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# Early Gaze Behaviours in Infants at High Familial Risk for Autism Spectrum Disorder: association with brain development

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**Abstract**

Children diagnosed with Autism Spectrum Disorder (ASD) show impairments in gaze-following and will seldom engage in joint attention (JA). The ability to initiate JA (IJA) can be more impaired than the ability to respond to JA (RJA). In a longitudinal study, 101 high-risk infants for ASD (62% males) completed MRI scans at 4 or 6 months of age. Subcortical volumes (thalamus, basal ganglia, hippocampus, amygdala) were extracted. Gaze and JA behaviours were assessed with standardized measures. The majority of infants were IJA non-responders (n=93, 92%), and over half were RJA non-responders (n=50, 52%). In the non-responder groups, models testing the association of subcortical volumes with later ASD diagnosis accounted for age, sex, and cerebral volumes. It was found that hippocampal and thalamic volumes predicted later ASD diagnosis. Findings suggest that these brain regions may present increased vulnerability early in life and might be key predictors of the development of ASD.

**Keywords**

Social Gaze

Brain Development

Autism Spectrum Disorder

Infants

MRI

## Summary for Lay Audience

Children diagnosed with Autism Spectrum Disorder (ASD) are more likely to present difficulties in following where the gaze of others is being directed and in engaging in shared attention with another person and a third object. These behaviours are called joint attention (JA). The ability to follow the gaze of another person, responding to joint attention (RJA), is different from initiating joint attention (IJA), which is when the child looks or points to something for the purpose of sharing interests with others. Difficulties in joint attention are observed in young children with ASD. These children have a harder time initiating shared interest than following somebody's gaze. In this study, 101 babies who had an older sibling diagnosed with ASD were assessed (62% males). Because ASD is considered a genetic disorder, these babies are considered to be at higher risk for developing ASD. Babies completed image exams when they were 4 or 6 months of age. From these images, volumes of some specific areas of the brain were extracted. Behavioural measures of early gaze and joint attention were assessed using 2 questionnaires. It was found that the majority of babies were non-responders to Initiating Joint Attention (n=93, 92%), and over half were non-responders to Responding to Joint Attention (n=50, 52%). We separated them into groups, responders and non-responders, and we tested the association of subcortical brain volumes with later ASD diagnosis. In the group that did not respond to Initiate Joint Attention, we found that volumes of some specific areas (hippocampus and thalamus) predicted later ASD diagnosis. Findings suggest that these brain areas might be key predictors of the development of ASD.

## Co-Authorship Statement

I hereby declare that this thesis incorporates material that is a result of joint research, which can be found in Chapter 2. The content of this chapter includes the manuscript entitled "*Joint Attention in Infants at High Familial Risk for Autism Spectrum Disorder and the association with Thalamic and Hippocampal Macrostructure*" which has the following co-authors: Dr. Diane Seguin and Dr. Emma Duerden. Both researchers were involved in the study design and are co-authors of the manuscript. The manuscript was submitted to a peer-reviewed journal, *Cerebral Cortex Communications,* and it is now under review. The data were collected as part of the Autism Centers of Excellence (ACE) Program and the Infant Brain Imaging Study (IBIS) Network and were made available by the National Database for Autism Research (NDAR) repository of the National Institute of Mental Health Data Archive (NDA). This data has open access to researchers in the field. As the first author, I wrote the manuscript independently and completed the analysis. Only my primary contributions towards the publication are included in this thesis, and the contribution of co-authors was primarily through providing assistance in study design and analysis, as well as contributing with valuable feedback on the refinement of ideas and editing of the manuscript.

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# Chapter 1

## 1.1 Introduction

Humans are inherently social creatures and seek interactions very early in life. Infants interact with others long before language development. Early social interactions are extremely important for the development of later social and communication skills. Direct gaze and following another person's gaze are important aspects of social interactions, and they provide information about the environment. However, the gaze cannot be limited to looking at others' eyes; the whole head, face, and eyes provide extremely valuable information important for social living (Emery, 2000). The eyes, in turn, can provide very subtle and complex information, and are also an important point of contact between infants and caregivers (Emery, 2000; Haith, 1977). The understanding of those subtle visual signals relies on the ability of the individual to interpret them, and this ability begins early in development (Emery, 2000). The ability to follow the gaze of others is called joint attention, and it allows infants to be able to gaze into another individual's eyes to get information about others' emotional expressions and potential intentions, and also to infer others' mental states, known as "theory of mind" (Emery, 2000; Guellai et al., 2020). In turn, gaze and joint attention are important tools for the development of social cognition, and consequently for language acquisition (Mundy & Crowson, 1997).

Social cognition allows individuals to process the information coming from the social world, make sense of it, and determine the appropriate action (Glynn & Watkiss, 2016). The development of social cognition and communication begins early in life and depends on the ability to respond spontaneously to social overtures coming from a variety of sources (Chawarska et al., 2013). Impairments in early social gaze and in joint attention are some of the earliest indicators of later development of Autism Spectrum Disorder (ASD) (Bottema-Beutel, 2016; Bruinsma et al., 2004; Charman, 2003; Ibanez et al., 2013; Werner, 2005). Infants who do not respond to social situations early in life may be at risk for navigating the social world. The Canadian Paediatric Society recommends developmental surveillance through standardized measures to identify those infants at risk for delay or disorder in order to allow flagged infants to access early interventions

(Zwaigenbaum et al., 2019). Accessing early interventions, in turn, can promote improved social and behavioural outcomes.

### 1.1.1 Early interactions and Social and Language Development

The early interactions between babies and their caregivers are extremely important for later social and language development. Infants are extremely sensitive to the social environment,  and it has been recognized that infants socialize with their caregivers very early in life (Tomasello et al., 2005). Such communication and interactions take place in various ways such as through gaze, gestures, and vocalizations (Tamis-LeMonda et al., 2014; Tomasello et al., 2005). Parents or caregivers, in turn, respond to those signals through actions and talking to infants, thereby facilitating language and social development (Tamis-LeMonda et al., 2014).

Those first early interactions between infants and caregivers are extremely important for scaffolding the later ability to communicate and participate in the social world. Before the 1960s, infants were seen as passive beings and merely as a system of reflexes reacting to the environment (Carmichael, 1946; Clifton, 2001; Malloch, 2009). Previous studies did not acknowledge the range of cognitive and social abilities human infants possessed (Malloch, 2009). Research later revealed that humans are inherently social creatures and are born willing to interact with others (Carpenter et al., 1998; Malloch, 2009). Despite the early notion that babies were born with limited social-communication skills, it was later recognized that babies not only are born ready to interact, but also those early interactions are extremely important for later social and language development. Acoustic analysis of baby-caregiver interactions demonstrated that the caregiver and baby listen to each other, and the babies' movements are coordinated to the rhythm of the caregiver's speech (Malloch, 1999, 2009). Those interactions between infants and adults happen dyadically, meaning that both adults and infants interact in a mutual fashion (Tomasello et al., 2005).

Even though young infants are not yet capable of producing verbal speech, they produce vocalizations that provoke responses from adults as early as 2 months of age (Gratier et al., 2015). These vocalizations are called 'protoconversations' because they have a conversational structure similar to an adult conversation (Bateson, 1975; Beebe, 1988; Gratier et al., 2015; Tomasello et al., 2005). These vocalizations are different from those in which the baby is exploring the available sounds and are accompanied by the infants' gaze to the adult's eyes (Gratier et al., 2015; Tomasello et al., 2005). This gaze during the protoconversations is called mutual gaze and is characterized by the infant's direct engagement with others (Tomasello et al., 2005).

The mutual gaze early in life, as well as the dyadic protoconversations between infants and caregivers, scaffold the later ability for social and communication abilities. Gratier et al. (2015) demonstrate in their study that 2 to 3-month-old infants actively participate in turn-taking conversations, as well as initiation with their caregivers, and that their caregivers adjust this turn-taking with the infants using co-regulation strategies. In other words, the baby and caregiver both adjust the turn-taking sequence of the conversation (Gratier et al., 2015; Tomasello et al., 2005). Caregivers' vocalizations directed to infants are longer and contain more pauses compared to the pauses in speech with another adult (Gratier et al., 2015). The turn-taking process tends to slow down around 9 months of age when infants acquire more complex communicative and social skills (Gratier et al., 2015). These findings highlight the importance of those early interactions in social and language development. However, those abilities also depend on the infant's condition to respond spontaneously to social cues (Chawarska et al., 2013).

### 1.1.2 Social Gaze & Social Cognition

One important aspect of human social interaction is social gaze. Social gaze is defined by "the orientation of the eyes within the face" (APA, 2020. Gaze definition). Previous studies have already demonstrated that infants perceive the gaze of others and prefer faces that engage in mutual gaze from the first days after birth (Farroni, 2002; Guellai et al., 2020). This means that infants prefer the direct gaze toward them instead of a gaze that looks in a different direction. This preference is also greater if the direct gaze is paired with a social situation, such as talking (Guellai et al., 2020). Direct eye gaze plays a role in social interactions, being an important communication channel (Guellai et al., 2020). The early sensitivity to direct gaze paired with other socially relevant experiences, lay the foundation for developing more complex forms of social skills

(Guellai et al., 2020). Eye gaze, in turn, is a main predictor for developing later social cognition (Farroni, 2002).

Social cognition is defined as “the cognition in which people perceive, think about, interpret, categorize, and judge their own social behaviours and those of others” (APA, 2020. Social cognition definition). Social cognitive processes include the ability to understand the perspective of others, to represent others’ emotional expressions and intentions, and also to interpret a variety of social cues (Mundy, 2018). Social cognition allows individuals to make sense of the world and is extremely important in social interactions; in turn, social cognition allows individuals to navigate the social world (Glynn & Watkiss, 2016). The development of social cognition and communication begins early in life and depends on the ability to respond spontaneously to social situations (Chawarska et al., 2013). It is now known that human infants are born willing to interact with others and pay special attention to others’ eyes (Carpenter et al., 1998; Farroni, 2002; Guellai et al., 2020; Tomasello et al., 2005).

### 1.1.3 Joint Attention

Infants, even during the first months of life, are aware of what is happening around them and they express themselves through gestures, vocalizations, and gaze so as to capture their caregiver’s attention and engage in an interaction (Malloch, 2009). Around 4 months of life, infants also become interested in objects and start to play with those objects, thereby sharing the interest with the caregiver (Malloch, 2009; Tomasello et al., 2005). This process of shared interaction begins with “primary intersubjectivity”, which is related to this dyadic relationship with the caregiver, and ends with “secondary intersubjectivity”, which makes it possible to integrate a third element (person or object) into this first dyadic relationship (Carpenter et al., 1998; Malloch, 2009; Tomasello et al., 2005). These interactions are characterized by the infant coordinating the attention with the person whom he/she is interacting, and an external object, person, or event (Tomasello et al., 2005). The infant, at this point, gradually shifts from an interaction with a partner, a dyadic interaction, to this alternated and coordinated attention with the partner and the object, a triadic interaction (Bruinsma et al., 2004; Tomasello et al., 2005).

This integration of a third element is called “joint attention” (JA) and allows infants to share a common focus on other individuals or objects (Gong & Shuai, 2012; Swanson et al., 2013; Tomasello et al., 2005). JA allows infants to be able to gaze into another individual's eyes to get information about the other individual's emotional state, and about where the eye gaze is being directed. One example of JA is when an infant looks at an adult who is looking at an object, for example, a ball. Then, the infant follows the adult’s gaze and looks towards the ball.  This process also allows the infant to direct the attention of others towards something else, for example, pointing to a ball in order to direct the adult’s gaze to the ball as well. The inclusion of this third element is what differentiates joint attention from imitation and face processing (Mundy, 2018). The ability to look into the eyes and follow gaze are prerequisites for engaging in JA.

JA is an umbrella term that includes different types of behaviours (Bruinsma et al., 2004). All JA behaviours include a common goal of sharing the third element with another person (Bottema-Beutel, 2016). Typically-developing infants at 3 months of age will begin exhibiting joint attention, which is considered to be solidified by 18 months of age (Carpenter et al., 1998). JA includes two constructs, responding to joint attention (RJA) and initiating joint attention (IJA). The ability or propensity of the child to respond to social situations with a partner and to follow where others’ gaze is being directed is called ‘Responding to Joint Attention’ (RJA) or ‘gaze-following’ (Bottema-Beutel, 2016). The ability to initiate an interaction with a partner in order to share interest or share affect about a third element, which could be an object or event, is called ‘Initiating Joint Attention’ (IJA) (Bottema-Beutel, 2016; Gong & Shuai, 2012; Swanson et al., 2013). IJA is considered a more complex ability than RJA, having the infant taking a more active role in IJA (Mundy, 2018). IJA requires the infant to intentionally initiate the interaction with another person (Bottema-Beutel, 2016). This initiation can take place in various ways such as looking, pointing, or using verbal language (Bottema-Beutel, 2016). The infant shifts from being a signal-receiver when displaying RJA behaviours to being a signal-sender in IJA, and motivation to engage in social interactions is required (Mundy, 2018). JA also precedes language acquisition and plays a role in transitioning to symbolic communication (Bottema-Beutel, 2016; Bruinsma et al., 2004).

### 1.1.4 Joint Attention Neural Networks

Specific neural networks have been associated with JA behaviours, which are different from those associated with face processing and imitation (Mundy, 2018). However, distinct neural networks have been identified for the different types of JA behaviours, RJA and IJA. Activation of the superior temporal cortex, intraparietal cortex, temporal pole, orbitofrontal cortex, and insular cortex was associated with RJA behaviours (Mundy, 2018). While activation of the frontal pole, dorsal frontal cortex, superior temporal and intraparietal cortex, posterior cingulate, and sensorimotor cortices was associated with IJA behaviours (Mundy, 2018). It has been suggested that some neural circuitries in both IJA and RJA could overlap, such as in the frontal and temporal cortices (Billeci et al., 2017). Some subcortical areas of the brain have also been suggested to be associated with JA behaviours, such as the amygdala, the striatum, and the hippocampus (Gordon et al., 2013; Mundy, 2018). Mundy (2018) suggested that sharing attention, especially in the first years of life, is fundamental to providing brain-behaviour experiences to build the neural systems necessary for later language development and for the development of social cognition.

### 1.1.5 Autism Spectrum Disorder and gaze and joint attention impairments

Autism Spectrum Disorder (ASD) is a heritable neurodevelopmental disorder that includes deficits in two domains: social communication and social interactions, and restricted and repetitive behaviours (American Psychiatric Association, 2013). Even though it is now recognized that early neurodevelopmental abnormalities are present in the first year of life, usually, the diagnosis of ASD is not made until the child is at least 3 years of age (Di Giorgio et al., 2016). The full manifestation of symptoms usually appears later in life, when communication delays are more easily identified (American Psychiatric Association, 2013; Zwaigenbaum, 2010). ASD is estimated to affect 1 in 66 children and youth in Canada, which means 1.5% of this population, according to the Public Health Agency of Canada report made in 2018 (Ofner, 2018). The prevalence in boys is 1 in 42, while in girls, it is 1 in 165 (Ofner, 2018). The prevalence rates, however, are higher in infants who have older biological siblings with ASD, with an increased prevalence of 19% (Ozonoff et al., 2011).

Infants who do not respond spontaneously to social bids and struggle with early social gaze may be more likely to be diagnosed with neurodevelopmental disorders, such as ASD. Individuals diagnosed with ASD seldom engage in joint attention, which is considered a core symptom of ASD diagnosis (Bruinsma et al., 2004; Dawson et al., 2004; Swanson et al., 2013). Difficulties in spontaneous gaze-following and in engaging in joint attention have also been studied as one of the earliest indicators of later ASD development (Bottema-Beutel, 2016; Bruinsma et al., 2004; Werner, 2005). Previous studies have shown that toddlers who were at high risk for developing ASD showed less joint attention skills and engagement than typically-developing toddlers, and this association became even greater for those who later developed ASD (Adamson et al., 2019). Individuals with ASD also have a preference, even early in life, for non-social stimuli rather than social ones (Di Giorgio et al., 2016). This tendency to orient to non-social situations has been studied as a possible predictor of ASD development later in childhood (Chawarska et al., 2013; Di Giorgio et al., 2016; Peltola et al., 2018; Pierce et al., 2011).

### 1.1.6 Subcortical brain alterations in ASD

Researchers have also investigated brain areas that could be associated with ASD core symptomatology. Previous studies found that individuals with ASD demonstrated alterations in some specific subcortical brain regions when compared to typically-developing individuals. Van Rooij et al. (2018) found an association between ASD and smaller volumes of subcortical areas such as the pallidum, putamen, nucleus accumbens, and amygdala. The findings suggest that alterations in those areas could be associated with social-motivational and cognitive/motor impairments in ASD, and with mechanisms of social reward differences observed in individuals with ASD (van Rooij et al., 2018). Other studies, however, found that the amygdala was enlarged in children with ASD, compared to the control group (Mosconi et al., 2009; Nordahl, 2012). Mosconi et al. (2009) also found an association between amygdala volumes and joint attention ability in 2- and 4-year-old children with ASD. Sussman et al. (2015) reported smaller subcortical volumes in brain areas, such as in the hippocampus, thalamus, and globus pallidus in children and adolescents with ASD, relative to total brain volume. Smaller thalamic volumes have also been reported in children diagnosed with ASD compared to control groups (Sussman et al., 2015; Tamura et al., 2010). These findings suggest that these alterations could be associated with memory and sensory impairments observed in individuals with ASD (Sussman et al., 2015).

