Specific Aims

Our laboratory recently discovered that a viewer's subjective assessment of the saliency of a stimulus can be quantified by measuring patterns of eye-blinking[1]. Given that blinking temporarily interrupts the flow of visual information to a viewer, our research revealed that viewers *unconsciously* adjust, on a moment-by-moment basis, the precise timing of their blinks to minimize the likelihood of missing critical visual information presented during a blink. Probabilistically, people are *least likely* to blink when looking at what they *perceive to be most important*[1,2] and *most likely* to blink during moments *perceived to be least important*[2,3]. Consequently, we can quantify the *perceived salience* of what viewers are looking at by measuring change in rate of eye-blinking relative to ongoing scene content.

The current application leverages this insight to advance our understanding of a population of patients whose condition is marked by atypical perceptions of the social world[4]: Autism Spectrum Disorder (ASD). Although social stimuli are reported to be *less salient* to individuals with ASD[5,6], a viewer's assessment of the saliency of a stimulus—an inherently *subjective* aspect of viewer experience—has traditionally been difficult to quantify objectively, limiting scientific inquiry into this defining feature of the condition. The present application uses our novel measure of perceived stimulus salience to open a new avenue of research into (1) what individuals with ASD perceive as most *salient* during free-viewing of naturalistic social scenes; (2) how perceived stimulus salience, in typical children and in children with ASD, relates to underlying brain systems; and (3) how perceived salience and active seeking of social information interacts with brain systems involved in social processing.

Unfortunately, very little is currently known about the neural systems involved in subjective perceptions of stimulus salience, in either typically-developing (TD) or ASD populations. We do know that active seeking of salient social information is inextricably linked to social information processing and may be a prerequisite for typical development of the social brain[7–9]. In contrast, the atypical engagement with social stimuli so often observed in ASD[1,5] is not only a defining feature of the condition[10], but is also a likely source of subsequent—and progressively increasing—differences in the development of brain and behavior[11]. Understanding what is perceived to be salient to children with ASD, as well as to their typical peers, and also identifying the neural systems involved in varying states of perceived salience, will provide new inroads into studying the etiology and underlying mechanisms of the disorder as well as new insights into the unique perspectives and experiences of individuals with ASD.

The current proposal combines novel measures of perceived stimulus salience (eye-blink inhibition) with functional magnetic resonance imaging (fMRI) to address the following aims in a pilot sample of TD children (*n*=15) and in children with ASD (*n*=15):

Aim 1: Identify neural systems recruited in varying states of perceived stimulus salience.

Hypothesis 1: Content perceived to be highly salient to TD children will activate brain systems relevant for reward and/or emotion processing (e.g. medial prefrontal cortex[12]) and for stimulus valence (e.g. amygdala[13], thalamus[14], nucleus accumbens[15], orbitofrontal cortex[16]), but will de-activate the default mode network[17]. Between-group comparisons will reveal whether children with ASD recruit these same or different brain systems in response to content perceived, *by participants with ASD themselves,* as highly salient.

Aim 2: Determine how perceived salience and the active seeking of social information (in this case, faces) interacts with the activity of brain systems involved in face processing.

Hypothesis 2: Faces perceived to be highly salient to TD children will increase activation in brain systems involved in face perception (e.g. occipital fusiform gyrus, mid-fusiform gyrus, and superior temporal sulcus[18]) and in thinking about the intentions of others (e.g. temporoparietal junction[19] and inferior parietal lobule[20]). Between-group comparisons will reveal whether children with ASD recruit these same or different brain systems when looking at faces perceived, *by participants with ASD themselves,* as highly salient.

Background & Significance

When we blink, the flow of visual information between the world and one's retina is interrupted. In that instant of blinking, visual stimulation from the external world is lost for 150–400 msec[21,22]. As a result, any visual information presented in those instants of blinking will be missed[23]. In this way, blinking sets a physical limit on visual attention because of its profound interruption of incoming visual information.

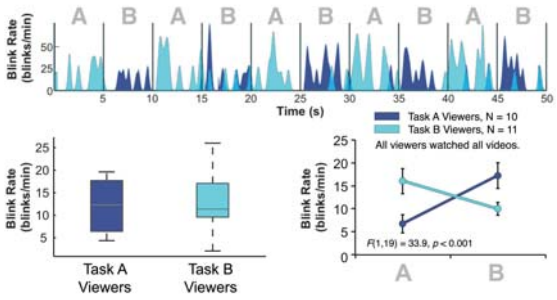

Figure 1. Rates of blinking are strongly modulated by the perceived salience of events. Viewers were arbitrarily assigned to attend to either type A or B events while watching a movie that alternated between scenes of type A or type B events (top panel). Despite having identical blink rates over the entire viewing session (bottom left), Task A viewers *decreased* their blink rate during type A events and *increased* their blink rate during type B events; Task B viewers did the same but for type B events (top panel). Bottom right: Significant interaction of viewer type by task.

