamics have been used to advance our understanding of the neural underpinnings of healthy and abnormal brain functioning and brain-behavior relationships. Autism spectrum disorder (ASD) in particular is a heterogeneous condition with core difficulties with cognitive and behavioral flexibility. This dissertation advances our understanding of how brain network dynamics underlie healthy cognitive flexibility across the lifespan, development of cognitive inflexibility in autism spectrum disorder, and early predictors of autism and emerging cognitive and behavioral inflexibility. The identification of neural signatures underlying cognitive flexibility, biomarkers of ASD, and behavioral difficulties may help increase early diagnosis and intervention and ultimately improve life outcomes in typical and atypical populations.

In chapter 2, resting-state intrinsic brain network dynamics was examined across the lifespan in a healthy sample. Further, brain network dynamics were examined in relation to flexible cognition and behavior across the lifespan. By investigating the co-activation patterns of the salience (SN), default mode (DMN), and central executive (CEN) networks, we found brain dynamics of coupled states of these networks (e.g., SN co-activated with CEN) had U-shaped trajectories across the lifespan. Next, between-network dynamics of a state consisting of the CEN and DMN co-activation moderated the relationship between aging and cognitive flexibility. Additionally, brain state transitions also moderated the relationship between aging and cognitive

flexibility. This is one of the first studies to investigate brain network dynamic changes across the lifespan and the relationship between brain network flexibility and cognitive and behavioral flexibility. This study revealed the importance of discovering brain-biomarkers for preventative measures and intervention for later developing cognitive flexibility problems, and highlighted the dynamic interactions among the SN, DMN, and CEN as promising biomarkers for cognitive and behavioral flexibility across the lifespan.

Chapter 3 extended these findings by investigating brain network dynamics in children with and without autism spectrum disorder (ASD; ages 7-12 years) during task and rest fMRI states. This study used the same co-activation pattern analysis method and further investigated the SN, DMN, and CEN dynamic interactions during a flexibility task and during a resting-state MRI scan. ASD children had altered brain dynamics in later stages of the flexibility task. During the fourth run of the task, ASD children had a greater frequency of occurrence of a state with CEN, and dwelled less in a state with co-active SN and CEN. Across both groups, a greater DMN dwell time was associated with stronger social skills. These findings support the importance of the SN, CEN, and DMN in development, flexible cognition and behavior, and in ASD. Additionally, these findings suggest atypical between-network coordination may underlie neural compensation in children with ASD but may be exacerbated by long durations of a task. Additionally, DMN dynamics may be important for social cognition and dysfunction in both ASD and healthy development. Taken together, these brain network dynamic studies in a healthy lifespan sample and in youth with and without ASD indicate the importance of brain network interactions underlying brain development and flexible and inflexible cognition. These results

also highlight the importance of the core neural networks (SN, DMN, and CEN) in atypical and typical development and predicting brain-behaviors.

The study in chapter 4 then investigated early brain network dynamics in toddlers later diagnosed with and without ASD and their relationship to flexible behaviors including restricted and repetitive behaviors and adaptive behaviors. ASD toddlers dwelled longer in a state consisting of co-activation of all three neural networks (SN, CEN, and DMN) compared with non-ASD toddlers. Next, various brain states consisting of the core neural networks were associated with RRBs and adaptive behaviors across both ASD and non ASD toddlers. The findings extended the findings in the previous studies such that states with SN and CEN co-activation were associated with positive flexible behaviors while states with DMN co-activation were associated with negative flexible behaviors. This is one of the first studies to investigate brain network dynamic interactions in toddlers with and without ASD, and how early brain network interactions may be predictive of later behaviors. This study is one of the first to show the importance of the core neural networks (SN, DMN, CEN) in early brain development and autism, and emerging behaviors.

Limitations and Future Directions

All of the studies discussed included a novel brain dynamic method and a fMRI scan at a single time point for each participant. The novel brain dynamic methods, although informative, need to be further assessed methodologically. Additionally, there are always concerns with head motion with time-varying methods which also limits the sample and may pose as a confound. Future studies need to test the time-varying methods further especially in neurodiverse

populations where head motion is a greater concern. Additionally, longitudinal studies of brain development are needed to elucidate causal interactions between brain network dynamics and behavior along the lifespan. Repeat fMRI scans may be useful to elucidate early neural signatures of autism, critical time periods for behavior and development, and the extent that brain networks can alter and change across the lifespan. Another limitation was the inconsistency of the timing of the fMRI scans especially in the toddlers investigated. Studies have begun to investigate and observe brain differences in those later diagnosed with autism as early as 6 weeks. Future work ideally would scan infants and toddlers at the same time points and repeatedly throughout the first three years of life. As the brain develops rapidly in the first years of life, consistent, repeat scans would potentially elucidate the neural mechanisms underlying autism and better identify early biomarkers for later developing autism and behavior outcomes. Another limitation of the studies was sample size making the studies not as generalizable to the population. Head motion and completion of the fMRI scan poses a challenge when scanning children and neurodiverse populations. It will be important for future studies to consider novel methods and strategies to include participants of varying abilities and across the lifespan.

Concluding Remarks

The ability to flexibly respond to a constantly changing environment is paramount to survival and positive life outcomes. Understanding the brain network dynamics underlying cognitive and behavioral flexibility and inflexibility is important to inform development and aging in typical and atypical populations. This thesis used functional magnetic resonance imaging and time-varying methods to investigate how three core neural networks dynamically

interact across the lifespan, and how they support cognitive flexibility during sleep, rest, and task states, and how they are altered in autism spectrum disorder during the first years of life. We found brain network interactions among the salience network (SN), default mode network (DMN), and central executive networks (CEN) supported cognitive flexibility across the lifespan, and are altered in children with autism, and in toddlers later diagnosed with autism compared with neurotypical toddlers. This dissertation lays the groundwork for further investigating the SN, DMN, and CEN dynamic interactions as a biomarker for autism spectrum disorder and underlying cognitive flexibility across the lifespan. The work in this dissertation also supports the development of targeted individualized treatments to improve the outcomes for those diagnosed with autism and those who face difficulties with cognitive and behavioral flexibility.