### 1.1.7 Early detection and early intervention

As infants who exhibit impairments in early social gaze and joint attention may be at risk for developing neurodevelopmental disorders, such as ASD, the Canadian Paediatric Society recommends developmental surveillance through standardized measures in order to identify those infants at risk for delay or disorder (Zwaigenbaum et al., 2019). This allows flagged infants to access early interventions. However, the general developmental assessment tools are not specifically designed to screen for ASD (e.g., Ages and Stages Questionnaire, Nipissing District Developmental Screen, Child Development Inventory, and Parents Evaluation of Developmental Status), and the other tools that assess early ASD symptoms usually focus on the second year of life (e.g., Modified Checklist for Autism in Toddlers, Autism Spectrum Rating Scales, and Infant Toddler Checklist) (Olliac et al., 2017). According to the Public Health Agency of Canada, only 56% of ASD-diagnosed children and youth had received that diagnosis by 6 years of age, and less than 20% had received a diagnosis by 3 years of age (Ofner, 2018). The median age of diagnosis is 5.3 years of age, with a range between 2 and 17 years old (Ofner, 2018). The delayed detection and diagnosis of those infants/children could hinder the opportunities for promptly accessing appropriate care to restore developmental trajectories.

Early detection of infants at risk, which does not mean necessarily receiving a diagnosis at an early age, is essential to allow those infants to access early interventions and fully benefit from them (Zwaigenbaum, 2010). A better understanding of early signs of ASD development, as well as of the neural mechanisms behind them, could help identify targets for intervention, as well as biomarkers, to promote improved social and behavioural outcomes in infants who are at high risk for the development of ASD. Early interventions have been associated with better social and behavioural outcomes, diminishing the societal costs of ASD to the individual, to their families, and to society (Horlin, 2014; Zwaigenbaum, 2010).

## 1.2 The Current Study

By conducting the current study, the goal is to better understand early gaze behaviours in infants who are at-risk for the development of ASD. Further, the goal was to examine early gaze behaviours in relation to subcortical macrostructural maturation in relation to ASD diagnosis. In the current work, JA abilities in young infants who carried a familial risk for the development of ASD were studied using behavioural assessments of early gaze behaviours. To study brain development, the infants who had a sibling diagnosed with ASD also underwent structural neuroimaging at 4 or 6 months of age. The subcortical brain volumes were automatically extracted. The association between subcortical brain volumes, behavioural measures of responding to and initiating JA, and ASD diagnostic status was examined. It was hypothesized that JA behaviours would be limited or absent in infants who were later diagnosed with ASD; and subcortical volumes would predict ASD diagnosis in infants. A better characterization of early joint attention behaviours and the underlying neural mechanisms could identify key windows for intervention to promote social gaze. Additionally, using neuroimaging methods in young infants, this could identify brain-based biomarkers to assess the efficacy of interventions overtime. Overall, better characterization of joint attention behaviours and the underlying neural mechanisms may lead to future hypothesis-driven research aimed at promoting improved social and behavioural outcomes in infants who are at-risk for the development of ASD (Zwaigenbaum, 2010).

### 1.3 References

Adamson, L. B., Bakeman, R., Suma, K., & Robins, D. L. (2019). An Expanded View of Joint Attention: Skill, Engagement, and Language in Typical Development and Autism. *Child Dev*, *90*(1), e1-e18. https://doi.org/10.1111/cdev.12973

American Psychiatric Association. (2013). *Diagnostic and statistical manual of mental disorders : DSM-5* (5th ed.). American Psychiatric Association.

American Psychological Association Dictionary (2020). *Gaze definition*. Retrieved March 19th, 2021. https://dictionary.apa.org/gaze

American Psychological Association Dictionary (2020). *Social Cognition definition*. Retrieved March 19th, 2021. https://dictionary.apa.org/social-cognition

Bateson, M. C. (1975). Mother-infant exchanges the epigenesis of conversational. *Annals New York Academy of Sciences*, 101-113.

Beebe, B., Alson, D., Jaffe, J., Feldstein, S.,Crown, C. . (1988). Vocal Congruence in Mother-Infant Play. *Journal of Psycholinguistic Research*, *17*, 245-259.

Billeci, L., Narzisi, A., Tonacci, A., Sbriscia-Fioretti, B., Serasini, L., Fulceri, F., . . . Muratori, F. (2017). An integrated EEG and eye-tracking approach for the study of responding and initiating joint attention in Autism Spectrum Disorders. *Sci Rep*, *7*(1), 13560. https://doi.org/10.1038/s41598-017-13053-4

Bottema-Beutel, K. (2016). Associations between joint attention and language in autism spectrum disorder and typical development: A systematic review and meta-regression analysis. *Autism Res*, *9*(10), 1021-1035. https://doi.org/10.1002/aur.1624

Bruinsma, Y., Koegel, R. L., & Koegel, L. K. (2004). Joint attention and children with autism: a review of the literature. *Ment Retard Dev Disabil Res Rev*, *10*(3), 169-175. https://doi.org/10.1002/mrdd.20036

Carmichael, L. (1946). The onset and early development of behavior. In *Manual of child psychology*. (pp. 45-166). John Wiley & Sons Inc. https://doi.org/10.1037/10756-002

Carpenter, M., Nagell, K., & Tomasello, M. (1998). Social cognition, joint attention, and communicative competence from 9 to 15 months of age. *Monogr Soc Res Child Dev*, *63*(4), i-vi, 1-143.

Charman, T. (2003). Why is joint attention a pivotal skill in autism? *Philos Trans R Soc Lond B Biol Sci*, *358*(1430), 315-324. https://doi.org/10.1098/rstb.2002.1199

Chawarska, K., Macari, S., & Shic, F. (2013). Decreased spontaneous attention to social scenes in 6-month-old infants later diagnosed with autism spectrum disorders. *Biol Psychiatry*, *74*(3), 195-203. https://doi.org/10.1016/j.biopsych.2012.11.022

Clifton, R. K. (2001). Lessons From Infants: 1960-2000. *Infancy*, *2*(3), 285-309.

Dawson, G., Toth, K., Abbott, R., Osterling, J., Munson, J., Estes, A., & Liaw, J. (2004). Early social attention impairments in autism: social orienting, joint attention, and attention to distress. *Dev Psychol*, *40*(2), 271-283. https://doi.org/10.1037/0012-1649.40.2.271

Di Giorgio, E., Frasnelli, E., Rosa Salva, O., Scattoni, M. L., Puopolo, M., Tosoni, D., . . . Vallortigara, G. (2016). Difference in Visual Social Predispositions Between Newborns at Low- and High-risk for Autism. *Sci Rep*, *6*, 26395. https://doi.org/10.1038/srep26395

Emery, N. J. (2000). The eyes have it: the neuroethology, function and evolution of social gaze. *Neuroscience and Biobehavioral Reviews*, *24*, 581-604.
Farroni, T., Csibra, G., Simion, F., Johnson, M. H. . (2002). Eye contact detection in humans from birth. *PNAS*, *99*(14), 9602–9605. https://doi.org/www.pnas.org/cgi/doi/10.1073/pnas.152159999
Glynn, M. A., & Watkiss, L. (2016). Social Cognition. In *The Palgrave Encyclopedia of Strategic Management* (pp. 1-4). https://doi.org/10.1057/978-1-349-94848-2_614-1
Gong, T., & Shuai, L. (2012). Modelling the coevolution of joint attention and language. *Proc Biol Sci*, *279*(1747), 4643-4651. https://doi.org/10.1098/rspb.2012.1431
Gordon, I., Eilbott, J. A., Feldman, R., Pelphrey, K. A., & Vander Wyk, B. C. (2013). Social, reward, and attention brain networks are involved when online bids for joint attention are met with congruent versus incongruent responses. *Soc Neurosci*, *8*(6), 544-554. https://doi.org/10.1080/17470919.2013.832374
Gratier, M., Devouche, E., Guellai, B., Infanti, R., Yilmaz, E., & Parlato-Oliveira, E. (2015). Early development of turn-taking in vocal interaction between mothers and infants. *Front Psychol*, *6*, 1167. https://doi.org/10.3389/fpsyg.2015.01167
Guellai, B., Hausberger, M., Chopin, A., & Streri, A. (2020). Premises of social cognition: Newborns are sensitive to a direct versus a faraway gaze. *Sci Rep*, *10*(1), 9796. https://doi.org/10.1038/s41598-020-66576-8
Haith, M. M., Bergman, T., Moore. M. J., . (1977). Eye Contact and Face Scanning in Early Infancy. *Science*, *198*(4319), 853-855.
Horlin, C., Falkmer, M., Parsons, R., Albrecht, M. A., Falkmer, T. . (2014). The Cost of Autism Spectrum Disorders. *PLoS One*, *9*(9), e106552-e106552. https://doi.org/10.1371/journal. pone.0106552
Ibanez, L. V., Grantz, C. J., & Messinger, D. S. (2013). The Development of Referential Communication and Autism Symptomatology in High-Risk Infants. *Infancy*, *18*(5). https://doi.org/10.1111/j.1532-7078.2012.00142.x
Malloch, S. (1999). Mother and Infants and Communicative Musicality. *Musicae Sclentiae*(Special Issue 1999-2000), 29-57.
Malloch, S., Trevarthen, C. (2009). Musicality: Communicating the vitality and interests of life. In *Communicative Musicality: Exploring the Basis of Human Companionship* (pp. 1-11). Oxford: Oxford University Press.
Mosconi, M. W., Cody-Hazlett, H., Poe, M. D., Gerig, G., Gimpel-Smith, R., & Piven, J. (2009). Longitudinal study of amygdala volume and joint attention in 2- to 4-year-old children with autism. *Arch Gen Psychiatry*, *66*(5), 509-516. https://doi.org/10.1001/archgenpsychiatry.2009.19
Mundy, P. (2018). A review of joint attention and social-cognitive brain systems in typical development and autism spectrum disorder. *Eur J Neurosci*, *47*(6), 497-514. https://doi.org/10.1111/ejn.13720
Mundy, P., & Crowson, M. (1997). Joint attention and early social communication: implications for research on intervention with autism. *J Autism Dev Disord*, *27*(6), 653-676.
Nordahl, C. W., Scholz, R., Yang, X., Buonocore, M.H., Simon, T., Rogers, S., Amaral, D.G., . (2012). Increased Rate of Amygdala Growth in Children Aged 2 to 4 Years With Autism Spectrum Disorders. *Arch Gen Psychiatry*, *69*(1), 53-61.

Ofner, M., Coles, A., Decou, M. L., Do, M. T., Bienek, A., Snider, J., Ugnat, A. M. (2018). *Autism Spectrum Disorder among children and youth in Canada 2018: A Report of the National Autism Spectrum Disorder Surveillance System* (1st ed.). Public Health Agency of Canada.

Olliac, B., Crespin, G., Laznik, M. C., Cherif Idrissi El Ganouni, O., Sarradet, J. L., Bauby, C., . . . Saint-Georges, C. (2017). Infant and dyadic assessment in early community-based screening for autism spectrum disorder with the PREAUT grid. *PLoS One*, *12*(12), e0188831. https://doi.org/10.1371/journal.pone.0188831

Ozonoff, S., Young, G. S., Carter, A., Messinger, D., Yirmiya, N., Zwaigenbaum, L., . . . Stone, W. L. (2011). Recurrence risk for autism spectrum disorders: a Baby Siblings Research Consortium study. *PEDIATRICS*, *128*(3), e488-495. https://doi.org/10.1542/peds.2010-2825

Peltola, M. J., Yrttiaho, S., & Leppanen, J. M. (2018). Infants' attention bias to faces as an early marker of social development. *Dev Sci*, *21*(6), e12687. https://doi.org/10.1111/desc.12687

Pierce, K., Conant, D., Hazin, R., Stoner, R., & Desmond, J. (2011). Preference for geometric patterns early in life as a risk factor for autism. *Arch Gen Psychiatry*, *68*(1), 101-109. https://doi.org/10.1001/archgenpsychiatry.2010.113

Sussman, D., Leung, R. C., Vogan, V. M., Lee, W., Trelle, S., Lin, S., . . . Taylor, M. J. (2015). The autism puzzle: Diffuse but not pervasive neuroanatomical abnormalities in children with ASD. *Neuroimage Clin*, *8*, 170-179. https://doi.org/10.1016/j.nicl.2015.04.008

Swanson, M. R., Serlin, G. C., & Siller, M. (2013). Broad autism phenotype in typically developing children predicts performance on an eye-tracking measure of joint attention. *J Autism Dev Disord*, *43*(3), 707-718. https://doi.org/10.1007/s10803-012-1616-7

Tamis-LeMonda, C. S., Kuchirko, Y., & Song, L. (2014). Why Is Infant Language Learning Facilitated by Parental Responsiveness? *Current Directions in Psychological Science*, *23*(2), 121-126. https://doi.org/10.1177/0963721414522813

Tamura, R., Kitamura, H., Endo, T., Hasegawa, N., & Someya, T. (2010). Reduced thalamic volume observed across different subgroups of autism spectrum disorders. *Psychiatry Res*, *184*(3), 186-188. https://doi.org/10.1016/j.pscychresns.2010.07.001

Tomasello, M., Carpenter, M., Call, J., Behne, T., & Moll, H. (2005). Understanding and sharing intentions: the origins of cultural cognition. *Behav Brain Sci*, *28*(5), 675-691; discussion 691-735. https://doi.org/10.1017/S0140525X05000129

van Rooij, D., Anagnostou, E., Arango, C., Auzias, G., Behrmann, M., Busatto, G. F., . . . Buitelaar, J. K. (2018). Cortical and Subcortical Brain Morphometry Differences Between Patients With Autism Spectrum Disorder and Healthy Individuals Across the Lifespan: Results From the ENIGMA ASD Working Group. *Am J Psychiatry*, *175*(4), 359-369. https://doi.org/10.1176/appi.ajp.2017.17010100

Werner, E., Dawson, G., . (2005). Validation of the Phenomenon of Autistic Regression Using Home Videotapes. *Arch Gen Psychiatry, 62*, 889-895.

Zwaigenbaum, L. (2010). Advances in the early detection of autism. *Curr Opin Neurol*, *23*(2), 97-102. https://doi.org/10.1097/WCO.0b013e3283372430

Zwaigenbaum, L., Brian, J. A., & Ip, A. (2019). Early detection for autism spectrum disorder in young children. *Paediatr Child Health*, *24*(7), 424-443. https://doi.org/10.1093/pch/pxz119

# Chapter 2

## 2 Joint Attention in Infants at High Familial Risk for Autism Spectrum Disorder and the association with Thalamic and Hippocampal Macrostructure

Autism Spectrum Disorder (ASD) is a neurodevelopmental disorder that includes deficits in social communication and social interactions, and restricted and repetitive behaviours (American Psychiatric Association, 2013). In Canada, ASD is estimated to affect 1 in 66 children and youth, or 1.5% of the population (Ofner, 2018). ASD diagnosis is male-biased, 1 in 42 males affected, and 1 in 165 females affected (Ofner, 2018). Additionally, there is an increased prevalence of ASD in children with an older biological sibling with an ASD diagnosis (Ozonoff et al., 2011). Usually, the diagnosis of ASD is not made until the child is at least 3 years of age when communication delays are more easily identified (Di Giorgio et al., 2016; Zwaigenbaum, 2010). The full manifestation of symptoms is expected to appear later in development, especially when social communication demands exceed the individual's abilities (American Psychiatric Association, 2013). Identification of the early signs of ASD during the first year of life is needed for infants to access early therapies, which can promote better social and behavioural outcomes.

Social gaze, or direct eye contact, is an important communication channel and plays a key role in social interactions (Guellai et al., 2020). The eyes provide very subtle and complex information, which contributes to social living (Emery, 2000; Haith, 1977). Understanding these subtle visual signals relies on the ability of the individual to correctly interpret them (Emery, 2000) and this ability begins very early in development. Several studies have demonstrated that infants, from the first days after birth, perceive the gaze of others and prefer faces that engage in mutual gaze (Farroni, 2002; Guellai et al., 2020). Social gaze lays the foundation for developing more complex forms of social cognition, such as following the gaze of others, regulating turn-taking in conversations, and inferring others' mental states (Emery, 2000; Guellai et al., 2020). The ability to follow gaze is necessary for the development of joint attention. Joint attention (JA) is the ability to look into the eyes and follow the gaze of others (Dawson et al., 2004). JA is essential for social emotional development and for later language acquisition (Mundy & Crowson, 1997).

An early clinical indicator in infants who later develop ASD is impairments in directing gaze and following the eye gaze of others (Charman, 2003; Ibanez et al., 2013). Typically-developing infants as young as 3 months will begin exhibiting JA, and this skill is solidified by 18 months of age (Carpenter et al., 1998). Impairments in spontaneous gaze-following and in engaging in JA are some of the earliest indicators of later ASD development (Bottema-Beutel, 2016; Bruinsma et al., 2004; Charman, 2003; Ibanez et al., 2013; Werner, 2005).

The ability to follow gaze cues of others is known as Responding to Joint Attention (RJA) or 'gaze following' (Bottema-Beutel, 2016; Mundy, 2018). JA also includes the ability to initiate an interaction with a partner in order to share interest about a third element, which could be an object or event (Bottema-Beutel, 2016; Mundy, 2018). This ability to seek interaction is known as Initiating Joint Attention (IJA) (Bottema-Beutel, 2016; Gong & Shuai, 2012; Swanson et al., 2013). Impairments in IJA are considered to be more prevalent than impairments in RJA in individuals with ASD (Gangi et al., 2014; Ibanez et al., 2013; Mundy, 2018). Previous studies have shown that toddlers who had a higher likelihood of developing ASD showed weaker JA skills and engagement than typically developing toddlers, and this association was even greater for those who later developed ASD (Adamson et al., 2019). The consistent practice of JA provides experiences for infants to build the neural systems necessary for social interactions and mentalizing (Mundy, 2018).