In previous research[2], we tested the hypothesis that viewers unconsciously adjust the timing of when they do or do not blink in order to minimize the likelihood of losing important visual information during a blink[1]. Specifically, we tested whether viewers blink *less* when watching content that is perceived, *by the viewers themselves*, as highly salient. We designed a visual task in which different types of scene content were explicitly manipulated to be differentially salient to two groups of viewers: during video watching, typical adults were arbitrarily assigned to one of two tasks that related to only certain events within the videos. Results showed a highly significant interaction of viewer type by task on blink rate: task A viewers spontaneously *decreased* their blink rate during type A events and *increased* their blink rate during type B events. Task B viewers changed their blink rate in the same way but for their own arbitrarily-assigned events (**Figure 1**).

We also tested whether the same effects would be observed in more ecologically-valid free-viewing of video scenes (*i.e.*, without an explicitly assigned task); in that case, the viewer's "task" is defined by the viewer's own internal goals and interests. To test these effects we recruited two groups of viewers known to have different internal goals and interests: typically-developing children and children with ASD[1,24,25]. Typical 2-year-olds inhibited their blinking when watching emotionally-charged scenes and when looking at the faces of onscreen characters. In contrast, two-year-olds with ASD inhibited their blinking when looking at physical objects, and at physical objects in motion. We observed similar results in older children: typical 8-year-olds most strongly inhibited their blinking when looking at faces, whereas 8-year-olds with ASD inhibited their blinking when looking at objects, particularly those in motion.

More strikingly, the results from this study illustrate the importance of measuring not only *where* a child is looking but also the *perceived salience* of what he or she is looking at. Our eye-tracking data revealed many instances when both groups of viewers were looking at the same onscreen location at the same moment in time. While looking behavior would otherwise suggest that both groups were attending to this content in similar ways, patterns of blinking revealed a different insight into the experiences of children with ASD. For instance, *even when children with ASD spontaneously look at faces at the same time as their typical peers, their patterns of eye-blinking reveal that they fail to assess those faces as salient information in the environment*[24]*.* This insight into the subjective experience of children with ASD, and the need to link this insight to underlying brain mechanisms, is at the heart of the current application.

<u>*Significance of Aim 1:*</u> The neural circuitry underlying a person's *unconscious, subjective assessment of perceived stimulus salience* is unknown in both typical development and in ASD. Aim 1 will identify neural circuitry involved in varying states of perceived stimulus salience during free-viewing of naturalistic scenes by using blink inhibition as a measure of viewers' unconscious appraisals of what is most salient at any given moment (**Figure 2**). Results from TD children will advance knowledge of a basic and critically important adaptive mechanism that fundamentally shapes brain development: over the course of development, the brain is shaped by selective experiences; perceived stimulus salience acts as the gatekeeper in guiding those

experiences. Further, because active seeking of salient social information is inextricably linked to social information processing and shapes typical development of the social brain[7–9], examining neural systems involved in the subjective perception of stimulus salience is critical for understanding the neural mechanisms of core social deficits in ASD.

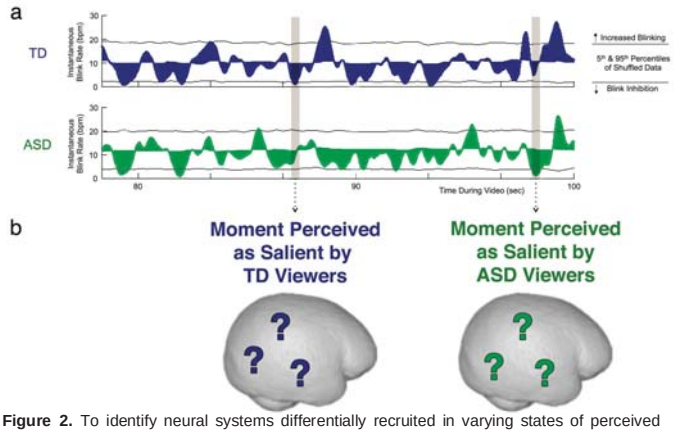

Figure 2. To identify neural systems differentially recruited in varying states of perceived stimulus salience, we will examine the brain systems recruited by TD children in response to content perceived—*by the TD children themselves*—as highly salient. To examine whether ASD children recruit the same or different brain systems in response to highly salient content, we will examine the brain systems recruited by children with ASD in response to content perceived—*by the children with ASD themselves*—as highly salient. (A) We identify moments perceived as highly salient—for each group s*eparately*—by identifying moments of statistically significant blink inhibition by comparison with permuted data as in Shultz et al, 2011[1]. Examples of moments perceived as highly salient to TD or ASD viewers are highlighted in gray. (B) Moments perceived as highly salient will be used as regressors to identify the neural circuitry underlying a person's *unconscious, subjective assessments of perceived stimulus salience*.

Significance of Aim 2: Examining how perceived salience and active seeking of social information interacts with brain systems involved in social processing offers an opportunity to test competing (though not necessarily mutually-exclusive) accounts of one of the most well-replicated neuroimaging findings in ASD: *reduced* activation in 'social brain' regions during social information processing tasks. For instance, one of the most consistent findings in ASD is hypoactivation of the fusiform gyrus (FG)[11,26,27], a region that responds strongly to faces in typical individuals, when looking at faces[28]. One interpretation of this result is that hypoactivation of the FG may play a causal role in face perception difficulties and other aspects of social disability in ASD[26]. Another interpretation is that years of reduced interest in faces[29,6] (i.e. years of perceiving faces as *less salient*) may strongly shape a child's developing brain systems[7,30–34]. In this case, FG hypoactivation may be a consequence of perceiving faces as less salient (a characteristic of ASD) rather than being an initial cause of disability. Our method offers a unique opportunity to test these interpretations by identifying moments when children with ASD *spontaneously fixate on faces that they perceive as highly salient* (**Figure 3**). If FG activation *increases* to normative levels when children with ASD fixate faces perceived, *by the children with ASD themselves,* as highly salient, then this would suggest that previous reports of FG hypoactivation reflect a reduced interest in the faces presented, rather than a general visual face processing deficit.

Each of these aims also has important implications for treatment in ASD: if differential perceptions of stimulus salience (of faces and of other social information) are key factors influencing and refining brain specialization, then treatments that augment a child's capacity to actively and successfully seek relevant social stimuli – ideally at points very early in life, prior to the time when atypical specialization becomes most pronounced[29] – might provide the greatest opportunity for optimal long-term outcomes[35,36], attenuating or even preventing some of the most deleterious consequences of ASD.

Innovation

Blink inhibition provides a measure of the unconscious assessment of perceived stimulus salience that is intrinsically linked to what a person is looking at in real time. This marks a significant advancement over other existing measures of arousal or reward, methods such as rating scales, surveys, or autonomic measures (*e.g.,* galvanic skin response or heart rate), each of which is highly multi-determined and also suffers from long latency and refractory periods[37,38]. In contrast, blink inhibition is intrinsic to the visual system and is time-locked, on a moment-by-moment basis, to what viewers are attending to. This is particularly well-suited to what we face in the real world, and also to what most often represents the greatest challenge to individuals with ASD (even those who are cognitively-able): dynamically-unfolding, fast-paced, and

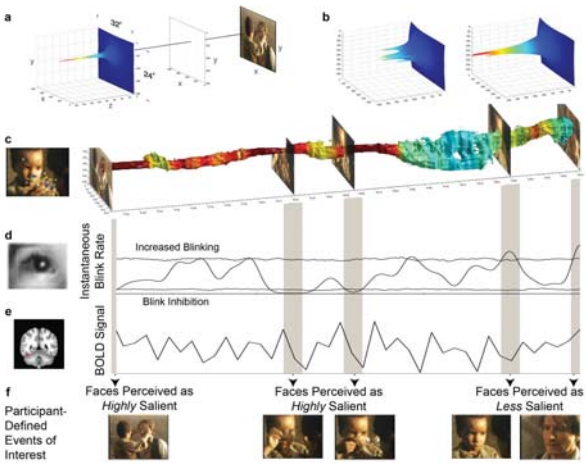

Figure 3. To determine how perceived salience interacts with the activity of brain systems involved in face processing, we will restrict our analyses to moments when viewers are fixating on faces, contrasting those perceived as highly salient with those perceived as less salient. To quantify time-varying visual scanning to dynamic stimuli, we (A) convolve a viewer's fixation location with a kernel function, then (B) compute kernel density estimates for all viewers in order to measure when visual scanning is divergent (left) or convergent (right). (C) Concatenating a series of such maps at every moment in the video, we can identify the area of greatest fixation density at each moment of 'convergent' visual scanning. In (D), (E), and (F), the main measures in this application are brought together via acquisition of simultaneous eye-tracking and fMRI data.

nuanced social situations. In such contexts, actively seeking the most relevant information at the right moment in time (i.e., dynamically reacting to and anticipating relevant social information as salient) is crucial for social functioning. To our knowledge, no studies have examined the neural circuitry underlying salience assessment during free-viewing of naturalistic scenes.

The current application now combines these methods with state-of-the-art imaging techniques. To do so, we have formed an outstanding team of investigators bringing together expertise in functional[39–41 42–44 45–47] and structural neuroimaging[48–50]; behavioral neuroscience and eye-tracking[5,29,51 1,52,53]; and patient training[54–56 57–59]. Our entire team worked to develop the combined data collection and analysis protocols:

- training protocols for mock scanning (that already resulted in successful pilot scans of 2 children with ASD and reduced head movement by 9.5-fold in mean absolute displacement (to <0.32mm) and 5.5-fold in maximum absolute displacement (to <2.9mm);
- implemented advanced motion-censoring techniques for fMRI data[60] to remove data volumes with excessive motion (especially beneficial for populations prone to movement);
- implemented high quality simultaneous eye-tracking and fMRI data collection—with calibration accuracy and signal-to-noise ratio *equal to or better than eye-tracking data collected outside the magnet* (owing to reduced head movement).

Experimental Design & Methods

Participants: A pilot sample of 15 TD and 15 ASD children (8 to 12 years-old) will be enrolled. Sample size is based upon an analysis of sample size and power in fMRI studies[61].