Distinct neural networks have been identified in RJA and IJA behaviours in infants. RJA involves activation of the superior temporal cortex, intraparietal cortex, temporal pole, orbitofrontal cortex, and insular cortex, while IJA is associated with activation of the frontal pole, dorsal frontal cortex, superior temporal and intraparietal cortex, posterior cingulate, and sensorimotor cortices (Mundy, 2018). Other studies suggest that some neural circuitries in both IJA and RJA could overlap such as in the frontal and temporal cortices (Billeci et al., 2017). Research studies report that subcortical brain areas such as the amygdala, the striatum, and hippocampus are also implicated in JA behaviours (Gordon et al., 2013; Mundy, 2018). Mosconi et al. (2009) found an association between increased amygdala volumes and decreased JA ability in 2 and 4-year-old children with

ASD. Whether comparable associations between JA behaviours and subcortical volumetric development occurs in infancy is currently understudied.

In the current work, we examined JA abilities in young infants who carried a familial risk for the development of ASD. Infants with a sibling diagnosed with ASD were assessed for joint attention and underwent structural neuroimaging at 4 or 6 months of age. We examined the association between subcortical brain volumes, behavioural measures of responding to and initiating JA, and ASD diagnostic status. We hypothesized that JA behaviours will be limited or absent in infants who are later diagnosed with ASD; and subcortical volumes will predict ASD diagnosis in infants. A better understanding of early joint attention and underlying neural mechanisms could identify key windows for intervention, as well as biomarkers to use as identification tools for accessing early interventions in order to promote improved social and behavioural outcomes in infants who have an increased likelihood for the development of ASD (Zwaigenbaum, 2010).

## 2.1 Methods

### 2.1.1 Participants

Initially, data from 107 infants (63% males) were collected through the National Database for Autism Research (NDAR) repository as part of the National Institute of Mental Health Data Archive (NDA) (Payakachat et al., 2016). Individuals were recruited as part of a longitudinal study to examine brain-based and behavioural phenotypes in infants who carry a familial risk for the development of ASD. All infants in the study had a sibling who was diagnosed with ASD. The infants were recruited from multiple sites through institutions that were part of the Autism Centers of Excellence (ACE) Program and the Infant Brain Imaging Study (IBIS) Network. The infants completed magnetic resonance imagining (MRI) scans assessments when they were 4 or 6 months of age and completed behavioural assessments at multiple time points, from 5 to 15 months of age. A total of 20 infants (18.7%) in the sample received a diagnosis of ASD, confirmed by the Autism Diagnostic Observational Schedule (ADOS) (Lord et al., 2000). A total of 17 of the children diagnosed were male (85%) and 3 were female (15%).

Data from Vineland Adaptive Behavior Scale-II (VABS-II) and Autism Observation Scale for Infants (AOSI) were used to examine early gaze and joint attention behaviours. For the current study, scores from the assessments completed at the closest time point to the MRI scans were used. The age of the first assessment completed for the VABS-II varied from 5 to 14 months of age, and from 6 to 15 months of age for the AOSI. As joint attention is a developmentally sensitive process, we chose to limit to the behavioural assessments that were performed at 9 months of age or below to ensure all behavioural and image data were collected within a comparable developmental period. All participants who were later diagnosed as ASD had data collected prior to 9 months of age and were included in the analysis. After applying the exclusion criteria, we had a total of 101 participants (62% males) in our sample.

### 2.1.2 Behavioural and Developmental Assessments

#### 2.1.2.1 Vineland Adaptive Behavioral Scale-II (VABS-II)

The VABS-II is a standardized norm-referenced measure of adaptive behaviour (Sparrow, 2011). The questionnaire assesses 4 adaptive domains: Communication, Daily Living Skills, Socialization, and Motor Skills. There is one additional domain, Maladaptive Behaviour, which is optional to complete (Community-University Partnership for the Study of Children, 2012). The questionnaire is suitable for infants from birth to adults of 90 years of age (Sparrow, 2011). The questionnaire is available as an interview form (semi-structured, open-ended interview), as well as a parent/caregiver form (Community-University Partnership for the Study of Children, 2012). The interview and parent-caregiver formats do not differ from each other in terms of content, but in how it is administrated (Community-University Partnership for the Study of Children, 2012). The scores for each of the items range from 0 to 2, indicating how often the child displays the behaviour (0=never; 1=sometimes/partially; 2=usually).

#### 2.1.2.2 Autism Observational Scale of Infants (AOSI)

The AOSI is a measure to detect early signs of ASD, particularly for infants who have a familial risk for ASD, to be used exclusively in research contexts (Bryson et al., 2008). The AOSI is composed of semi-structured activities administered by an expert examiner

(Bryson et al., 2008). The activities are divided into 19 tasks in which the examiner observes specific signs of autism in infants (Bryson & Zwaigenbaum, 2014). The AOSI was created based on infants' developmental trajectories (Bryson et al., 2008). Its administration is characterized by an interactive play between an infant and an examiner, while assessing infants' target behaviours (e.g., Visual Tracking, Disengagement of Attention, Orientation to Name, Reciprocal Social Smiling, Differential Response to Facial Emotion, Social Anticipation and Imitation) (Bryson et al., 2008). The measure can be used to assess infants from 6 to 18 months of age for ASD (Bryson et al., 2008). The scores for each of the items range from 0 to 3, indicating if the child displays typical behaviour (0=typical behaviour; 1=inconsistent/partial behaviour; 2=impaired/atypical behaviour; 3=total lack of behaviour).

### 2.1.3 MRI Acquisition

All images were acquired on a Siemens 3T scanner. T1-weighted MR images were acquired with 160 sagittal slices using parameters: repetition time (TR) and the echo time (TE) - TR/TE = 2400/3.16 ms, and voxel resolution=1×1×1 mm3. For this study, T1-weighted images obtained in babies aged 4 or 6 months of age were selected for subsequent image segmentation.

### 2.1.4 MR Image Processing

The quality of the acquired images was visually inspected for motion and other artifacts. The T1-weighted images were subsequently analyzed using recon-all command using Infant FreeSurfer (de Macedo Rodrigues et al., 2015; Fischl, 2012; Zollei et al., 2020). The automatic regional segmentation by the Infant FreeSurfer pipeline was visually qualified on the graphic interface FreeView, available with the Freesurfer suite of tools (http://surfer.nmr.mgh.harvard.edu/). Further manual segmentation was employed to correct segmentation errors in the subcortical gray matter using ITK-SNAP (http://www.itksnap.org/). The quantified measurements of the cortical grey, subcortical white matter, subcortical regions (thalamus, pallidum, putamen, caudate, amygdala, hippocampus) were extracted using age-specific brain atlases at 4 and 6 months depending on the postmenstrual age of the infant at the time of scan. The "brainvol" for global measurements of brain volumes, the "aseg" (Fischl, 2002) for segmentation of subcortical regions including the basal ganglia, cerebellum, and brainstem and the "aparc" (Desikan et al., 2006) or automatic cortical parcellation. A total of 26 volumes and 204 regionally distributed measurements (regional volume, surface area, and cortical thickness) were extracted from the "aseg" (Fischl, 2002) and the "aparc" (Desikan et al., 2006) in each subject, respectively. Segmentation results for representative participants with and without ASD, anatomically annotated (Desikan et al., 2006), are shown in Figure 1.

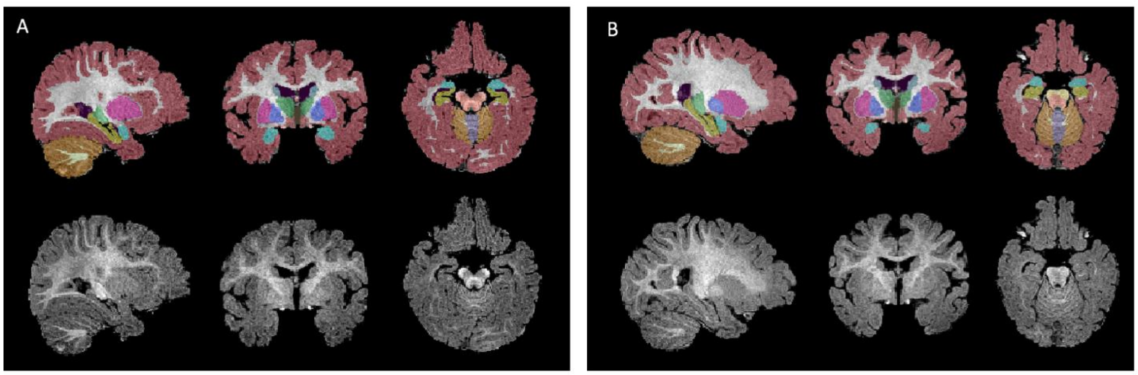


**Figure 1.** *Post-processed T1-weighted MRI scans that were segmented using infant Freesurfer in (A) a high-risk infant who was later diagnosed with ASD and (B) a high-risk infant who did not receive an ASD diagnosis.*

### 2.1.5 Statistical Analysis

Statistical analyses were performed using Statistical Package for the Social Sciences (v.27 SPSS, Chicago, IL). The aim of our study relates to the prediction of joint attention behaviours and subcortical volumes in relation to ASD diagnosis in infants with a familial risk for developing ASD. A comprehensive final model was built to address the two hypotheses of our aim: (1) joint attention behaviours will be limited or absent in those infants who are later are diagnosed with ASD; (2) subcortical volumes of the thalamus, ventral diencephalon, hippocampus, basal ganglia, and amygdala will predict ASD diagnoses in infants. First, from all questionnaires' items (VABS-II and AOSI), we searched for key words related to joint attention, which included: look, eye, watch, point, share, social interest, and attention. Two additional readers reviewed the key words and agreed that those were the best terms to identify JA behaviours, with an agreement rate of 100%. We selected 11 items from both questionnaires that contained one or more key words. In order to classify the behavioural data from both questionnaires, data reduction methods were applied. A Principal Component Analysis (PCA) was applied to the 11 selected items. The PCA allowed for the creation of new constructs combining data from both questionnaires. Using Varimax rotation, 5 components were extracted (Eigenvalues >1). The model was tested for sample adequacy (KMO=0.493) and for sphericity (Bartlett's test $p<0.001$). From the 5 components, two composite measures which best defined joint attention behaviours were chosen from the results of the PCA analysis: Responding to Joint Attention (RJA) and Initiating Joint Attention (IJA). Subsequently, based on the composites scores, participants were divided into groups: responders to RJA, non-responders to RJA, responders IJA, and non-responders to IJA, whereby infants with scores of 0 on the Vineland were classified as non-responders and scores on the AOSI >1 were classified as non-responders.

Data from the non-responder groups were analyzed using Binomial Logistic Regression. The dependent variable was later ASD diagnosis. The independent variables were cortical (grey, white matter) and subcortical volumes (thalamus, hippocampus, amygdala, nucleus accumbens, ventral diencephalon, cerebellum), controlling for age, sex and total cerebral volumes. As we had two hypotheses for our aim regarding the predictive ability of volumes and later ASD diagnosis in RJA and IJA non-responders, alpha level was set to $p=0.05/2$ or $p<0.03$ using the Bonferroni correction method.

## 2.2 Results

### 2.2.1 Joint Attention Composites

The PCA analysis of the behavioural data revealed 5 components, which can be found in Table 1. Two composite scores that best described Joint Attention were chosen. Three components that described visual tracking and auditory processing were excluded. The first composite score included items related to eye gaze and shared affect (i.e., eye contact score and social interest and shared affect score), which are aspects related to Responding to Joint Attention. The second composite score, Initiating Joint Attention, included questions related to pointing and initiating JA (i.e., points to object he or she wants that is out of reach; points or gestures to indicate preference when offered a choice).

**Table 1** *Principal components analysis of gaze data*

| | | Eigenvalues | | |
|---|---|---|---|---|
| | Questions | Total | % of Variance | Cumulative % |
| Component 1 | Eye contact score<br>Social interest and shared affect score | 1.977 | 17.969 | 17.969 |
| Component 2 | Looks at face of parent or caregiver.<br>Watches (that is, follows with eyes) someone moving by crib or bed for 5 seconds or more. | 1.811 | 16.463 | 34.432 |
| Component 3 | Turns eyes and head toward sound.<br>Looks toward parent or caregiver when hearing parent's or caregiver's voice. | 1.708 | 15.526 | 49.959 |
| Component 4 | Points to object he or she wants that is out of reach.<br>Points or gestures to indicate preference when offered a choice (for example: "Do you want this one or that one?" etc.) | 1.52 | 13.82 | 63.779 |
| Component 5 | Points to common objects in a book or magazine as they are named (for example, dog, car, cup, key, etc.).<br>Makes sounds or gestures (for example, waves arms) to get parent's or caregivers' attention.<br>Coordination of eye gaze and action score | 1.085 | 9.868 | 73.646 |

*Note*. Principal components analysis applied to questionnaire items from the Vineland Adaptive Behavior Scales, 2nd edition (VABS-II) and the Autism Observation Scale for Infants (AOSI). Questionnaire items from component 1 were used to create the responding to joint attention grouping variable (RJA). Questionnaire items from component 4 were used for the initiating joint attention grouping variable (IJA).

### 2.2.2 Responders and non-responders

From the two composites' scores, IJA and RJA, infants were classified as IJA responders, IJA non-responders, RJA responders, and RJA non-responders. Data from 6 participants were excluded for IJA group (n=101) and from 11 participants for RJA group (n=96) as behavioural assessments were completed after 9 months of age. Data revealed that for the IJA scores (n=101), the majority of the infants were IJA non-responders (n=93, 92%), while approximately half of the sample were RJA non-responders (n=50, 52%).

### 2.2.3 Responding to Joint Attention (RJA), Initiating Joint Attention (IJA), brain volume and ASD diagnosis

Data from the IJA non-responder group (n=93) was analyzed using a regression method. In the Binary logistic regression analysis, subcortical volumes for the thalamus, ventral diencephalon, hippocampus, basal ganglia, and amygdala were entered as predictors in the model, and ASD diagnosis was used as the outcome variable, controlling for age, sex, and total cerebral volumes. The omnibus test of model coefficient (p=0.049) and the Hosmer and Lemeshow goodness of fit test (p=0.211) demonstrated our model was appropriate. The model performed well and correctly classified 87.1% of ASD diagnoses. From the subcortical volumes, left-hippocampus was a significant predictor of ASD diagnosis (B=-0.009, aOR=0.991, p=0.025). The association between hippocampal volumes and ASD diagnosis in the non-responder IJA group is shown in Figure 2. The right-thalamus was also a significant predictor of ASD (B=-0.016, aOR=0.984, p=0.026), as well as the left-thalamus (B=0.015, aOR=1.015, p=0.019). Age was not significant in the model (p>0.05) and sex was borderline (B=2.315, OR=10.122, p=0.052). No significant associations were evident amongst the subcortical volumes and ASD diagnosis (all, p>0.03) for the RJA non-responder group.

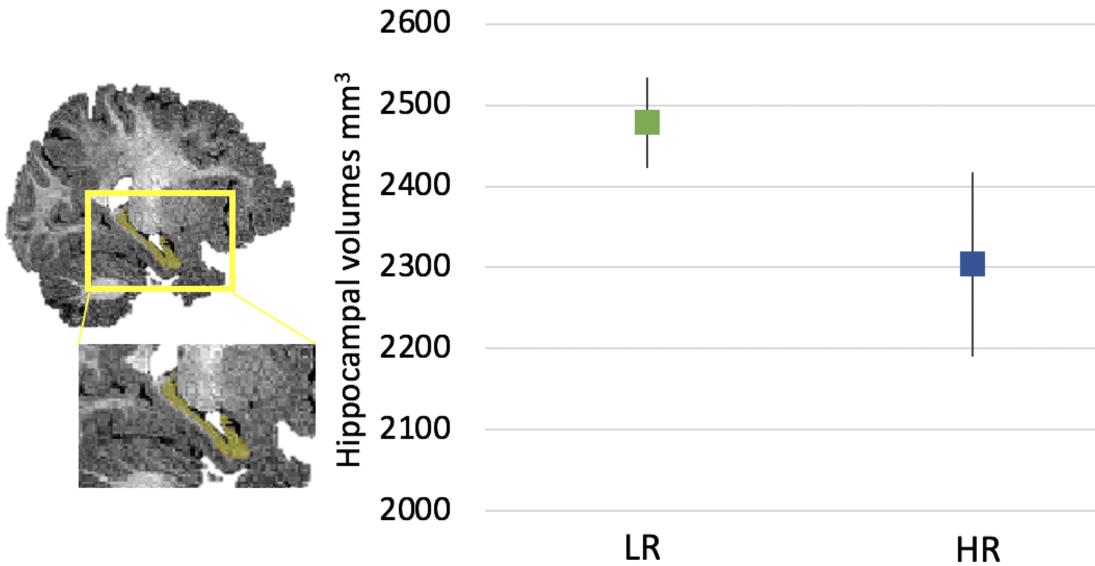


**Figure 2.** *The association between hippocampal volumes and ASD diagnosis in the non-responder IJA group who were low (LR) or high risk (HR) for the later development of ASD.*

**2.3 Discussion**

In the current study, we examined JA in young infants who had an increased familial likelihood for ASD. We used a data-driven approach to identify core constructs for IJA and RJA using clinical assessments. We identified that most infants were not-responding to IJA, while a larger proportion of infants engaged in RJA. Overall, in the IJA non-responder group, hippocampal and thalamic volumes were predictive of a later ASD diagnosis, indicating that these brain regions may show enhanced vulnerability early in life and may be key predictors of development of ASD. In the RJA non-responder group, no association between brain volumes and later ASD diagnosis was evident. Findings indicate that the absence of IJA behaviours may be associated with early changes in brain development, which are later associated with an ASD diagnosis.