Experimental Procedures: All participants will complete the training procedure, the MRI procedure, and participant characterization. **Training Procedure:** To facilitate the inclusion of participants who represent the full-range of intellectual and social disability characteristic of ASD, all participants will complete a training protocol developed by Drs. Call and Shillingsburg. Training sessions will be conducted using a mock scanner and will consist of baseline assessments and operant training procedures (using contingent reinforcement and shaping procedures tailored to each child). The terminal goal will be < 3 mm of child movement for three periods of up to eight minutes in duration. The protocol has already been used successfully in children with ASD. **MRI Procedure:** MRI data will be collected on a 3T Siemens Tim Trio scanner (Siemens Medical Solutions, Malvern, PA) at the Wesley Woods Center for Systems Imaging using a 32-channel head coil. Eye-tracking data will be acquired simultaneously with functional MRI (fMRI) data, using hardware and software created by Senso Motoric Instruments (SMI) with modifications from our engineering team. Because of the challenges associated with requiring pediatric populations to remain still for long durations, we optimized our protocols so that scanning can be segmented into runs as short as 3 to 8 minutes so that participants may move or take breaks between runs. *Structural MRI:* High-resolution structural images will be acquired for anatomical referencing and image registration. Images will be acquired using a T1-weighted 3D MPRAGE sequence with the following parameters: TE=2.31ms, TR=2400ms, flip angle=8^{o}, matrix=256, FOV=256x256mm, 208 sagittal slices, thickness=1mm, bandwidth=210

Hz/pixel. Total scan duration is 5 min, 54 s. *Simultaneous fMRI and Eye-Tracking:* In piloting sessions, we have had successful collections of simultaneous eye-tracking and fMRI with 4 of 4 adults and 2 of 2 children with ASD. fMRI data will be acquired using a T2*-weighted EPI imaging sequence with the following parameters: TE=25ms, TR=2s, flip angle=90^{o}, matrix=64x64, FOV=224 mm, 37 axial slices without gap, thickness=3.5mm, BW=2604 Hz/pixel. Age-appropriate movie scenes, as in Rice et al[78], will be presented for a total scan time of approximately 15 minutes. **Participant Characterization:** Children will be characterized with measures of social ability (the SCQ[62] and the SRS[63]), and intellectual functioning (the Differential Abilities Scale[64]). Social disability in ASD will be characterized with the ADOS[65]. *TD Inclusionary Criteria:* (1) no developmental delays (all IQ scores > 70); (2) does not meet criteria for ASD on the SCQ[62]; (3) no family history of ASD; and (4) a consensus clinical classification as TD. *ASD Inclusionary Criteria:* (1) meet criteria for ASD on the ADOS-2[65]; and (2) receive a consensus best estimate clinical diagnosis of ASD. *Exclusionary Criteria for all children:* history of head trauma, seizures, or visual or auditory abnormalities, and any contraindication for MRI.

Data Processing: **fMRI Data:** Preprocessing will include slice timing correction, volume registration, artifact reduction, and spatial smoothing (FWHM = 5 mm). **Eye-tracking Data:** Eye movements identified as fixations will be coded relative to onscreen regions-of-interest (ROIs, i.e., face, body, and object). Moments perceived as highly salient will be determined by the following steps, as in[1]: (1) individual blink data are recorded as time series binary values indicating whether an individual is blinking or not; (2) instantaneous blink rate is calculated at each moment in the time series; (3) statistically significant blink inhibition will be identified through permutation testing. An exact statistic of $p<0.05$ will be used to identify periods of statistically significant blink inhibition (indicating moments perceived as highly salient) and periods of statistically significant increase in blink rate (indicating moments perceived as relatively less salient) (**Figure 2a**). During periods of statistically significant blink inhibition, we will identify *where* the viewers are fixating by the following steps: (1) patterns of visual scanning will be quantified at each time point by convolving a kernel density function with viewers' fixation coordinates (**Figure 3a**); (2) this process is applied to time-locked data from all viewers to derive kernel density estimates for the group, yielding a measure of where visual fixations are distributed within each movie frame (**Figure 3b**); (3) finally, viewers' fixation locations during moments of simultaneous blink inhibition and convergent visual scanning will be assessed relative to region of fixation location (face, body, or object) (**Figure 3c-d**).

Statistical Analyses: **Aim 1:** Two first-level analyses will be performed on fMRI data using a standard GLM approach: continuous data correlation and event-related regression. The former approach correlates instantaneous blink rate with fMRI signal. The advantage of this approach is that it utilizes the entire time series of blink data thereby providing higher statistical power. The latter approach estimates impulse response functions related to events of interest (*i.e.*, moments of significant blink inhibition or increased blinking). This approach focuses exclusively on significant modulation of blink rate and is therefore more robust to effects of noise. In the first analysis (continuous data correlation), the regressor will consist of group (TD or ASD) instantaneous blink rate recorded throughout the scan. The second analysis (event-related regression) will include regressors for each condition of interest: (1) moments perceived as highly salient by TD or ASD viewers, and (2) moments perceived as relatively less salient by TD or ASD viewers. A functional activation map will be obtained for each individual by an F-test contrasting model fit with and without regressors of interest. First-level analyses will then be combined into group-level analyses using a voxel-wise one-sample t-test, contrasting regression coefficients with 0. For between-group comparison, differences in regression coefficients will be tested by two-sample t-test. **Aim 2:** fMRI data will be analyzed as in Aim 1 using a standard GLM approach with regressors for each condition of interest: (1) faces perceived by TD or ASD viewers as highly salient and (2) faces perceived by TD or ASD viewers as relatively less salient.

References

1. Shultz, S.; Klin, A.; Jones, W. Inhibition of Eye Blinking Reveals Subjective Perceptions of Stimulus Salience. *Proceedings of the National Academy of Sciences* **2011**, *108* , 21270–21275.
2. Ranti, C., Ramsay, G., Klin, A., Jones, W., Shultz, S. Assessing the Use of Blink Inhibition As a Measure of an Individual's Level of Engagement with Ongoing Content. In *International Meeting for Autism Research*; Atlanta, 2014.
3. Nakano, T.; Yamamoto, Y.; Kitajo, K.; Takahashi, T.; Kitazawa, S. Synchronization of Spontaneous Eyeblinks While Viewing Video Stories. *Proceedings of the Royal Society B: Biological Sciences* **2009**, *276* , 3635–3644.
4. Klin, A.; Jones, W.; Schultz, R.; Volkmar, F.; Cohen, D. VIsual Fixation Patterns during Viewing of Naturalistic Social Situations as Predictors of Social Competence in Individuals with Autism. *Archives of General Psychiatry* **2002**, *59*, 809–816.
5. Klin, A.; Lin, D. J.; Gorrindo, P.; Ramsay, G.; Jones, W. Two-Year-Olds with Autism Orient to Non-Social Contingencies rather than Biological Motion. *Nature* **2009**, *459*, 257–261.
6. Jones, W.; Carr, K.; Klin, A. Absence of Preferential Looking to the Eyes of Approaching Adults Predicts Level of Social Disability in 2-Year-Old Toddlers with Autism Spectrum Disorder. *Archives of General Psychiatry* **2008**, *65*, 946–954.
7. Grand, R. Le; Mondloch, C. J.; Maurer, D.; Brent, H. P. Neuroperception: Early Visual Experience and Face Processing. *Nature* **2001**, *410*, 890.
8. Nelson, C. A. The Development and Neural Bases of Face Recognition. *Infant and Child Development* **2001**, *10*, 3–18.
9. Gottlieb, G. Experiential Canalization of Behavioral Development: Theory. *Dev. Psychol.* **1991**, *27*, 4–13.
10. Kanner, L. Autistic Disturbances of Affective Contact. *Nervous child* **1943**, *2*, 217–250.
11. Grelotti, D. J.; Klin, A. J.; Gauthier, I.; Skudlarski, P.; Cohen, D. J.; Gore, J. C.; Volkmar, F. R.; Schultz, R. T. fMRI Activation of the Fusiform Gyrus and Amygdala to Cartoon Characters but Not to Faces in a Boy with Autism. *Neuropsychologia* **2005**, *43*, 373–85.
12. O'Doherty, J. P. Reward Representations and Reward-Related Learning in the Human Brain: Insights from Neuroimaging. *Current Opinion in Neurobiology* **2004**, *14*, 769–776.
13. Adolphs, R. What Does the Amygdala Contribute to Social Cognition? *Annals of the New York Academy of Sciences* **2010**, *1191*, 42–61.
14. Robinson, D. L.; Petersen, S. E. The Pulvinar and Visual Salience. *Trends in Neurosciences* **1992**, *15*, 127–132.
15. Dichter, G. S.; Felder, J. N.; Green, S. R.; Rittenberg, A. M.; Sasson, N. J.; Bodfish, J. W. Reward Circuitry Function in Autism Spectrum Disorders. *Social cognitive and affective neuroscience* **2012**, *7*, 160–72.
16. Critchley, H. D. Neural Mechanisms of Autonomic, Affective, and Cognitive Integration. *The Journal of comparative neurology* **2005**, *493*, 154–66.
17. Zhang, S.; Li, C.-S. R. A Neural Measure of Behavioral Engagement: Task-Residual Low-Frequency Blood Oxygenation Level-Dependent Activity in the Precuneus. *NeuroImage* **2010**, *49*, 1911–8.
18. Engell, A. D.; McCarthy, G. Probabilistic Atlases for Face and Biological Motion Perception: An Analysis of Their Reliability and Overlap. *NeuroImage* **2013**, *74*, 140–151.
19. Saxe, R.; Kanwisher, N. People Thinking about Thinking People: The Role of the Temporo-Parietal Junction in "theory of Mind." *NeuroImage* **2003**, *19*, 1835–1842.
20. Fogassi, L.; Ferrari, P. F.; Gesierich, B.; Rozzi, S.; Chersi, F.; Rizzolatti, G. Parietal Lobe: From Action Organization to Intention Understanding. *Science* **2005**, *308*, 662–667.
21. Baumstimler, Y.; Parrot, J. Stimulus Generalization and Spontaneous Blinking in Man Involved in a Voluntary Activity. *J. Exp. Psychol.* **1971**, *88*, 95–102.