Atypical JA behaviours have previously been reported in infants who were high risk for developing ASD compared to typically developing infants (Ibanez et al., 2013). Although differences in both IJA and RJA abilities have been observed in previous studies, the absence of IJA responses were more prominent, suggesting that IJA is a better predictor of later ASD diagnosis and symptomatology than RJA (Charman, 2003; Gangi et al., 2014; Ibanez et al., 2013). IJA behaviours require not just the ability to follow gaze, but also the infant's motivation to share interest or affect with others (Dawson et al., 2004; Gangi et al., 2014; Mundy, 2018). In this sense, IJA requires social information processing in a complex manner, having the infant taking a more active role in IJA than in RJA (Mundy, 2018). The infant shifts from being a signal-receiver when displaying RJA behaviours to being a signal-sender in IJA, and motivation to engage in social interactions is required (Mundy, 2018). In our study, we found that the majority of infants were IJA non-responders. In the IJA non-responder group, thalamus and hippocampal volumes were predictive of ASD diagnosis, while no association was evident between brain volumes and diagnosis in the non-responder RJA group.

Previous literature indicates that infants with a greater likelihood of developing ASD show deficits in communicative and social functioning, and in joint attention (Dawson, 2002; Ibanez et al., 2013; Zwaigenbaum et al., 2005). A study by Ozonoff et al. (2011) reported that only 19% of this population will later be diagnosed with ASD. As IJA requires more skills to be able to process different sources of information (e.g., their own position in space and position of others, direction of gaze, sensory information, emotional or affective information of others etc.), it is considered a more complex ability than RJA (Mundy, 2018). In this sense, IJA requires the infant not just to master those skills, but also to play an active role in engaging with others. That difference, in terms of the complexity of the behaviour, can be hypothesized as a key contributor to higher percentage of IJA non-responders compared to RJA.

Impairments in early joint attention have been identified as one of the earliest indicators of later ASD development (Bottema-Beutel, 2016; Bruinsma et al., 2004; Charman, 2003; Dawson et al., 2004; Ibanez et al., 2013; Werner, 2005). Previous studies have found that infants are sensitive to the gaze of others, even from the first days after birth, and prefer faces that engage in mutual gaze (Farroni, 2002; Guellai et al., 2020). However, infants and young children later diagnosed with ASD tend to prefer non-social stimuli rather than social ones (Chawarska et al., 2013; Di Giorgio et al., 2016; Gale et al., 2019; Peltola et al., 2018; Pierce et al., 2011), and fail to spontaneously orient to the social situation in the environment (Dawson et al., 2004).The failure to orient to social stimuli early in life could possibly lead to impairments in engaging in IJA, and consequently, to later social and communication impairments commonly observed in individuals with ASD (Dawson et al., 2004). The lack of early propensity to spontaneously engage in IJA with others might hinder opportunities to build and strengthen brain networks necessary for developing social cognition, contributing to a variety of social and language impairments. High-risk infants who do not respond spontaneously to social situations and struggle with early social gaze and JA engagement may be more likely to be diagnosed with ASD.

In our study, infants who were IJA non-responders and who had smaller thalamic and hippocampal volumes were more likely to later be diagnosed with ASD. The thalamus plays a critical role in the early specialization of the neocortex (Nair et al., 2021). Smaller thalamic volumes have been reported in children diagnosed with ASD compared to control groups (Sussman et al., 2015; Tamura et al., 2010). Evidence from infant and child studies suggests that altered thalamocortical connectivity is associated with ASD symptomatology (Chen et al., 2016; Iidaka et al., 2019; Nair et al., 2015; Nair et al., 2013). Early alterations in thalamic development and its connectivity have been reported in 6-week-old infants who were high risk for the development of ASD (Nair et al., 2021). Additionally, in a sample of young children and adolescents with ASD, structural alterations in thalamocortical pathways were associated with social communication impairments, along with repetitive behaviours (Nair et al., 2015). Structural alterations in the hippocampus have also been reported in individuals with ASD (Barnea-Goraly et al., 2014; Sussman et al., 2015). The thalamus is a relay center that receives sensory periphery information, such as visual and auditory information, and sends it to the cortex (Chen et al., 2016; Fu et al., 2019; Nair et al., 2015). An infant’s environment presents a variety of sensory information that is processed in their brains. To produce coherent perceptual representations and adequate behaviour, this information must be perfectly integrated (Murray et al., 2016; Stevenson et al., 2014). Impairments in sensory function and processing have been observed in individuals with ASD, and the failure to integrate sensory information coming from various sources put those individuals at risk for navigating the social world (Baum et al., 2015; Stevenson et al., 2014). Social and language cues come from different sensory inputs; in turn, integration of multisensory information plays an important role in social and communication function (Baum et al., 2015). In previous studies, alterations in thalamus-temporal cortex connectivity were associated with language and communication impairments observed in ASD (Chen et al., 2016).

The hippocampus is an essential brain region for learning and memory but has also been implicated in shared attention (Anand & Dhikav, 2012; Nummenmaa & Calder, 2009). In turn, early alterations in the development of these structures in high-risk infants may contribute to impairments with social gaze processing that require active engagement with others.

In our study, we found volumetric differences in the thalamus in infants who later developed ASD, a complex brain structure associated with filtering a variety of sensory information. Previous studies have found increased connectivity between the thalamus and sensory networks in individuals diagnosed with ASD compared to control groups (Fu et al., 2019). Lewis et al. (2017) suggested that alterations in brain networks in ASD individuals are present early in development, especially in low-level sensory processing, rather than higher-level cognitive processes. It was suggested that the deficits in social communication skills in ASD, which are higher-level cognitive processes, have a cascade effect due to deficits in lower-level processing of sensory information (Baum et al., 2015; Lewis et al., 2017).

In our study, we found that thalamic volumes predicted ASD diagnosis in high-risk infants who were not responding to IJA. Previous studies have found atypical thalamic connectivity and suggest this may contribute to impairments in orienting to social information in high-risk infants (Nair et al., 2021). Alterations in thalamic-prefrontal connectivity have been associated with diminished social attention and engagement in high-risk infants, and alterations in thalamic-occipital networks were associated with ASD symptomatology (Nair et al., 2015; Nair et al., 2021). These regions have been associated with the development of social cognition, as well as with processing visual information (Nair et al., 2015; Nair et al., 2021). It is possible that our findings related to alterations in thalamic volumes in high-risk infants underlie the atypical social development in this population and explain why we found that the majority of infants were non responders to IJA.

Infants who later develop ASD also fail to orient to social stimuli (Dawson et al., 2004) and one explanation for that could be rooted in the social motivation theory of ASD (Dawson et al., 2004; Mundy & Crowson, 1997; Tomasello et al., 2005). It has been observed that children with ASD lack the coordination to respond to joint attention but particularly display few IJA behaviours (Tomasello et al., 2005). This major deficit in IJA skills suggests that ASD children may lack motivation for sharing interests and emotions with others (Tomasello et al., 2005). Social motivation is crucial for IJA, which could explain the greater impairments in IJA rather than in RJA we have reported. It is possible that infants at risk for developing ASD do not experience the social interaction and sharing affect as a reward for continuing to seek interaction throughout their development. These abnormalities in reward neurological systems might explain the failure to attribute reward to social interactions (Dawson et al., 2004; Mundy, 2018). Activation of other associated areas of the brain could be associated with motivation and social reward such as the amygdala, the striatum and the hippocampus (Gordon et al., 2013). Previous studies found that IJA behaviours increase activation in brain areas related to reward, such as the striatum and the hippocampus (Schilbach, 2010). Altered hippocampal volumes, as found in our study, may be associated with atypical reward pathways in the brain. Some individuals with ASD may not process social interactions as rewarding, which in turn results in diminished social motivation. This lack of motivation to engage with others could have led to the impairments in IJA observed in our study. The hippocampus might show a greater vulnerability early in life and may be associated with the lack of social motivation in infants who later develop ASD.

Having a sample composed exclusively of infants with autistic siblings, and thus a familial risk of developing ASD, for a longitudinal study is rare, yet further investigation of heterogenous samples is needed to support our findings. As our sample was composed exclusively of infants who had an elevated risk of developing ASD, we cannot presume the same associations between brain volumes, JA, and later ASD diagnosis exist in typically-developing infants who are at low risk for an ASD diagnosis. As IJA is developmentally sensitive and is first exhibited around 3 months of age, it is possible that these behaviours could not had been completely developed at 4 to 6 months of age in some participants, which might have impacted our findings. Investigation of the development of JA over longer developmental periods than we were able to include are necessary to determine whether the trajectory of JA behaviours in later infancy (>9 months) show similar associations that we have reported in the current work.

### 2.3.1 Conclusions

In sum, we examined the association between subcortical brain volumes, behavioural measures of joint attention, and ASD development in at-risk infants. Using a data driven method, we identified constructs related to both IJA and RJA. We found that the vast majority of infants in our sample were non-responders to IJA, and from this group, we found that hippocampal and thalamic volumes predicted later ASD diagnosis. These findings suggest that these brain regions may have enhanced vulnerability early in life and may be key predictors of ASD development in infants who are high risk. A better understanding of early signs of social gaze and joint attention, as well as the neural mechanisms behind those behaviours, could help identify targets for intervention as well as biomarkers to promote improved social and behavioural outcomes in infants who are high risk for the development of ASD.

## 2.4 References

Adamson, L. B., Bakeman, R., Suma, K., & Robins, D. L. (2019). An Expanded View of Joint Attention: Skill, Engagement, and Language in Typical Development and Autism. *Child Dev*, *90*(1), e1-e18. https://doi.org/10.1111/cdev.12973

American Psychiatric Association. (2013). *Diagnostic and statistical manual of mental disorders : DSM-5* (5th ed.). American Psychiatric Association.

Anand, K. S., & Dhikav, V. (2012). Hippocampus in health and disease: An overview. *Ann Indian Acad Neurol*, *15*(4), 239-246. https://doi.org/10.4103/0972-2327.104323

Barnea-Goraly, N., Frazier, T. W., Piacenza, L., Minshew, N. J., Keshavan, M. S., Reiss, A. L., & Hardan, A. Y. (2014). A preliminary longitudinal volumetric MRI study of amygdala and hippocampal volumes in autism. *Prog Neuropsychopharmacol Biol Psychiatry*, *48*, 124-128. https://doi.org/10.1016/j.pnpbp.2013.09.010

Baum, S. H., Stevenson, R. A., & Wallace, M. T. (2015). Behavioral, perceptual, and neural alterations in sensory and multisensory function in autism spectrum disorder. *Prog Neurobiol*, *134*, 140-160. https://doi.org/10.1016/j.pneurobio.2015.09.007

Billeci, L., Narzisi, A., Tonacci, A., Sbriscia-Fioretti, B., Serasini, L., Fulceri, F., . . . Muratori, F. (2017). An integrated EEG and eye-tracking approach for the study of responding and initiating joint attention in Autism Spectrum Disorders. *Sci Rep*, *7*(1), 13560. https://doi.org/10.1038/s41598-017-13053-4

Bottema-Beutel, K. (2016). Associations between joint attention and language in autism spectrum disorder and typical development: A systematic review and meta-regression analysis. *Autism Res*, *9*(10), 1021-1035. https://doi.org/10.1002/aur.1624

Bruinsma, Y., Koegel, R. L., & Koegel, L. K. (2004). Joint attention and children with autism: a review of the literature. *Ment Retard Dev Disabil Res Rev*, *10*(3), 169-175. https://doi.org/10.1002/mrdd.20036

Bryson, S. E., & Zwaigenbaum, L. (2014). Autism Observation Scale for Infants. In *Comprehensive Guide to Autism* (pp. 299-310). https://doi.org/10.1007/978-1-4614-4788-7_12

Bryson, S. E., Zwaigenbaum, L., McDermott, C., Rombough, V., & Brian, J. (2008). The Autism Observation Scale for Infants: scale development and reliability data. *J Autism Dev Disord*, *38*(4), 731-738. https://doi.org/10.1007/s10803-007-0440-y

Carpenter, M., Nagell, K., & Tomasello, M. (1998). Social cognition, joint attention, and communicative competence from 9 to 15 months of age. *Monogr Soc Res Child Dev*, *63*(4), i-vi, 1-143.

Charman, T. (2003). Why is joint attention a pivotal skill in autism? *Philos Trans R Soc Lond B Biol Sci*, *358*(1430), 315-324. https://doi.org/10.1098/rstb.2002.1199

Chawarska, K., Macari, S., & Shic, F. (2013). Decreased spontaneous attention to social scenes in 6-month-old infants later diagnosed with autism spectrum disorders. *Biol Psychiatry*, *74*(3), 195-203. https://doi.org/10.1016/j.biopsych.2012.11.022

Chen, H., Uddin, L. Q., Zhang, Y., Duan, X., & Chen, H. (2016). Atypical effective connectivity of thalamo-cortical circuits in autism spectrum disorder. *Autism Res*, *9*(11), 1183-1190. https://doi.org/10.1002/aur.1614

Community-University Partnership for the Study of Children, Y. a. F. (2012). *Early Childhood Measurement and Evaluation Tool Review*.

Dawson, G., Toth, K., Abbott, R., Osterling, J., Munson, J., Estes, A., & Liaw, J. (2004). Early social attention impairments in autism: social orienting, joint attention, and attention to distress. *Dev Psychol*, *40*(2), 271-283. https://doi.org/10.1037/0012-1649.40.2.271

Dawson, G., Webb, S., Schellenberg, G. D., Dager, S., Friedman, S., Aylward, E., Richards, T., . (2002). Defining the broader phenotype of autism: Genetic, brain, and behavioral perspectives. *Development and Psychopathology*, *14*, 581–611. https://doi.org/DOI: 10.1017.S0954579402003103

de Macedo Rodrigues, K., Ben-Avi, E., Sliva, D. D., Choe, M. S., Drottar, M., Wang, R., . . . Zollei, L. (2015). A FreeSurfer-compliant consistent manual segmentation of infant brains spanning the 0-2 year age range. *Front Hum Neurosci*, *9*, 21. https://doi.org/10.3389/fnhum.2015.00021

Desikan, R. S., Segonne, F., Fischl, B., Quinn, B. T., Dickerson, B. C., Blacker, D., . . . Killiany, R. J. (2006). An automated labeling system for subdividing the human cerebral cortex on MRI scans into gyral based regions of interest. *Neuroimage*, *31*(3), 968-980. https://doi.org/10.1016/j.neuroimage.2006.01.021

Di Giorgio, E., Frasnelli, E., Rosa Salva, O., Scattoni, M. L., Puopolo, M., Tosoni, D., . . . Vallortigara, G. (2016). Difference in Visual Social Predispositions Between Newborns at Low- and High-risk for Autism. *Sci Rep*, *6*, 26395. https://doi.org/10.1038/srep26395

Emery, N. J. (2000). The eyes have it: the neuroethology, function and evolution of social gaze. *Neuroscience and Biobehavioral Reviews*, *24*, 581-604.

Farroni, T., Csibra, G., Simion, F., Johnson, M. H. . (2002). Eye contact detection in humans from birth. *PNAS*, *99*(14), 9602–9605. https://doi.org/www.pnas.org/cgi/doi/10.1073/pnas.152159999

Fischl, B. (2012). FreeSurfer. *Neuroimage*, *62*(2), 774-781. https://doi.org/10.1016/j.neuroimage.2012.01.021

Fischl, B., Salat, D. H., Busa, E. Albert. M., Dieterich, M., Haselgrove, C., van der Kouwe, A., Killiany, R., Kennedy, K., Klaveness, S., Montillo, A., Makris, N., Rosen, B., Dale, A. M. (2002). Whole Brain Segmentation: Automated Labeling of Neuroanatomical Structures in the Human Brain. *Neuron*, *33*, 341–355.

Fu, Z., Tu, Y., Di, X., Du, Y., Sui, J., Biswal, B. B., . . . Calhoun, V. D. (2019). Transient increased thalamic-sensory connectivity and decreased whole-brain dynamism in autism. *Neuroimage*, *190*, 191-204. https://doi.org/10.1016/j.neuroimage.2018.06.003

Gale, C. M., Eikeseth, S., & Klintwall, L. (2019). Children with Autism show Atypical Preference for Non-social Stimuli. *Sci Rep*, *9*(1), 10355. https://doi.org/10.1038/s41598-019-46705-8

Gangi, D. N., Ibanez, L. V., & Messinger, D. S. (2014). Joint attention initiation with and without positive affect: risk group differences and associations with ASD symptoms. *J Autism Dev Disord*, *44*(6), 1414-1424. https://doi.org/10.1007/s10803-013-2002-9

Gong, T., & Shuai, L. (2012). Modelling the coevolution of joint attention and language. *Proc Biol Sci*, *279*(1747), 4643-4651. https://doi.org/10.1098/rspb.2012.1431

Gordon, I., Eilbott, J. A., Feldman, R., Pelphrey, K. A., & Vander Wyk, B. C. (2013). Social, reward, and attention brain networks are involved when online bids for joint attention are met with congruent versus incongruent responses. *Soc Neurosci*, *8*(6), 544-554. https://doi.org/10.1080/17470919.2013.832374

Guellai, B., Hausberger, M., Chopin, A., & Streri, A. (2020). Premises of social cognition: Newborns are sensitive to a direct versus a faraway gaze. *Sci Rep*, *10*(1), 9796. https://doi.org/10.1038/s41598-020-66576-8

Haith, M. M., Bergman, T., Moore. M. J., . (1977). Eye Contact and Face Scanning in Early Infancy. *Science*, *198*(4319), 853-855.

Ibanez, L. V., Grantz, C. J., & Messinger, D. S. (2013). The Development of Referential Communication and Autism Symptomatology in High-Risk Infants. *Infancy*, *18*(5). https://doi.org/10.1111/j.1532-7078.2012.00142.x

Iidaka, T., Kogata, T., Mano, Y., & Komeda, H. (2019). Thalamocortical Hyperconnectivity and Amygdala-Cortical Hypoconnectivity in Male Patients With Autism Spectrum Disorder. *Front Psychiatry*, *10*, 252. https://doi.org/10.3389/fpsyt.2019.00252

Lewis, J. D., Evans, A. C., Pruett, J. R., Jr., Botteron, K. N., McKinstry, R. C., Zwaigenbaum, L., . . . Infant Brain Imaging Study, N. (2017). The Emergence of Network Inefficiencies in Infants With Autism Spectrum Disorder. *Biol Psychiatry*, *82*(3), 176-185. https://doi.org/10.1016/j.biopsych.2017.03.006

Lord, C., Risi, S., Lambrecht, L., Cook, E. H., Leventhal, B. L., DiLavore, P. C., ... & Rutter, M. (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), 205-223.