22. VanderWerf, F.; Brassinga, P.; Reits, D.; Aramideh, M.; Ongerboer de Visser, B. Eyelid Movements: Behavioral Studies of Blinking in Humans Under Different Stimulus Conditions. *Journal of Neurophysiology* **2003**, *89* , 2784–2796.
23. Kevin O'Regan, J.; Deubel, H.; Clark, J. J.; Rensink, R. A. Picture Changes During Blinks: Looking Without Seeing and Seeing Without Looking. *Visual Cognition* **2000**, *7*, 191–211.
24. Shultz, S.; Klin, A.; Jones, W. Inhibition of Eye Blinking Reveals Subjective Perceptions of Stimulus Salience in Children with Autism Spectrum Disorder. In *International Meeting for Autism Research*; San Sebastian, 2013.
25. Shultz, S.; Klin, A.; Jones, W. What Engages Children with Autism Spectrum Disorders When Viewing Naturalistic Social Scenes? In *International Meeting for Autism Research*; Toronto, 2012.
26. Schultz, R. T. Developmental Deficits in Social Perception in Autism: The Role of the Amygdala and Fusiform Face Area. *International journal of developmental neuroscience : the official journal of the International Society for Developmental Neuroscience* **2005**, *23*, 125–41.
27. Schultz, R. T.; Grelotti, D. J.; Klin, A.; Kleinman, J.; Gaag, C. Van der; Marois, R.; Skudlarski, P. The Role of the Fusiform Face Area in Social Cognition: Implications for the Pathobiology of Autism. *Philosophical Transactions of the Royal Society of London. Series B: Biological Sciences* **2003**, *358*, 415–427.
28. McCarthy, G.; Puce, A.; Gore, J. C.; Allison, T. Face-Specific Processing in the Human Fusiform Gyrus. *Journal of Cognitive Neuroscience* **1997**, *9*, 605–610.
29. Jones, W., Klin, A. Attention to Eyes Is Present But in Decline in 2-6 Month-Olds Later Diagnosed with Autism. *Nature*.
30. Grelotti, D. J.; Gauthier, I.; Schultz, R. T. Social Interest and the Development of Cortical Face Specialization: What Autism Teaches Us about Face Processing. *Developmental Psychobiology* **2002**, *40*, 213–225.
31. Pierce, K. Face Processing Occurs Outside the Fusiform "Face Area" in Autism: Evidence from Functional MRI. *Brain* **2001**, *124*, 2059–2073.
32. Dalton, K. M.; Nacewicz, B. M.; Johnstone, T.; Schaefer, H. S.; Gernsbacher, M. A.; Goldsmith, H. H.; Alexander, A. L.; Davidson, R. J. Gaze Fixation and the Neural Circuitry of Face Processing in Autism. *Nat Neurosci* **2005**, *8*, 519–526.
33. Hadjikhani, N.; Joseph, R. M.; Snyder, J.; Chabris, C. F.; Clark, J.; Steele, S.; McGrath, L.; Vangel, M.; Aharon, I.; Feczko, E.; Harris, G. J.; Tager-Flusberg, H. Activation of the Fusiform Gyrus When Individuals with Autism Spectrum Disorder View Faces. *NeuroImage* **2004**, *22*, 1141–50.
34. Klin, A.; Jones, W.; Schultz, R.; Volkmar, F.; Cohen, D. Visual Fixation Patterns during Viewing of Naturalistic Social Situations as Predictors of Social Competence in Individuals with Autism. *Archives of general psychiatry* **2002**, *59*, 809–16.
35. Chevallier, C.; Kohls, G.; Troiani, V.; Brodkin, E. S.; Schultz, R. T. The Social Motivation Theory of Autism. *Trends in cognitive sciences* **2012**, *16*, 231–9.
36. Kohls, G.; Chevallier, C.; Troiani, V.; Schultz, R. T. Social "Wanting" Dysfunction in Autism: Neurobiological Underpinnings and Treatment Implications. *Journal of neurodevelopmental disorders* **2012**, *4*, 10.
37. Sequeira, H.; Hot, P.; Silvert, L.; Delplanque, S. Electrical Autonomic Correlates of Emotion. *International Journal of Psychophysiology* **2009**, *71*, 50–56.
38. Berntson, G. G.; Cacioppo, J. T.; Grossman, P. Whither Vagal Tone. *Biological Psychology* **2007**, *74*, 295–300.
39. Shultz, S.; Lee, S. M.; Pelphrey, K.; McCarthy, G. The Posterior Superior Temporal Sulcus Is Sensitive to the Outcome of Human and Non-Human Goal-Directed Actions. *Social Cognitive and Affective Neuroscience* **2011**, *6*, 602–611.