Mosconi, M. W., Cody-Hazlett, H., Poe, M. D., Gerig, G., Gimpel-Smith, R., & Piven, J. (2009). Longitudinal study of amygdala volume and joint attention in 2- to 4-year-old children with autism. *Arch Gen Psychiatry*, *66*(5), 509-516. https://doi.org/10.1001/archgenpsychiatry.2009.19

Mundy, P. (2018). A review of joint attention and social-cognitive brain systems in typical development and autism spectrum disorder. *Eur J Neurosci*, *47*(6), 497-514. https://doi.org/10.1111/ejn.13720

Mundy, P., & Crowson, M. (1997). Joint attention and early social communication: implications for research on intervention with autism. *J Autism Dev Disord*, *27*(6), 653-676.

Murray, M. M., Lewkowicz, D. J., Amedi, A., & Wallace, M. T. (2016). Multisensory Processes: A Balancing Act across the Lifespan. *Trends Neurosci*, *39*(8), 567-579. https://doi.org/10.1016/j.tins.2016.05.003

Nair, A., Carper, R. A., Abbott, A. E., Chen, C. P., Solders, S., Nakutin, S., . . . Muller, R. A. (2015). Regional specificity of aberrant thalamocortical connectivity in autism. *Hum Brain Mapp*, *36*(11), 4497-4511. https://doi.org/10.1002/hbm.22938

Nair, A., Jalal, R., Liu, J., Tsang, T., McDonald, N. M., Jackson, L., . . . Dapretto, M. (2021). Altered Thalamocortical Connectivity in 6-Week-Old Infants at High Familial Risk for Autism Spectrum Disorder. *Cereb Cortex, 31*(9), 4191-4205. https://doi.org/10.1093/cercor/bhab078

Nair, A., Treiber, J. M., Shukla, D. K., Shih, P., & Muller, R. A. (2013). Impaired thalamocortical connectivity in autism spectrum disorder: a study of functional and anatomical connectivity. *Brain*, *136*(Pt 6), 1942-1955. https://doi.org/10.1093/brain/awt079

Nummenmaa, L., & Calder, A. J. (2009). Neural mechanisms of social attention. *Trends Cogn Sci*, *13*(3), 135-143. https://doi.org/10.1016/j.tics.2008.12.006

Ofner, M., Coles, A., Decou, M. L., Do, M. T., Bienek, A., Snider, J., Ugnat, A. M. (2018). *Autism Spectrum Disorder among children and youth in Canada 2018: A Report of the National Autism Spectrum Disorder Surveillance System* (1st ed.). Public Health Agency of Canada.

Ozonoff, S., Young, G. S., Carter, A., Messinger, D., Yirmiya, N., Zwaigenbaum, L., . . . Stone, W. L. (2011). Recurrence risk for autism spectrum disorders: a Baby Siblings Research Consortium study. *PEDIATRICS*, *128*(3), e488-495. https://doi.org/10.1542/peds.2010-2825

Payakachat, N., Tilford, J. M., & Ungar, W. J. (2016). National Database for Autism Research (NDAR): Big Data Opportunities for Health Services Research and Health Technology Assessment. *Pharmacoeconomics*, *34*(2), 127-138. https://doi.org/10.1007/s40273-015-0331-6

Peltola, M. J., Yrttiaho, S., & Leppanen, J. M. (2018). Infants' attention bias to faces as an early marker of social development. *Dev Sci*, *21*(6), e12687. https://doi.org/10.1111/desc.12687

Pierce, K., Conant, D., Hazin, R., Stoner, R., & Desmond, J. (2011). Preference for geometric patterns early in life as a risk factor for autism. *Arch Gen Psychiatry*, *68*(1), 101-109. https://doi.org/10.1001/archgenpsychiatry.2010.113

Schilbach, L., Wilms, M., Eickhoff, S.B., Romanzetti, S., Tepest, R., Bente, G. Jon Shah, N. J., Fink, G. R., Vogeley, K. . (2010). Minds made for sharing: initiating joint attention recruits reward-related neurocircuitry. *Journal of Cognitive Neuroscience*, *22*(12), 2702–2715.

Sparrow, S. S. (2011). Vineland Adaptive Behavior Scales. In *Encyclopedia of Clinical Neuropsychology* (pp. 2618-2621). https://doi.org/10.1007/978-0-387-79948-3_1602

Stevenson, R. A., Siemann, J. K., Woynaroski, T. G., Schneider, B. C., Eberly, H. E., Camarata, S. M., & Wallace, M. T. (2014). Evidence for diminished multisensory integration in autism spectrum disorders. *J Autism Dev Disord*, *44*(12), 3161-3167. https://doi.org/10.1007/s10803-014-2179-6

Sussman, D., Leung, R. C., Vogan, V. M., Lee, W., Trelle, S., Lin, S., . . . Taylor, M. J. (2015). The autism puzzle: Diffuse but not pervasive neuroanatomical abnormalities in children with ASD. *Neuroimage Clin*, *8*, 170-179. https://doi.org/10.1016/j.nicl.2015.04.008

Swanson, M. R., Serlin, G. C., & Siller, M. (2013). Broad autism phenotype in typically developing children predicts performance on an eye-tracking measure of joint attention. *J Autism Dev Disord*, *43*(3), 707-718. https://doi.org/10.1007/s10803-012-1616-7

Tamura, R., Kitamura, H., Endo, T., Hasegawa, N., & Someya, T. (2010). Reduced thalamic volume observed across different subgroups of autism spectrum disorders. *Psychiatry Res*, *184*(3), 186-188. https://doi.org/10.1016/j.pscychresns.2010.07.001

Tomasello, M., Carpenter, M., Call, J., Behne, T., & Moll, H. (2005). Understanding and sharing intentions: the origins of cultural cognition. *Behav Brain Sci*, *28*(5), 675-691; discussion 691-735. https://doi.org/10.1017/S0140525X05000129

Torrico, T. J., Munakomi, S. . (2021). Neuroanatomy, Thalamus. *NCBI Bookshelf. A service of the National Library of Medicine, National Institutes of Health.*

Werner, E., Dawson, G., . (2005). Validation of the Phenomenon of Autistic Regression Using Home Videotapes. *Arch Gen Psychiatry*, *62*, 889-895.

Zollei, L., Iglesias, J. E., Ou, Y., Grant, P. E., & Fischl, B. (2020). Infant FreeSurfer: An automated segmentation and surface extraction pipeline for T1-weighted neuroimaging data of infants 0-2 years. *Neuroimage*, *218*, 116946. https://doi.org/10.1016/j.neuroimage.2020.116946

Zwaigenbaum, L. (2010). Advances in the early detection of autism. *Curr Opin Neurol*, *23*(2), 97-102. https://doi.org/10.1097/WCO.0b013e3283372430

Zwaigenbaum, L., Bryson, S., Rogers, T., Roberts, W., Brian, J., & Szatmari, P. (2005). Behavioral manifestations of autism in the first year of life. *Int J Dev Neurosci*, *23*(2-3), 143-152. https://doi.org/10.1016/j.ijdevneu.2004.05.001

# Chapter 3

## 3 Study outcomes

In this study, young infants who had an increased familial risk of developing ASD were more likely to be non-responders to initiating joint attention (IJA) compared with responding to joint attention (RJA). When analyzing brain volumes in both responders and non-responders to IJA groups, we found that hippocampal and thalamic volumes were predictive of a later ASD diagnosis in the IJA non-responder group. These findings also indicate that the absence of IJA behaviours may be associated with early changes in brain development that contribute to ASD diagnosis. Further, findings indicate that early in life, these brain regions may show enhanced vulnerability and may be key predictors of ASD development.

### 3.1 Implications

Early alterations in brain development in high-risk infants may contribute to impairments in social engagement with others and may be key predictors of the development of ASD. Identifying those infants at risk for the development of ASD very early in life can facilitate access to early interventions and promote improved outcomes. Although it is highly recognized that early signs can be detected in the first year of life, usually the diagnosis of ASD is not made until the child is at least 3 years of age, when socio-communication delays are more easily identified (American Psychiatric Association, 2013; Di Giorgio et al., 2016; Zwaigenbaum, 2010). Currently, the median age of diagnosis in Canada is above 5 years of age, according to the Public Health Agency of Canada report published in 2018 (Ofner, 2018). This same report shows that more than half of the ASD-diagnosed children had received that diagnosis by 6 years of age, and only a minority (<20%) had received a diagnosis by 3 years of age (Ofner, 2018). It is also common for children to be diagnosed after entering the school system, when concerns related to socio-communication skills are more evident (Vietze, 2020). The delayed detection and diagnosis of those infants or children could hinder the opportunities for accessing appropriate care in a timely manner. Identifying infants who struggle with social interactions early in life could help identify targets for intervention, as well as biomarkers to promote better social and behavioural outcomes, even before these children start school.

Infants who show impairments in early JA skills, especially in initiating shared attention with others (IJA) may be at higher risk for developing ASD later in life. JA abilities are associated with language acquisition and with transitioning to symbolic communication, which can be significantly impaired in children diagnosed with ASD (Bottema-Beutel, 2016; Bruinsma et al., 2004). Therapies that focus on the improvement of JA in children diagnosed with ASD have been associated with greater improvements in language development (Bono, 2004). It has been recognized that JA abilities begin in early infancy and tend to solidify during the second year of life (Carpenter et al., 1998). Given the importance of JA in social development and communication, early therapies that focus on JA may be a key focus of intervention for flagged infants to develop better outcomes. Previous research suggested that JA is a pivotal skill when measuring the effects of an intervention (Bono, 2004). These findings suggested that JA may be an important target of intervention for those infants who experiencing impairments in social interactions early in life.

Some of the therapies that focus on developing and increasing JA skills are behavioural interventions, speech communication therapy, and structured educational interventions, among others (Bono, 2004). Bono (2004) found an association between better JA skills and enhanced language development in young children diagnosed with ASD, regardless of the type of intervention. Other studies found that interventions targeting JA skills improved language outcomes; these studies confirmed that these interventions also improved shared attention initiated by the child (IJA) in young children diagnosed with ASD (Kasari et al., 2006; Kasari et al., 2008). These findings highlight the importance of focusing on JA skills as a target of intervention.

One therapy commonly used for social communication and learning skills in children with ASD is Applied Behavioural Analysis (ABA). Previous studies found that community-based ABA interventions are associated with improvements in socio-communication skills, as well as with adaptive behaviour in toddlers with suspected developmental delay, including those diagnosed with ASD (Vietze, 2020). Also based on the ABA model, the Early Start Denver Model (ESDM) combines ABA with developmental and relationship-based approaches (Dawson et al., 2010). Currently, this intervention can be offered for preschool-aged children as early as 12 months of age and has been associated with improvements in IQ, language, and adaptive behaviour, as well as with diminished severity in ASD symptomatology in children aged 18-30 months (Dawson et al., 2010). ESDM uses teaching techniques that comprise aspects of JA, such as shared affect and engagement with others (Dawson et al., 2010). The Canadian Paediatric Society recommends these therapeutic approaches including ABA, which focus on affective engagement between the child and caregiver/therapist, because of its role in developing social relationships (Ip et al., 2019). Also, more naturalistic approaches that are delivered in a natural environment using materials that are familiar to the child have been recommended for very young children at risk for ASD (Ip et al., 2019; Schreibman, 2015).

Even though it is widely accepted that interventions should be accessed as early as possible, even before the diagnosis is made (Ip et al., 2019), there is still a gap in early interventions for young children and infants (Shonkoff, 2003; Vietze, 2020). According to Landa (2018), the age of interventions has been considered a significant predictor of social communication outcomes across multiple studies. However, access to early interventions is often impacted by inconsistent criteria in defining who is eligible to receive those services (Landa, 2018). Based on our findings and on the extensive literature that indicates brain alterations behind the early social impairments, it seems reasonable that early detection of signs that could indicate later ASD development should be paired with early ASD interventions, even before a conclusive diagnosis is made. In that way, infants could benefit from interventions as soon as social impairments have been identified.

Early interventions are also important to reduce barriers to learning, even before those children enter the school system. Research has shown that not only social engagement skills, but also social communication skills, are important factors for learning (Krstovska-Guerrero, 2016; Schreibman et al., 2015). It is now known that socio-emotional competence is key for cognitive and language development, emotional regulation, and strong peer relationships, all of which are foundational skills for academic success and, later, quality of life. Children start to develop those skills from birth in the context of positive interactions and engagement with others (Schreibman et al., 2015; Shonkoff, 2003). These skills are crucial for future school readiness (Shonkoff, 2003). Children diagnosed with ASD often struggle with early social interactions and communication, as well as with regulating their emotions, thereby placing these children at higher risk for having difficulties with learning. Early interventions can help these children to overcome developmental barriers and improve the outcomes when they reach school age and start in an educational setting (Landa, 2018). Improving social engagement early in life for infants who struggle with early social engagement can promote the development of a variety of other skills necessary for amplifying the repertoire of more complex forms of social cognition and, consequently, for learning (Landa, 2018).

Promoting interventions at an earlier age supports early social interactions that scaffold later social skills and can interrupt the cascade effects that result in atypical development (Landa, 2018). In this sense, as soon as early signs related to social communication delays are detected, early interventions should be provided (Landa, 2018). Early interventions can promote child learning and increase the development of social skills, thereby diminishing the effects of ASD symptoms in young children (Landa, 2018). Given that ASD is a neurodevelopmental disorder, interventions should be provided as early as possible to maximize the gains and alter developmental trajectories. Early intervention would also diminish the costs of treatment over time (Horlin, 2014; Zwaigenbaum, 2010). Those benefits extend not just to the individual, but to their families, as well as to society (Horlin, 2014).

### 3.2 Limitations and Future work

The current study was designed and implemented using data available by the National Database for Autism Research (NDAR) repository as part of the National Institute of Mental Health Data Archive (NDA) (Payakachat et al., 2016). This data is available for research and is used to advance the scientific knowledge related to ASD and its treatments (Payakachat et al., 2016). The available data made it possible to have access to a rare sample, composed exclusively of infants with autistic siblings; however, it was not possible to collect our data for this study. The behavioural data was collected also through parental reports and not only behavioural observation, which could have biased our results. The open access to the database for researchers made this study possible during the COVID-19 pandemic, when access to in-person data collection was highly restricted.

As our sample was composed exclusively of infants with an increased likelihood of developing ASD, we could not extend the same associations we found to typically developing infants. Given that JA is developmentally sensitive, it is possible that IJA behaviours were not completely developed by the time the data were collected. Future studies could potentially focus on the investigation of the development of JA over longer developmental periods, especially in later infancy (>9 months). Also, in the IJA composite, items related to pointing to objects were considered aspects that are related to initiation of shared attention; however, this behaviour could be considered Initiating Behavioral Request (IBR), which is less frequently discussed in the field of Joint Attention. This behaviour has also been associated with later ASD diagnosis as they also require motivation to share or request objects with others but is considered to require less motivation than IJA (Ibanez et al., 2013). Future studies should be more hypothesis-driven and would be necessary to determine whether the trajectory of JA behaviours in later infancy shows similar associations as we have reported in our investigations.

### 3.3 Final conclusions

In sum, the association between subcortical brain volumes, behavioural measures of joint attention, and ASD development in infants who had a familial risk of later developing ASD was examined. Using a machine learning approach applied to infants' behavioural data, key gaze actions for both IJA and RJA were identified. The vast majority of infants in the sample were non-responders to IJA, and from this group, it was found that hippocampal and thalamic volumes predicted later ASD diagnosis. Findings suggest that these brain regions may present increased vulnerability early in life and may be key predictors of the development of ASD. Integrating a better understanding of the behavioural signs of ASD development early in life and the neural mechanisms behind them could help identify targets for intervention, as well as biomarkers to promote improved social and behavioural outcomes in infants who are at high risk for the development of ASD.

### 3.4 References

American Psychiatric Association. (2013). *Diagnostic and statistical manual of mental disorders : DSM-5* (5th ed.). American Psychiatric Association.

Bono, M. A., Daley, T., Sigman, M. . (2004). Relations Among Joint Attention, Amount of Intervention and Language Gain in Autism. *Journal of Autism and Developmental Disorders*, *34*(5), 495-505.

Bottema-Beutel, K. (2016). Associations between joint attention and language in autism spectrum disorder and typical development: A systematic review and meta-regression analysis. *Autism Res*, *9*(10), 1021-1035. https://doi.org/10.1002/aur.1624

Bruinsma, Y., Koegel, R. L., & Koegel, L. K. (2004). Joint attention and children with autism: a review of the literature. *Ment Retard Dev Disabil Res Rev*, *10*(3), 169-175. https://doi.org/10.1002/mrdd.20036

Carpenter, M., Nagell, K., & Tomasello, M. (1998). Social cognition, joint attention, and communicative competence from 9 to 15 months of age. *Monogr Soc Res Child Dev*, *63*(4), i-vi, 1-143.