40. Kaiser, M. D.; Hudac, C. M.; Shultz, S.; Lee, S. M.; Cheung, C.; Berken, A. M.; Deen, B.; Pitskel, N. B.; Sugrue, D. R.; Voos, A. C.; Saulnier, C. A.; Ventola, P.; Wolf, J. M.; Klin, A.; Wyk, B. C. Vander; Pelphrey, K. A. Neural Signatures of Autism. *Proceedings of the National Academy of Sciences* **2010**, *107* , 21223–21228.
41. Shultz, S.; McCarthy, G. Goal-Directed Actions Activate the Face-Sensitive Posterior Superior Temporal Sulcus and Fusiform Gyrus in the Absence of Human-like Perceptual Cues. *Cerebral Cortex* **2011**.
42. Li, Z.; Moore, A. B.; Tyner, C.; Hu, X. Asymmetric Connectivity Reduction and Its Relationship to "HAROLD" in Aging Brain. *Brain Research* **2009**, *1295*, 149–158.
43. Li, Z.; Santhanam, P.; Coles, C. D.; Lynch, M. E.; Hamann, S.; Peltier, S.; Hu, X. Increased "default Mode" Activity in Adolescents Prenatally Exposed to Cocaine. *Human Brain Mapping* **2011**, *32*, 759–770.
44. Li, Z., Santhanam, P., Coles, C.D., Lynch, M.E., Hamann, S., Peltier, S., Hu, X. Prenatal Cocaine Exposure Alters Functional Activation in Ventral Prefrontal Cortex and Its Neural Connectivity with Amygdala. *Psychiat. Res. Neuroim.* **2013**, *213*.
45. Coles, C. D.; Goldstein, F. C.; Lynch, M. E.; Chen, X.; Kable, J. A.; Johnson, K. C.; Hu, X. Memory and Brain Volume in Adults Prenatally Exposed to Alcohol. *Brain and Cognition* **2011**, *75*, 67–77.
46. Chen, X.C., Coles, C.D., Lynch, M.E., Hu, X. Understanding Specific Effects of Prenatal Alcohol Exposure on Brain Structure in Young Adults. *Human Brain Mapp.*
47. Santhanam P, Li Z, Lynch ME, Coles CD, and H. X. Default Mode Network Dysfunction in Adults with Prenatal Alcohol Exposure. *Psych Res: Neuroimaging*.
48. Li, L.; Hu, X.; Preuss, T. M.; Glasser, M. F.; Damen, F. W.; Qiu, Y.; Rilling, J. Mapping Putative Hubs in Human, Chimpanzee and Rhesus Macaque Connectomes via Diffusion Tractography. *NeuroImage* **2013**, *80*, 462–474.
49. Li, L.; Rilling, J. K.; Preuss, T. M.; Glasser, M. F.; Hu, X. The Effects of Connection Reconstruction Method on the Interregional Connectivity of Brain Networks via Diffusion Tractography. *Human Brain Mapping* **2012**, *33*, 1894–1913.
50. Li, L.; Rilling, J. K.; Preuss, T. M.; Glasser, M. F.; Damen, F. W.; Hu, X. Quantitative Assessment of a Framework for Creating Anatomical Brain Networks via Global Tractography. *NeuroImage* **2012**, *61*, 1017–1030.
51. Jones, W.; Klin, A. Heterogeneity and Homogeneity across the Autism Spectrum: The Role of Development. *Journal of the American Academy of Child and Adolescent Psychiatry* **2009**, *48*, 471–3.
52. Shultz, S.; Vouloumanos, A. Three-Month-Olds Prefer Speech to Other Naturally Occurring Signals. *Language Learning and Development* **2010**, *6*, 241–257.
53. Pelphrey, K. A.; Shultz, S.; Hudac, C. M.; Wyk, B. C. Vander Research Review: Constraining Heterogeneity: The Social Brain and Its Development in Autism Spectrum Disorder. *Journal of Child Psychology and Psychiatry* **2011**, *52*, 631–644.
54. Call, N. A.; Pabico, R. S.; Lomas, J. E. Use of Latency to Problem Behavior to Evaluate Demands for Inclusion in Functional Analyses. *Journal of Applied Behavior Analysis* **2009**, *42*, 723–728.
55. Call, N. A.; Findley, A. J.; Reavis, A. R. The Effects of Conducting a Functional Analysis on Problem Behavior in Other Settings. *Research in Developmental Disabilities* **2012**, *33*, 1990–1995.
56. Call, N.; Parks, N.; Reavis, A. Treating Severe Problem Behavior Within Intensive Day-Treatment Programs. In *Handbook of Crisis Intervention and Developmental Disabilities SE - 21*; Reed, D. D.; DiGennaro Reed, F. D.; Luiselli, J. K., Eds.; Issues in Clinical Child Psychology; Springer New York, 2013; pp. 367–391.
57. Shillingsburg, M. A.; Valentino, A. L.; Bowen, C. N.; Bradley, D.; Zavatkay, D. Teaching Children with Autism to Request Information. *Research in Autism Spectrum Disorders*

2011, *5*, 670–679.
58. Shillingsburg, M. A.; Kelley, M. E.; Roane, H. S.; Kisamore, A.; Brown, M. R. Evaluation and Training of yes-no Responding across Verbal Operants. *Journal of Applied Behavior Analysis* **2009**, *42*, 209–223.
59. Call, N.A., Trosclair-Lasserre, N.M., Findley, A.J., Reavis, A., Shillingsburg, M. A. Correspondence between Single versus Daily Preference Assessment Outcomes and Reinforcer Efficacy under Progressive-Ratio Schedules. *J. Appl. Behav. Anal.* **2012**, *45*.
60. Power, J. D.; Barnes, K. A.; Snyder, A. Z.; Schlaggar, B. L.; Petersen, S. E. Spurious but Systematic Correlations in Functional Connectivity MRI Networks Arise from Subject Motion. *Neuroimage* **2012**, *59*, 2142–2154.
61. Desmond, J. E.; Glover, G. H. Estimating Sample Size in Functional MRI (fMRI) Neuroimaging Studies: Statistical Power Analyses. *Journal of Neuroscience Methods* **2002**, *118*, 115–128.
62. Rutter, M., Bailey, A., Lord, C. Social Communication Questionnaire. *Western Psychological Services* **2003**.
63. Constantino, J. N.; Gruber, C. P. The Social Responsiveness Scale. *Los Angeles: Western Psychological Services* **2002**.
64. Elliot, C. D. *Differential Ability Scales - Second Edition*; Harcourt: San Antonio Tx, 2007.
65. Lord, C. Autism Diagnostic Observation Schedule, Second Edition. *Western Psychological Services* **2012**.

Extramural Funding Track Record: Drs. Shultz (PI on the current application), Li, and Jones (Investigators on the current application) were awarded a Pediatric Center Seed Grant in 2014 ('Acquiring high-quality magnetic resonance data for understanding brain connectivity in infants at-risk for autism'). This seed grant generated pilot data that led to an NIH-funded R21 (submitted within the 1st year of the Pediatric Seed Grant) and an NIH K01 grant application (PI: Dr. Shultz; submitted within the first year following the Pediatric Seed Grant). The K01 application was favorably reviewed (impact score of 12) and is likely to be funded for a 5-year period beginning in September 2016. *Given the funding amounts of the R21 ($362,436) and the K01 ($871,770), the return on investment of the 2014 Pediatric Center Seed Grant ($50,000) is likely to be 2368.4%.* In addition to securing external funding, the seed grant lay the foundation for the creation of a new 'Pediatric Neuroimaging Core' to expand research at the Marcus Autism Center to include investigation of the neural mechanisms underlying ASD. The work supported by this seed grant led to the development of novel infant-optimized MRI protocols that, relative to conventional techniques, offer significant improvements in the speed and quality of data acquisition from naturally sleeping infants. These methodological breakthroughs were critical in establishing MRI protocols and infrastructure for the first-ever pediatric neuroimaging facility at the Wesley Woods Center for Systems Imaging (a combined Emory/CHOA/Marcus Autism Center initiative, led by Drs. Shultz and Li). ***Future Plans Related to Current Proposal:*** Just as our 2014 Pediatric Seed Grant launched a program of research on brain development in *infancy*, the current application will launch a program of research focused on understanding *the neural mechanisms of social disability in school-age children with ASD*. The innovative technologies and behavior protocols supported by this application will create the infrastructure for a new program of research in developmental neuroimaging and will produce pilot data that will lead directly to an NIH R01 application (PA-13-216, 'Research on Autism Spectrum Disorders', to be submitted October 5, 2017, estimated 5-year budget of $1,750,000). The focus of the present grant application—identifying neural markers of social disability in children with ASD—is well aligned with both the research objectives of the R01 funding announcement ("studies of brain mechanisms underlying the development, regulation, and modulation of behaviors characterizing ASD, particularly those involved in communication and social interaction") and the funding priorities identified by the NIH Interagency Autism Coordinating Committee ("neural markers of autism and neural mechanisms of social disability"), strengthening the prospect of future NIH grant applications. ***Leveraging of Resources:*** This application brings together the unique strengths of several research cores at the Marcus Autism Center: (1) the Pediatric Neuroimaging Core (expertise in pediatric brain imaging); (2) the Social Neuroscience Core (expertise in eye-tracking measures of child behavior in response to naturalistic social stimuli); and (3) the Behavior Treatment Clinics (expertise in behavior analysis and behavior modification). Collaborations between the Pediatric Neuroimaging Core and the Social Neuroscience Core will lead to the development of novel technology for simultaneous measurement of a child's engagement with naturalistic social stimuli (via eye-tracking) and brain activity (via fMRI). Collaborations between the Pediatric Neuroimaging Core and the Behavior Treatment Clinics will lead to the development of behavior training protocols to teach children (including those with severe intellectual and social disabilities) the skills to successfully complete an MRI scan, a unique resource not commonly available to pediatric neuroimaging studies. These collaborations will set the stage for a generative interdisciplinary research program by opening new avenues to the study of social disability in a sample of children that is actually representative of the range of intellectual functioning and social disability characteristic of ASD. These efforts are also likely to stimulate additional collaborations and interactions with GRA-affiliated colleagues. For instance, our simultaneous eye-tracking and fMRI technology is currently used by Emory's Department of Radiology and Imaging Sciences, while our behavior training protocols have been shared with Emory colleagues in Psychology and Psychiatry.