Dawson, G., Rogers, S., Munson, J., Smith, M., Winter, J., Greenson, J., . . . Varley, J. (2010). Randomized, Controlled Trial of an Intervention for Toddlers With Autism: The Early Start Denver Model. *Pediatrics*, *125*(1), e17-e23. https://doi.org/10.1542/peds.2009-0958

Di Giorgio, E., Frasnelli, E., Rosa Salva, O., Scattoni, M. L., Puopolo, M., Tosoni, D., . . . Vallortigara, G. (2016). Difference in Visual Social Predispositions Between Newborns at Low- and High-risk for Autism. *Sci Rep*, *6*, 26395. https://doi.org/10.1038/srep26395

Horlin, C., Falkmer, M., Parsons, R., Albrecht, M. A., Falkmer, T. (2014). The Cost of Autism Spectrum Disorders. *PLoS ONE*, *9*(9), e106552-e106552. https://doi.org/10.1371/journal. pone.0106552

Ibanez, L. V., Grantz, C. J., & Messinger, D. S. (2013). The Development of Referential Communication and Autism Symptomatology in High-Risk Infants. *Infancy*, *18*(5). https://doi.org/10.1111/j.1532-7078.2012.00142.x

Ip, A., Zwaigenbaum, L., & Brian, J. A. (2019). Post-diagnostic management and follow-up care for autism spectrum disorder. *Paediatrics & Child Health*, *24*(7), 461-468. https://doi.org/10.1093/pch/pxz121

Kasari, C., Freeman, S., & Paparella, T. (2006). Joint attention and symbolic play in young children with autism: a randomized controlled intervention study. *Journal of Child Psychology and Psychiatry*, *47*(6), 611-620. https://doi.org/10.1111/j.1469-7610.2005.01567.x

Kasari, C., Paparella, T., Freeman, S., & Jahromi, L. B. (2008). Language outcome in autism: randomized comparison of joint attention and play interventions. *J Consult Clin Psychol*, *76*(1), 125-137. https://doi.org/10.1037/0022-006X.76.1.125

Krstovska-Guerrero, I., Jones, E. A. (2016). Social-Communication Intervention for Toddlers with Autism Spectrum Disorder: Eye Gaze in the Context of Requesting and Joint Attention. *J Dev Phys Disabil*, *28*, 289–316. https://doi.org/10.1007/s10882-015-9466-9

Landa, R. J. (2018). Efficacy of early interventions for infants and young children with, and at risk for, autism spectrum disorders *INTERNATIONAL REVIEW OF PSYCHIATRY*, *30*(1), 25–39. https://doi.org/https://doi.org/10.1080/09540261.2018.1432574

Ofner, M., Coles, A., Decou, M. L., Do, M. T., Bienek, A., Snider, J., Ugnat, A. M. (2018). *Autism Spectrum Disorder among children and youth in Canada 2018: A Report of the National Autism Spectrum Disorder Surveillance System* (1st ed.). Public Health Agency of Canada.

Payakachat, N., Tilford, J. M., & Ungar, W. J. (2016). National Database for Autism Research (NDAR): Big Data Opportunities for Health Services Research and Health Technology Assessment. *Pharmacoeconomics*, *34*(2), 127-138. https://doi.org/10.1007/s40273-015-0331-6

Schreibman, L., Dawson, G., Stahmer, A. C., Landa, R., Rogers, S. J., Mcgee, G. G., . . . Halladay, A. (2015). Naturalistic Developmental Behavioral Interventions: Empirically Validated Treatments for Autism Spectrum Disorder. *Journal of Autism and Developmental Disorders*, *45*(8), 2411-2428. https://doi.org/10.1007/s10803-015-2407-8

Schreibman, L., Dawson, G., Stahmer, A. C., Landa, R., Rogers, S. J., McGee, G. G., Kasari, C., Ingersoll, B., Kaiser, A.. P., Bruinsma, Y., McNerney, E., Wetherby, E., Halladay, A. (2015). Naturalistic Developmental Behavioral Interventions: Empirically Validated Treatments for Autism Spectrum Disorder *J Autism Dev Disord*, *45*, 2411–2428. https://doi.org/DOI 10.1007/s10803-015-2407-8

Shonkoff, K. P. (2003). From Neurons to Neighborhoods: Old and New Challenges for Developmental and Behavioral Pediatrics *Developmental and Behavioral Pediatrics*, *24*(1), 70-76.

Vietze, P. L., L. E. (2020). Early Intervention ABA for Toddlers with ASD: Effect of Age and Amount. *Current Psychology*, *39*, 1234–1244. https://doi.org/10.1007/s12144-018-9812-z

Zwaigenbaum, L. (2010). Advances in the early detection of autism. *Curr Opin Neurol*, *23*(2), 97-102. https://doi.org/10.1097/WCO.0b013e3283372430

# 4 Appendices

## 4.1 Appendix A: *Vineland Adaptive Behavior Scales, 2nd edition (VABS-II) Items*

| Vineland Questions | | | |
|---|---|---|---|
| **Subject ID:** | | | |
| **Age in months at the time of the interview:** | | | |
| **Sex of subject at birth:** | | | |
| | Never | Sometimes /Partially | Usually |
| Turns eyes and head toward sound. | 0 | 1 | 2 |
| Looks toward parent or caregiver when hearing parent's or caregiver's voice. | 0 | 1 | 2 |
| Responds to his or her name spoken (for example, turns toward speaker, smiles, etc.). | 0 | 1 | 2 |
| Demonstrates understanding of the meaning of no, or word or gesture with the same meaning (for example, stops current activity briefly). | 0 | 1 | 2 |
| Demonstrates understanding of the meaning of yes, or word or gesture with the same meaning (for example, continues activity, smiles, etc.). | 0 | 1 | 2 |
| Listens to story for at least 5 minutes (that is, remains relatively still and directs attention to the storyteller or reader). | 0 | 1 | 2 |
| Points to at least three major body parts when asked (for example, nose, mouth, hands, feet, etc.). | 0 | 1 | 2 |
| Points to common objects in a book or magazine as they are named (for example, dog, car, cup, key, etc.). | 0 | 1 | 2 |
| Listens to instructions. | 0 | 1 | 2 |
| Follows instructions with one action and one object (for example, "Bring me the book"; "Close the door"; etc.). | 0 | 1 | 2 |
| Points to at least five minor body parts when asked (for example, fingers, elbows, teeth, toes, etc.). | 0 | 1 | 2 |
| Follows instructions with two actions or an action and two objects (for example, "Bring me the crayons and the paper"; "Sit down and eat your lunch"; etc.). | 0 | 1 | 2 |
| Follows instructions in "if-then" form (for example, "If you want to play outside, then put your things away."; etc.)." | 0 | 1 | 2 |
| Listens to a story for at least 15 minutes. | 0 | 1 | 2 |
| Listens to a story for at least 30 minutes. | 0 | 1 | 2 |
| Follows three-part instructions (for example "Brush your teeth get dressed and make your bed" etc.). | 0 | 1 | 2 |
| Follows instructions or directions heard 5 minutes before. | 0 | 1 | 2 |
| Understands sayings that are not meant to be taken word for word (for example, "Button your lip"; "Hit the road"; etc.). | 0 | 1 | 2 |
| Listens to an informational talk for at least 15 minutes. | 0 | 1 | 2 |
| Listens to an informational talk for at least 30 minutes. | 0 | 1 | 2 |
| Cries or fusses when hungry or wet. | 0 | 1 | 2 |
| Smiles when you smile at him or her. | 0 | 1 | 2 |
| Makes sounds of pleasure (for example, coos, laughs, etc.). | 0 | 1 | 2 |
| Makes nonword baby sounds (that is, babbles). | 0 | 1 | 2 |
| Makes sounds or gestures (for example, waves arms) to get parent's or care-givers attention. | 0 | 1 | 2 |
| Makes sounds or gestures (for example, shakes head) if he or she wants an activity to stop or keep going. | 0 | 1 | 2 |

| Vineland Questions | | | |
|---|---|---|---|
| **Subject ID:** | | | |
| **Age in months at the time of the interview:** | | | |
| **Sex of subject at birth:** | | | |
| | Never | Sometimes /Partially | Usually |
| Waves good-bye when another person waves or parent or caregiver tells him or her to wave. | 0 | 1 | 2 |
| Says "Da-da" "Ma-ma" or another name for parent or caregiver including parents or caregivers first name or nickname. | 0 | 1 | 2 |
| Points to object he or she wants that is out of reach. | 0 | 1 | 2 |
| Points or gestures to indicate preference when offered a choice (for example: "Do you want this one or that one?" etc.) | 0 | 1 | 2 |
| "Repeats or tries to repeat common words immediately upon hearing for them (for example, ball, car, go, etc.)" | 0 | 1 | 2 |
| Names at least three objects (for example, bottle, dog, favorite toy, etc.) | 0 | 1 | 2 |
| Says one-word requests (for example, up, more, out, etc.) | 0 | 1 | 2 |
| Uses first names or nicknames of brothers, sisters, or friends, or says their names when asked. | 0 | 1 | 2 |
| Answers or tries to answer with words when asked a question. | 0 | 1 | 2 |
| Names at least 10 objects. | 0 | 1 | 2 |
| States own first name or nickname (for example, Latesha, Little Sister, etc.) when asked. | 0 | 1 | 2 |
| Uses phrases with a noun and a verb (for example, "Katie stay"; "Go home"; etc.). | 0 | 1 | 2 |
| Asks questions by changing inflection of words or simple phrases (Mine?; Me go?; etc.). Grammar is not important. | 0 | 1 | 2 |
| Says at least 50 recognizable words. | 0 | 1 | 2 |
| Uses simple words to describe things (for example, dirty, pretty, big, loud, etc.). | 0 | 1 | 2 |
| Asks questions beginning with what or where (for example, "What's that?"; "Where doggie go?"; etc.). | 0 | 1 | 2 |
| Uses negatives in sentences (for example, "Me no go"'; "I won't drink it"; etc.); grammar is not important. | 0 | 1 | 2 |
| Tells about experiences in simple sentences (for example, "Ginger and I play"; "Dan read me a book"; etc.). | 0 | 1 | 2 |
| Says correct age when asked. | 0 | 1 | 2 |
| Says at least 100 recognizable words. | 0 | 1 | 2 |
| Uses in, on, or under in phrases or sentences (for example, "Ball go under chair"; "Put it on the table"; etc.). | 0 | 1 | 2 |
| Uses "and" in phrases or sentences (for example: "Mom and Dad"; "I want ice cream and cake"; etc.). | 0 | 1 | 2 |
| Says first and last name when asked. | 0 | 1 | 2 |
| Identifies and names most common colors (red, blue, green, yellow, orange, purple, brown, and black). | 0 | 1 | 2 |
| Asks questions beginning with who or why (for example, "Who's that?"; "Why do I have to go?"; etc.). | 0 | 1 | 2 |
| Uses present tense verbs ending in -ing (for example, "is singing"; "is playing"; etc.). | 0 | 1 | 2 |
| Uses possessives in phrases or sentences (for example, "That's her book"; "This is Carlos's ball"; etc.). | 0 | 1 | 2 |
| Uses pronouns in phrases or sentences; must use correct gender and form of the pronoun, but sentences need not be grammatically correct (for example, "He done it"; "They went"; etc.). | 0 | 1 | 2 |

| Vineland Questions | | | |
|---|---|---|---|
| **Subject ID:** | | | |
| **Age in months at the time of the interview:** | | | |
| **Sex of subject at birth:** | | | |
| | Never | Sometimes /Partially | Usually |
| Asks questions beginning with when (for example, "When is dinner?"; "When can we go home?"; etc.). | 0 | 1 | 2 |
| Uses regular past tense verbs (for example, walked, baked, etc.); may use irregular past tense verbs ungrammatically (for example, "I runned away"; etc.). | 0 | 1 | 2 |
| Uses behind or in front of in phrases or sentences (for example: "I walked in front of her"; "Terrell is behind you" etc.). | 0 | 1 | 2 |
| Pronounces words clearly without sound substitutions (for example, does not say wabbit for rabbit, Thally for Sally, etc.). | 0 | 1 | 2 |
| Tells basic parts of a story, fairy tale, or television show plot; does not need to include great detail or recount in perfect order. | 0 | 1 | 2 |
| Says month and day of birthday when asked. | 0 | 1 | 2 |
| Modulates tone of voice, volume, and rhythm appropriately (for example, does not consistently speak too loudly, too softly, or in a monotone, etc.). | 0 | 1 | 2 |
| Tells about experiences in detail (for example, tells who was involved, where activity took etc.). | 0 | 1 | 2 |
| Gives simple directions (for example, on how to play a game). | 0 | 1 | 2 |
| Uses between in phrases or sentences (for example, "The ball went between the cars"; etc.). | 0 | 1 | 2 |
| Says own telephone number when asked. | 0 | 1 | 2 |
| Easily moves from one topic to another in conversation. | 0 | 1 | 2 |
| Stays on topic in conversations; does not go off on tangents. | 0 | 1 | 2 |
| Explains ideas in more than one way (for example, "This was a good book. It was exciting and fun to read."; etc.). | 0 | 1 | 2 |
| Has conversations that last 10 minutes (for example, relates experiences, contributes ideas, shares etc.). | 0 | 1 | 2 |
| Uses irregular plurals correctly (for example, children, geese, mice, women, etc.). | 0 | 1 | 2 |
| Says complete home address (that is, street or rural route, apartment number, city, and state), with or without zip code, when asked. | 0 | 1 | 2 |
| Describes a short-term goal and what he or she needs to do to reach it (for example says "I want to get an A on my test so I'm going to study hard"; etc.). | 0 | 1 | 2 |
| Gives complex directions to others (for example, to a distant location). | 0 | 1 | 2 |
| Describes a realistic long-range goal that can be done in 6 months or more (for example: says "I want to buy a bike so I'll babysit and run errands to earn enough money to buy it"; etc.). | 0 | 1 | 2 |
| Identifies one or more alphabet letters as letters and distinguishes them from numbers. | 0 | 1 | 2 |
| Recognizes own name in printed form. | 0 | 1 | 2 |
| Identifies at least 10 printed letters of the alphabet. | 0 | 1 | 2 |
| Prints or writes using correct orientation (for example, in English from left to right; in some languages from right to left or top to bottom). | 0 | 1 | 2 |
| Copies own first name. | 0 | 1 | 2 |
| Identifies all printed letters of the alphabet, upper-and lowercase. | 0 | 1 | 2 |
| Prints at least three simple words from example (for example, cat, see, bee, etc.). | 0 | 1 | 2 |
| Prints or writes own first and last name form memory. | 0 | 1 | 2 |

| Vineland Questions | | | |
|---|---|---|---|
| **Subject ID:** | | | |
| **Age in months at the time of the interview:** | | | |
| **Sex of subject at birth:** | | | |
| | Never | Sometimes /Partially | Usually |
| Reads at least 10 words aloud. | 0 | 1 | 2 |
| Prints at least 10 simple words from memory (for example, hat, ball, the, etc.). | 0 | 1 | 2 |
| Reads simple stories aloud (that is, stories with sentences of three to five words). | 0 | 1 | 2 |
| Prints simple sentences of three or four words; may make small errors in spelling or sentence structure. | 0 | 1 | 2 |
| Prints more than 20 words from memory; may make small spelling errors. | 0 | 1 | 2 |
| Reads and understands material of at least second-grade level. | 0 | 1 | 2 |
| Puts lists of words in alphabetical order. | 0 | 1 | 2 |
| Writes simple correspondence at least three sentences long (for example, postcards, thank-you notes, e-mail, etc.). | 0 | 1 | 2 |
| Reads and understands material of at least fourth-grade level. | 0 | 1 | 2 |
| Writes reports, papers, or essays at least one page long; may use computer. | 0 | 1 | 2 |
| Writes complete mailing and return addresses on letters or packages. | 0 | 1 | 2 |
| Reads and understands material of least sixth-grade level. | 0 | 1 | 2 |
| Edits or corrects own written work before handing it in (for example, checks punctuation, spelling, grammar, etc.). | 0 | 1 | 2 |
| Writes advanced correspondence at least 10 sentences long; may use computer. | 0 | 1 | 2 |
| Reads and understand material of at least ninth-grade level. | 0 | 1 | 2 |
| Reads at least two newspaper articles weekly (print or electronic version). | 0 | 1 | 2 |
| Writes business letters (for example, requests information, makes complaint, places order, etc.); may use computer. | 0 | 1 | 2 |
| Opens mouth when food is offered. | 0 | 1 | 2 |
| Eats solid foods (for example, cooked vegetables, chopped meats, etc.). | 0 | 1 | 2 |
| Sucks or chews on finger foods (for example, crackers, cookies, toast, etc.). | 0 | 1 | 2 |
| Drinks from a cup or glass; may spill. | 0 | 1 | 2 |
| Let someone know when he or she has wet or soiled diaper or pants (for example, points, vocalizes, pulls at diaper, etc.). | 0 | 1 | 2 |
| Feeds self with spoon; may spill. | 0 | 1 | 2 |
| Sucks from straw. | 0 | 1 | 2 |
| Takes off clothing that opens in the front (for example, a coat or sweater); does not have to unbutton or unzip the clothing. | 0 | 1 | 2 |
| Pulls up clothing with elastic waistbands (for example, underwear or sweatpants). | 0 | 1 | 2 |
| Feeds self with fork; may spill. | 0 | 1 | 2 |
| Drinks from cup or glass without spilling | 0 | 1 | 2 |
| Feeds self with spoon without spilling. | 0 | 1 | 2 |
| Urinates in toilet or potty chair. | 0 | 1 | 2 |
| Puts on clothing that opens in the front (for example, a coat or sweater); does not have to zip or button the clothing. | 0 | 1 | 2 |
| Asks to use toilet. | 0 | 1 | 2 |

| Vineland Questions | | | |
|---|---|---|---|
| **Subject ID:** | | | |
| **Age in months at the time of the interview:** | | | |
| **Sex of subject at birth:** | | | |
| | Never | Sometimes /Partially | Usually |
| Defecates in toilet or potty chair. | 0 | 1 | 2 |
| Is toilet-trained during the day. | 0 | 1 | 2 |
| Zips zippers that are fastened at the bottom (for example, in pants, on backpacks, etc.). | 0 | 1 | 2 |
| Wipes or blows nose using tissue or handkerchief. | 0 | 1 | 2 |
| Is toilet-trained during the night. | 0 | 1 | 2 |
| Puts shoes on correct feet; does not need to tie laces. | 0 | 1 | 2 |
| Fastens snaps. | 0 | 1 | 2 |
| Holds spoon, fork, and knife correctly. | 0 | 1 | 2 |
| Washes and dries face using soap and water. | 0 | 1 | 2 |
| Brushes teeth. | 0 | 1 | 2 |
| Buttons large buttons in front, in correct buttonholes. | 0 | 1 | 2 |
| Covers mouth and nose when coughing and sneezing. | 0 | 1 | 2 |
| Buttons small buttons in front, in correct buttonholes. | 0 | 1 | 2 |
| Connects and zips zippers that are not fastened at the bottom (for example, in jackets, sweatshirts, etc.). | 0 | 1 | 2 |
| Turns faucets on and adjusts temperature by adding hot or cold water. | 0 | 1 | 2 |
| Wears appropriate clothing during wet or cold weather (for example, raincoat, boots, sweater, etc.). | 0 | 1 | 2 |
| Bathes or showers and dries self. | 0 | 1 | 2 |
| Finds and uses appropriate public restroom for his or her gender. | 0 | 1 | 2 |
| Washes and dries hair (with towel or hair dryer). | 0 | 1 | 2 |
| Cares for minor cuts (for example, cleans wound, puts on bandage, etc.). | 0 | 1 | 2 |
| Takes medicine as directed (that is, follows directions on label). | 0 | 1 | 2 |
| Uses thermometer to take own or another's temperature. | 0 | 1 | 2 |
| Seeks medical help in emergency (for example, recognizes symptoms of serious illness or injury, such as shortness of breath, chest pain or uncontrolled bleeding). May mark "N/O" for No Opportunity if individual has not been in medical emergency. | 0 | 1 | 2 |
| Follows directions for health care procedures special diet or medical treatments. You may mark "N/O" for No Opportunity if the individual does not have a health concern that requires special procedures diet or treatments. | 0 | 1 | 2 |
| Keeps track of medications (non-prescription and prescription) and refills them as needed. | 0 | 1 | 2 |
| Makes appointments for regular medical and dental checkups. | 0 | 1 | 2 |
| Is careful around hot objects (for example, the stove or oven, an open fire, etc.). | 0 | 1 | 2 |
| Helps with simple household chores (for example, dusts, picks up clothes or toys, feeds pet, etc.). | 0 | 1 | 2 |
| Clears unbreakable items from own place at table. | 0 | 1 | 2 |
| Cleans up play or work area at end of an activity (for example, finger painting, model building, etc.). | 0 | 1 | 2 |

| Vineland Questions | | | |
|---|---|---|---|
| **Subject ID:** | | | |
| **Age in months at the time of the interview:** | | | |
| **Sex of subject at birth:** | | | |
| | Never | Sometimes /Partially | Usually |
| Puts away personal possessions (for example, toys, books, magazines, etc.). | 0 | 1 | 2 |
| Is careful when using sharp objects (for example, scissors, knives, etc.). | 0 | 1 | 2 |
| Clears breakable items from own place at table. | 0 | 1 | 2 |
| Helps prepare foods that require mixing and cooking (for example, cake or cookie mixes, macaroni and cheese, etc.). | 0 | 1 | 2 |
| Uses simple appliances (for example, a toaster, can opener, bottle opener, etc.). | 0 | 1 | 2 |
| Uses microwave oven for heating, baking, or cooking (that is, sets time and power setting, etc.). | 0 | 1 | 2 |
| Puts clean clothes away in proper place (for example, in drawers or closet, on hooks, etc.). | 0 | 1 | 2 |
| Uses tools (for example, a hammer to drive nails, a screwdriver to screw and unscrew screws, etc.). | 0 | 1 | 2 |
| Washes dishes by hand, or loads and uses dishwasher. | 0 | 1 | 2 |
| Sweeps mops or vacuums floors thoroughly. | 0 | 1 | 2 |
| Clears table completely (for example, scrapes and stacks dishes, throws away disposable items, etc.). | 0 | 1 | 2 |
| Uses household products correctly (for example, laundry detergent, furniture polish, glass cleaner, etc.). | 0 | 1 | 2 |
| Prepares basic foods that do not need mixing but require cooking (for example, rice, soup, vegetables, etc.). | 0 | 1 | 2 |
| Cleans one or more rooms other than own bedroom. | 0 | 1 | 2 |
| Uses sharp knife to prepare food. | 0 | 1 | 2 |
| Uses stove or oven for heating, baking, or cooking (that is, turns burners on and off, sets oven temperature, etc.). | 0 | 1 | 2 |
| Prepares food from ingredients that require measuring, mixing, and cooking. | 0 | 1 | 2 |
| Washes clothing as needed. | 0 | 1 | 2 |
| Performs maintenance tasks as needed (for example, replaces light bulbs, changes vacuum cleaner bag, etc.). | 0 | 1 | 2 |
| Plans and prepares main meal of the day. | 0 | 1 | 2 |
| Demonstrates understanding of function of telephone (for example, pretends to talk on phone, etc.). | 0 | 1 | 2 |
| Talks to familiar person on telephone. | 0 | 1 | 2 |
| Uses TV or radio without help (for example, turns equipment on, accesses channel or station, selects program, etc.). | 0 | 1 | 2 |
| Counts at least 10 objects, one by one. | 0 | 1 | 2 |
| Is aware of and demonstrates appropriate behavior while riding in car (for example, keeps seat belt on, refrains from distracting driver, etc.). | 0 | 1 | 2 |
| Demonstrates understanding of the function of money (for example, says, "Money is what you need to buy things at the store"; etc.). | 0 | 1 | 2 |
| Uses sidewalk (where available) or shoulder of road when walking or using wheeled equipment (skates, scooter, tricycle, etc.). | 0 | 1 | 2 |
| Demonstrates understanding of function of clock (for example, says, "Clocks tell time"; "What time can we go?"; etc.). | 0 | 1 | 2 |
| Follows household rules (for example, no running in the house, no jumping on the furniture, etc.). | 0 | 1 | 2 |

| Vineland Questions | | | |
|---|---|---|---|
| **Subject ID:** | | | |
| **Age in months at the time of the interview:** | | | |
| **Sex of subject at birth:** | | | |
| | Never | Sometimes /Partially | Usually |
| Demonstrates computer skills necessary to play games or start programs with computer turned on; does not need to turn computer on by self. | 0 | 1 | 2 |
| Summons to the telephone the person receiving a call or indicates that the person is not available. | 0 | 1 | 2 |
| Identifies penny, nickel, dime, and quarter by name when asked; does not need to know the value of coins. | 0 | 1 | 2 |
| Looks both ways when crossing streets or roads. | 0 | 1 | 2 |
| Says current day of the week when asked. | 0 | 1 | 2 |
| Demonstrates understanding of right to personal privacy for self and others (for example, while using restroom or changing clothes, etc.). | 0 | 1 | 2 |
| Demonstrates knowledge of what phone number to call in an emergency when asked. | 0 | 1 | 2 |
| Tells time using a digital clock or watch. | 0 | 1 | 2 |
| States value of penny (1 cent), nickel (5 cents), dime (10 cents), and quarter (25 cents). | 0 | 1 | 2 |
| Discriminates between bills of different denominations (for example, refers to $1 bills, $5 bills, etc., in conversation; etc.). | 0 | 1 | 2 |
| Obeys traffic lights and "Walk" and "Don't Walk" signs. | 0 | 1 | 2 |
| Points to current or other date on calendar when asked. | 0 | 1 | 2 |
| Demonstrates understanding that some items cost more than others (for example, says, "I have enough money to buy gum but not a candy bar"; "Which pencil costs less?"; etc.). | 0 | 1 | 2 |
| Tells time by the half hour on analog clock (for example, 1:30, 2:00, etc.). | 0 | 1 | 2 |
| Makes telephone calls to others, using standard or cell phone. | 0 | 1 | 2 |
| Orders a complete meal in a fast-food restaurant. | 0 | 1 | 2 |
| Carries or stores money safely (for example, in wallet, purse, money belt, etc.). | 0 | 1 | 2 |
| Tells time by 5-minute segments on analog clock (for example, 1:05, 1:10, etc.). | 0 | 1 | 2 |
| Obeys curfew parent or caregiver sets. | 0 | 1 | 2 |
| Watches or listens to programs for information (for example, weather report, news, educational program, etc.). | 0 | 1 | 2 |
| Counts change from a purchase. | 0 | 1 | 2 |
| Demonstrates computer skills necessary to carry out complex tasks (for example, word processing, accessing the Internet, installing software, etc.). | 0 | 1 | 2 |
| Evaluates quality and price when selecting items to purchase. | 0 | 1 | 2 |
| Obeys time limits for breaks (for example, lunch or coffee breaks, etc.). | 0 | 1 | 2 |
| Travels at least 5 to 10 miles to familiar destination (that is, bikes, uses public transportation, or drives self). | 0 | 1 | 2 |
| Demonstrates understanding of right to complain or report legitimate problems when dissatisfied with services or situations. | 0 | 1 | 2 |
| Notifies school or supervisor when he or she will be late or absent. | 0 | 1 | 2 |
| Uses savings or checking account responsibly (for example, keeps some money in account, tracks balance carefully, etc.). | 0 | 1 | 2 |
| Travels at least 5 to 10 miles to unfamiliar destination (that is, bikes, uses public transportation, or drives self). | 0 | 1 | 2 |

| Vineland Questions | | | |
|---|---|---|---|
| **Subject ID:** | | | |
| **Age in months at the time of the interview:** | | | |
| **Sex of subject at birth:** | | | |
| | Never | Sometimes /Partially | Usually |
| Earns money at part-time job (that is, at least 10 hours a week) for 1 year. | 0 | 1 | 2 |
| Attempts to improve job performance after receiving constructive criticism from supervisor. | 0 | 1 | 2 |
| Manages own money (for example, pays most or all own expenses, uses checks or money orders for purchases as needed, etc.). | 0 | 1 | 2 |
| Has held full-time job for 1 year. | 0 | 1 | 2 |
| Budgets for monthly expenses (for example, utilities, rent, etc.). | 0 | 1 | 2 |
| Applies for and uses personal credit card responsibly (for example, does not exceed credit limit, pays on time, etc.). | 0 | 1 | 2 |
| Looks at face of parent or caregiver. | 0 | 1 | 2 |
| Watches (that is, follows with eyes) someone moving by crib or bed for 5 seconds or more. | 0 | 1 | 2 |
| Shows two or more emotions (for example, laughs, cries, screams, etc.). | 0 | 1 | 2 |
| Smiles or makes sounds when approached by a familiar person. | 0 | 1 | 2 |
| Makes or tries to make social contact (for example, smiles, makes noises, etc.). | 0 | 1 | 2 |
| Reaches for familiar person when person holds out arms to him or her. | 0 | 1 | 2 |
| Shows preference for certain people and objects (for example, smiles, reaches for or moves toward person or object, etc.). | 0 | 1 | 2 |
| Shows affection to familiar persons (for example, touches, hugs, kisses, cuddles, etc.). | 0 | 1 | 2 |
| Imitates or tries to imitate parent's or caregiver's facial expressions (for example, smiles, frowns, etc.). | 0 | 1 | 2 |
| Moves about looking for parent or caregiver or other familiar person nearby. | 0 | 1 | 2 |
| Shows interest in children the same age, other than brothers or sisters (for example, watches them, smiles at them, etc.). | 0 | 1 | 2 |
| Imitates simple movements (for example, claps hands, waves good-bye, etc.). | 0 | 1 | 2 |
| Uses actions to show happiness or concern for others (for example, hugs, pats arm, holds hands, etc.). | 0 | 1 | 2 |
| Shows desire to please others (for example, shares a snack or toy, tries to help even if not capable, etc.). | 0 | 1 | 2 |
| Demonstrates friendship-seeking behavior with others the same age (for example says "Do you want to play?" or takes another child by the hand etc.). | 0 | 1 | 2 |
| Imitates relatively complex actions as they are being performed by another person (for example, shaving, putting on makeup, hammering nails, etc.). | 0 | 1 | 2 |
| Answers when familiar adults make small talk (for example: if asked "How are you?" says "I'm fine"; if told "You look nice" says "Thank you"; etc.). | 0 | 1 | 2 |
| Repeats phrases heard spoken before by an adult (for example: "Honey I'm home"; "No dessert until you clean your plate"; etc.). | 0 | 1 | 2 |
| Uses words to express own emotions (for example, "I'm happy"; "I'm scared"; etc.). | 0 | 1 | 2 |
| Has best friend or shows preference for certain friends (of either sex) over others. | 0 | 1 | 2 |
| Imitates relatively complex actions several hours after watching someone else perform them (for example, shaving, putting on makeup, hammering nails, etc.). | 0 | 1 | 2 |

| Vineland Questions | | | |
|---|---|---|---|
| **Subject ID:** | | | |
| **Age in months at the time of the interview:** | | | |
| **Sex of subject at birth:** | | | |
| | Never | Sometimes /Partially | Usually |
| Uses words to express happiness or concern for others (for example, says, "Yeah! You won"; "Are you all right?"; etc.). | 0 | 1 | 2 |
| Acts when another person needs a helping hand (for example, holds door open, picks up dropped items, etc.). | 0 | 1 | 2 |
| Recognizes the likes and dislikes of others (for example, says, "Chow likes soccer"; "Susie doesn't eat pizza"; etc.). | 0 | 1 | 2 |
| Shows same level of emotion as others around him or her (for example, does not downplay or over dramatize a situation, etc.). | 0 | 1 | 2 |
| Keeps comfortable distance between self and others in social situations (for example, does not get too close to another person when talking, etc.). | 0 | 1 | 2 |
| Talks with others about shared interests (for example, sports, TV shows, summer plans, etc.). | 0 | 1 | 2 |
| Starts small talk when meets people he or she knows (for example, says, "How are you?"; "What's up?"; etc.). | 0 | 1 | 2 |
| Meets with friends regularly. | 0 | 1 | 2 |
| Chooses not to say embarrassing or mean things or ask rude questions in public. | 0 | 1 | 2 |
| Places reasonable demands on friendship (for example, does not expect to be a person's only friend or to have the friend always available, etc.). | 0 | 1 | 2 |
| Understands that others do not know his or her thoughts unless he or she says them. | 0 | 1 | 2 |
| Is careful when talking about personal things. | 0 | 1 | 2 |
| Cooperates with others to plan or be part of an activity (for example, a birthday party, sports event, etc.). | 0 | 1 | 2 |
| Demonstrates understanding of hints or indirect cues in conversation (for example: knows that yawns may mean "I'm bored" or a quick change of subject may mean "I don't want to talk about that"; etc.). | 0 | 1 | 2 |
| Starts conversations by talking about things that interest others (for example, "Tyrone tells me you like computers"; etc.). | 0 | 1 | 2 |
| Goes on group dates. | 0 | 1 | 2 |
| Goes on single dates. | 0 | 1 | 2 |
| Responds when parent or caregiver is playful (for example, smiles, laughs, claps hands, etc.). | 0 | 1 | 2 |
| Shows interest in where he or she is (for example, looks or moves around, touches objects or people, etc.). | 0 | 1 | 2 |
| Plays simple interaction games with others (for example, peek-a-boo, patty-cake, etc.). | 0 | 1 | 2 |
| Plays near another child, each doing different things. | 0 | 1 | 2 |
| Chooses to play with other children (for example, does not stay on the edge of a group or avoid others). | 0 | 1 | 2 |
| Plays cooperatively with one or more children for up to 5 minutes. | 0 | 1 | 2 |
| Plays cooperatively with more than one child for more than 5 minutes. | 0 | 1 | 2 |
| Continues playing with another child with little fussing when parent or caregiver leaves. | 0 | 1 | 2 |
| Shares toys or possessions when asked. | 0 | 1 | 2 |
| Plays with others with minimal supervision. | 0 | 1 | 2 |
| Uses common household objects or other objects for make-believe activities (for example, pretends a block is a car, a box is a house, etc.). | 0 | 1 | 2 |

| Vineland Questions | | | |
|---|---|---|---|
| **Subject ID:** | | | |
| **Age in months at the time of the interview:** | | | |
| **Sex of subject at birth:** | | | |
| | Never | Sometimes /Partially | Usually |
| Protects self by moving away from those who destroy things or cause injury (for example, those who bite, hit, throw things, pull hair, etc.). | 0 | 1 | 2 |
| Plays simple make-believe activities with others (for example, plays dress-up, pretends to be superheroes, etc.). | 0 | 1 | 2 |
| Seeks out others for companionship (for example, invites others home, goes to another's home, plays with others on the playground, etc.). | 0 | 1 | 2 |
| Takes turns when asked while playing games or sports. | 0 | 1 | 2 |
| Plays informal, outdoor group games (for example, tag, jump rope, catch, etc.). | 0 | 1 | 2 |
| Shares toys or possessions without being asked. | 0 | 1 | 2 |
| Follows rules in simple games (relay races, spelling bees, electronic games, etc.). | 0 | 1 | 2 |
| Takes turns without being asked. | 0 | 1 | 2 |
| Plays simple card or board game based only on chance (for example: Go Fish Crazy Eights Sorry etc.). | 0 | 1 | 2 |
| "Goes places with friends during the day with adult supervision (for example, to a shopping mall, park, community center, etc.)." | 0 | 1 | 2 |
| Asks permission before using objects belonging to or being used by another. | 0 | 1 | 2 |
| Refrains from entering group when nonverbal cues indicate that he or she is not welcome. | 0 | 1 | 2 |
| Plays simple games that require keeping score (for example, kickball, pickup basketball, etc.). | 0 | 1 | 2 |
| Shows good sportsmanship (that is, follows rules, is not overly aggressive, congratulates other team on winning, and does not get mad when losing). | 0 | 1 | 2 |
| Plays more than one board, card, or electronic game requiring skill and decision making (for example, Monopoly, Cribbage, etc.). | 0 | 1 | 2 |
| Goes places with friends in evening with adult supervision (for example, to a concert, lecture, sporting event, movie, etc.). | 0 | 1 | 2 |
| Follows rules in complex games or sports (for example, football, soccer, volleyball, etc.). | 0 | 1 | 2 |
| Goes places with friends during the day without adult supervision (for example, to a shopping mall, park, community center, etc.). | 0 | 1 | 2 |
| Plans fun activities with more than two things to be arranged (for example, a trip to a beach or park that requires planning transportation, food, recreational items, etc.). | 0 | 1 | 2 |
| Goes places with friends in evening without adult supervision (for example, to a concert, lecture, sporting event, movie, etc.). | 0 | 1 | 2 |
| Changes easily from one at-home activity to another. | 0 | 1 | 2 |
| Says "thank you" when given something. | 0 | 1 | 2 |
| Changes behavior depending on how well he or she knows another person (for example, acts differently with family member than with stranger, etc.). | 0 | 1 | 2 |
| Chews with mouth closed. | 0 | 1 | 2 |
| Says "please" when asking for something. | 0 | 1 | 2 |
| Ends conversations appropriately (for example, says, "Good-bye"; "See you later"; etc.). | 0 | 1 | 2 |
| Cleans or wipes face and hands during and/or after meals. | 0 | 1 | 2 |
| Responds appropriately to reasonable changes in routine (for example, refrains from complaining, etc.). | 0 | 1 | 2 |

| Vineland Questions | | | |
|---|---|---|---|
| **Subject ID:** | | | |
| **Age in months at the time of the interview:** | | | |
| **Sex of subject at birth:** | | | |
| | Never | Sometimes /Partially | Usually |
| Says that he or she is sorry for unintended mistakes (for example, bumping into someone, etc.). | 0 | 1 | 2 |
| Chooses not to taunt, tease, or bully. | 0 | 1 | 2 |
| Acts appropriately when introduced to strangers (for example, nods, smiles, shakes hands, greets them, etc.). | 0 | 1 | 2 |
| Changes voice level depending on location or situation (for example, in a library, during a movie or play, etc.). | 0 | 1 | 2 |
| Says he or she is sorry after hurting another's feelings. | 0 | 1 | 2 |
| Refrains from talking with food in mouth. | 0 | 1 | 2 |
| Talks with others without interrupting or being rude. | 0 | 1 | 2 |
| Accepts helpful suggestions or solutions from others. | 0 | 1 | 2 |
| Controls anger or hurt feelings when plans change for reason(s) that cannot be helped (for example, bad weather, car trouble, etc.). | 0 | 1 | 2 |
| Keeps secrets or confidences for longer than one day. | 0 | 1 | 2 |
| Says he or she is sorry after making unintentional mistakes or errors in judgment (for example, when unintentionally leaving someone out of a game, etc.). | 0 | 1 | 2 |
| Shows understanding that gentle teasing with family and friends can be a form of humor or affection. | 0 | 1 | 2 |
| Tells parent or caregiver about his or her plans (for example, what time he or she is leaving and returning, where he or she is going, etc.). | 0 | 1 | 2 |
| Chooses to avoid dangerous or risky activities (for example, jumping off high places, picking up a hitchhiker, driving recklessly, etc.). | 0 | 1 | 2 |
| Controls anger or hurt feelings when he or she does not get his or her way (for example, when not allowed to watch television or attend a party; when suggestion is rejected by friend or supervisor; etc.). | 0 | 1 | 2 |
| Follows through with arrangements (for example, if promises to meet someone, meets that person; etc.). | 0 | 1 | 2 |
| Stops or stays away from relationships or situations that are hurtful or dangerous (for example, being bullied or made fun of, being taken advantage of sexually or financially, etc.). | 0 | 1 | 2 |
| Controls anger or hurt feelings due to constructive criticism (for example, correction of misbehavior, discussion of test score or grade, performance review, etc.). | 0 | 1 | 2 |
| Keeps secrets or confidences for as long as needed. | 0 | 1 | 2 |
| Thinks about what could happen before making decisions (for example, refrains from acting impulsively, thinks about important information, etc.). | 0 | 1 | 2 |
| Is aware of potential danger and uses caution when encountering risky social situations (for example, binge drinking parties, Internet chat rooms, personal ads, etc.). | 0 | 1 | 2 |
| Shows respect for co-workers (for example, does not distract or interrupt others who are working, is on time for meetings, etc.). | 0 | 1 | 2 |
| Holds head erect for at least 15 seconds when held upright in parent's or caregiver's arms. | 0 | 1 | 2 |
| Sits supported (for example, in a chair, with pillows, etc.) for at least 1 minute. | 0 | 1 | 2 |
| Sits without support for at least 1 minute. | 0 | 1 | 2 |
| Creeps or moves on stomach across floor. | 0 | 1 | 2 |

| Vineland Questions | | | |
| --- | --- | --- | --- |
| **Subject ID:** | | | |
| **Age in months at the time of the interview:** | | | |
| **Sex of subject at birth:** | | | |
| | Never | Sometimes /Partially | Usually |
| Sits without support for at least 10 minutes. | 0 | 1 | 2 |
| Raises self to sitting position and sits without support for at least 1 minute. | 0 | 1 | 2 |
| Crawls at least 5 feet on hands and knees, without stomach touching floor. | 0 | 1 | 2 |
| Pulls self to standing position. | 0 | 1 | 2 |
| Crawls up stairs. | 0 | 1 | 2 |
| Takes at least two steps. | 0 | 1 | 2 |
| Stands alone for 1 to 3 minutes. | 0 | 1 | 2 |
| Rolls ball while sitting. | 0 | 1 | 2 |
| Climbs on and off low objects (for example, chair, step stool, slide, etc.). | 0 | 1 | 2 |
| Crawls down stairs. | 0 | 1 | 2 |
| Stands for at least 5 minutes. | 0 | 1 | 2 |
| Walks across room; may be unsteady and fall occasionally. | 0 | 1 | 2 |
| Throws ball. | 0 | 1 | 2 |
| Walks to get around; does not need to hold on to anything. | 0 | 1 | 2 |
| Climbs on and off adult-sized chair. | 0 | 1 | 2 |
| Runs without falling; may be awkward and uncoordinated. | 0 | 1 | 2 |
| Walks up stairs, putting both feet on each step; may use railing. | 0 | 1 | 2 |
| Kicks ball. | 0 | 1 | 2 |
| Runs smoothly without falling. | 0 | 1 | 2 |
| Walks downstairs, facing forward, putting both feet on each step; may use railing. | 0 | 1 | 2 |
| Jumps with both feet off floor. | 0 | 1 | 2 |
| Throws ball of any size in specific direction. | 0 | 1 | 2 |
| Catches beach ball-sized ball with both hands from a distance of 2 or 3 feet. | 0 | 1 | 2 |
| Walks up stairs, alternating feet; may use railing. | 0 | 1 | 2 |
| Pedals tricycle or other three-wheeled toy for at least 6 feet. | 0 | 1 | 2 |
| Jumps or hops forward at least three times. | 0 | 1 | 2 |
| Hops on one foot at least once without falling; may hold on to something for balance. | 0 | 1 | 2 |
| Climbs on and off high objects (for example, jungle gym, 4-foot slide ladder, etc.). | 0 | 1 | 2 |
| Walks downstairs, alternating feet; may use railing. | 0 | 1 | 2 |
| Runs smoothly, with changes in speed and direction. | 0 | 1 | 2 |
| Rides bicycle with training wheels for at least 10 feet. | 0 | 1 | 2 |
| Catches beach ball-sized ball (from at least 6 feet away) with both hands. | 0 | 1 | 2 |
| Hops forward on one foot with ease. | 0 | 1 | 2 |
| Skips at least 5 feet. | 0 | 1 | 2 |
| Catches tennis or baseball-sized ball from at least 10 feet away), moving to catch it if necessary. | 0 | 1 | 2 |

| Vineland Questions | | | |
|---|---|---|---|
| **Subject ID:** | | | |
| **Age in months at the time of the interview:** | | | |
| **Sex of subject at birth:** | | | |
| | Never | Sometimes /Partially | Usually |
| Rides bicycle with no training wheels without falling. | 0 | 1 | 2 |
| Reaches for toy or object. | 0 | 1 | 2 |
| Picks up small objects (no larger than 2 inches on any side); may use both hands | 0 | 1 | 2 |
| Moves object from one hand to the other. | 0 | 1 | 2 |
| Squeezes squeaky toy or object. | 0 | 1 | 2 |
| Picks up small object with thumb and fingers. | 0 | 1 | 2 |
| Removes object (for example, a block or clothespin) from a container. | 0 | 1 | 2 |
| Puts object (for example, a block or clothespin) into a container. | 0 | 1 | 2 |
| Turns pages of board, cloth, or paper book, one at a time. | 0 | 1 | 2 |
| Stacks at least four small blocks or other small objects; stack must not fall. | 0 | 1 | 2 |
| Opens doors by turning doorknobs. | 0 | 1 | 2 |
| Unwraps small objects (for example, gum or candy). | 0 | 1 | 2 |
| Completes simple puzzle of at least two pieces or shapes. | 0 | 1 | 2 |
| Turns book or magazine pages one by one. | 0 | 1 | 2 |
| Uses twisting hand-wrist motion (for example, winds up toy, screws/unscrews lid of jar, etc.). | 0 | 1 | 2 |
| Holds pencil in proper position (not with fist) for writing or drawing. | 0 | 1 | 2 |
| Colors simple shapes; may color outside lines. | 0 | 1 | 2 |
| Builds three-dimensional structures (for example, a house, bridge, vehicle, etc.) with at least five small blocks. | 0 | 1 | 2 |
| Opens and closes scissors with one hand. | 0 | 1 | 2 |
| Glues or pastes two or more pieces together (for example, for art or science projects, etc.). | 0 | 1 | 2 |
| Uses tape to hold things together (for example, torn page, art project, etc.). | 0 | 1 | 2 |
| Draws more than one recognizable form (for example, person, house, tree, etc.). | 0 | 1 | 2 |
| Makes recognizable letters or numbers. | 0 | 1 | 2 |
| Draws circle freehand while looking at example. | 0 | 1 | 2 |
| Uses scissors to cut across paper along a straight line. | 0 | 1 | 2 |
| Colors simple shapes; colors inside the lines. | 0 | 1 | 2 |
| Cuts out simple shapes (for example, circles, squares, rectangles, etc.). | 0 | 1 | 2 |
| Uses eraser without tearing paper. | 0 | 1 | 2 |
| Draws square freehand while looking at example. | 0 | 1 | 2 |
| Draws triangle freehand while looking at example. | 0 | 1 | 2 |
| Ties knot. | 0 | 1 | 2 |
| Draws straight line using a ruler or straightedge. | 0 | 1 | 2 |
| Unlocks dead bolt; key; or combination locks that require twisting. | 0 | 1 | 2 |
| Cuts out complex shapes (for example, stars, animals, alphabet letters, etc.). | 0 | 1 | 2 |

| Vineland Questions | | | |
|---|---|---|---|
| **Subject ID:** | | | |
| **Age in months at the time of the interview:** | | | |
| **Sex of subject at birth:** | | | |
| | Never | Sometimes /Partially | Usually |
| Uses keyboard, typewriter, or touch screen to type name or short words; may look at keys. | 0 | 1 | 2 |
| Ties secure bow. | 0 | 1 | 2 |
| Uses a keyboard to type up to 10 lines; may look at the keys. | 0 | 1 | 2 |
| Is overly dependent (that is, clings to caregiver, teacher, brother, or sister). | 0 | 1 | 2 |
| Avoids others and prefers to be alone. | 0 | 1 | 2 |
| Has eating difficulties (for example, eats too fast or too slowly, hoards food, overeats, refuses to eat, etc.). | 0 | 1 | 2 |
| Has sleep difficulties (for example, sleepwalks, has frequent nightmares, sleeps significantly more or less than typical for his or her age). | 0 | 1 | 2 |
| Refuses to go to school or work because of fear, feelings of rejection or isolation, etc. | 0 | 1 | 2 |
| Is overly anxious or nervous. | 0 | 1 | 2 |
| Cries or laughs too easily. | 0 | 1 | 2 |
| Has poor eye contact (that is, does not look at or face others when speaking or spoken to). | 0 | 1 | 2 |
| Is sad for no clear reason. | 0 | 1 | 2 |
| Avoids social interaction. | 0 | 1 | 2 |
| Lacks energy or interest in life. | 0 | 1 | 2 |

**4.2 Appendix B**: *Autism Observational Scale for Infants (AOSI) Individual Items*

| AOSI Items |
|---|
| Visual tracking |
| Disengagement of attention |
| Orients to name |
| Differential Response to Facial Emotion |
| Imitation of Action |
| Anticipatory Responses |
| Social babbling |
| Eye contact |
| Reciprocal social smile |
| Coordination of eye gaze and action |
| Reactivity |
| Social interest and shared affect |
| Transitions |
| Motor Control and Behavior |
| Atypical motor behaviors |
| Atypical sensory behaviors |
| Engagement of attention |
| Insistence on having or playing with particular objects |
| Sharing interest |

**Curriculum Vitae**

| | |
|---|---|
| **Name:** | Julia Teixeira Pinto Montenegro |
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| **Honours and Awards:** | Ministry of Education Scholarship – Federative Republic of Brazil<br>2012-2014<br><br>FUNDAP Scholarship – Government of State of São Paulo<br>2011-2012 |
| **Related Work Experience** | Research Assistant<br>The University of Western Ontario - Developing Brain Lab<br>2020-2022<br><br>Research Assistant<br>The University of Western Ontario - Sensory Perception Lab<br>2018 – 2020<br><br>Psychologist at Babies and their Parents Research and Intervention Group<br>Langage Institute – São Paulo, SP – Brazil<br>2015-2021 |

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2014-2014

**Publications:**

Montenegro, J. T. P., Seguin, D., & Duerden, E. G. Joint Attention in Infants at High Familial Risk for Autism Spectrum Disorder and the association with Thalamic and Hippocampal Macrostructure. *Under review*.

Montenegro, J.T.P.; Santos, J. V.; Riquena, C. A psicose na adolescência e suas possibilidades de escuta In: Dos primórdios à adolescência: desafios e perspectivas. 1st ed. São Paulo: Langage (2020), p. 261-272. Brazilian/Portuguese, Print, ISBN: 9788562686399

Casetto, S. J.; Henz, A. O.; Garcia, M. L.; Aguiar, F. B.; Montenegro, J. T.; Unzueta, L. B.; Capozzolo, A. A. (2016) A good training based on insufficiency: Work in health care as an ethics. *Journal of Health Psychology,* Vol. 21(3) 291–301

**Conference Presentations:**

Montenegro, J. T. P., Seguin, D., & Duerden, E. G. Joint Attention in 4- and 6-month-old Infants at High Familial Risk for Autism Spectrum Disorder: association with brain development. Accepted for presentation at the 7th Annual Dr. Benjamin Goldberg Developmental Disabilities Research Day, 2022. London, Canada [Oral Presentation].

Montenegro, J. T. P., Seguin, D., & Duerden, E. G. Joint Attention in 4- and 6-month-old Infants at High Familial Risk for Autism Spectrum Disorder: association with brain development. Accepted for presentation at the Child Health Research Day Conference, 2022. London, Canada [Poster].

Montenegro, J. T. P.; Seguin, D.; Christiaans, E.; Duerden, E. Baby Gaze: a study of early social gaze. Presented at VII Séminaire International Transdisciplinaire sur le bébé 2021. Langage Institute, Pitié-Salpêtrière, Université Pierre et Marie Curie. Paris, France. Online [Oral Presentation]

Montenegro, J. T. P. The early experience of pain in infants: reflections from current research. Presented at VI Congrès International Transdisciplinaire sur l'enfant et l'adolescent 2020. Langage Institute, UFMG and Université Paris Diderot. Brazil. Online [Oral Presentation]

Shafai, F. L., Scheerer, N., Feldman, J. I., Gateman, E., Altoum, E., Iarocci, G., Woynaroski, T. G., Montenegro, J. T. P., Stevenson, R. A. Pupil Dilation Responses in Autism: The Impact of Emotional Category and Intensity at INSAR 2020. Online [Oral Presentation]

Montenegro, J. T. P. The baby nowadays: competences and sensory abilities at VI Congrés Internacional Transdisciplinaire Sur Le bébé 2019. Langage Institute and Université Paris Diderot. Paris, France [Oral Presentation]

Dalal, T.C., Muller, A.M., Brierley, N.J., Sehl, C.G., Abraham, A., Tran, B., Haque, A., Shafai, F., Gateman, E., Montenegro, J., & Stevenson, R.A. The relationship between multisensory perception and schizotypal personality traits at 48th Annual Lake Ontario Visionary Establishment (L.O.V.E) Conference 2019. Niagara Falls, Canada [Poster]

Montenegro, J. T. P. Psychosis in adolescence and treatments' possibilities at V Congrès International Transdisciplinaire sur l'enfant et l'adolescent 2018. Langage Institute, UFMG and Université Paris Diderot. Belo Horizonte, Brazil [Oral Presentation]

Carvalho, T. C., Lucirio, C., Montenegro, J. T. P., Boaventura; S., Costa, S., Diaz, A. C., Parlato-Oliveira, E.; The diagnosis effect on psychoanalytic clinic in early childhood at I Congresso Internacional Transdisciplinar Portugal-Brazil sobre o bebê 2018. Langage Institute and Universidade de Évora. Évora, Portugal [Oral Presentation]

C., Lucirio, C., Montenegro, J. T. P., Boaventura S., Costa, S., Gandolfo, P., Carvalho, T. Diaz, A. C., Banzato, D., Parlato-Oliveira, E. Listening to early childhood: a psychoanalytic clinic at Séminaire Transdisciplinaire Sur Le bébé 2017. Langage Institute and Université Paris Diderot. Paris, France [Oral Presentation]