Piloting a Battery to Evaluate Parasympathetic Reactivity and Externalizing Behaviors during Early Childhood in Autism Spectrum Disorder

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Abstract

Background Respiratory Sinus Arrhythmia Reactivity (RSA-R) correlates both positively and negatively with externalizing behavior in autistic individuals. These inconsistencies may result from task-based differences. Furthermore, RSA-R is understudied in young autistic children, despite particular challenges with externalizing behavior. This pilot study measured RSA-R in 4-to 6-year-olds, across two time-points, using four validated tasks with matched baseline and challenge periods. Methods RSA and parent-reported externalizing behavior were collected from 17 children (Mage=5.57 years). RSA-R was measured by the difference score of the challenge task minus its corresponding comparison task. Correlations were computed to evaluate relationships between RSA-R and behavior. Results RSA was reliably measured for 3/4 tasks (.694 [?] ICCs [?] .896). Only RSA-R during a social task correlated with externalizing behavior. These results support using a battery that measures a range of challenges, differing in social demands, to characterize how arousal contributes to emotion regulation demands among young autistic children.

Introduction

Autism spectrum disorder (ASD) is a neurodevelopmental disability characterized by the presence of restricted and repetitive behaviors and challenges with social communication (American Psychiatric Association, 2013). For autistic children, externalizing behaviors (e.g., aggression and hyperactivity) are more prevalent compared to neurotypical peers (McClintock, Hall, & Oliver, 2003, Mahan and Matson, 2011, Totsika et al. 2011) and are linked with worsened learning opportunities (Lauderdale-Litten et al., 2013), lower peer acceptance (Burt et al., 2008; Deater-Deckard., 2001), and poor quality of life (Kuhlthau et al. 2010). Externalizing behaviors also relate to ASD features, namely emotional response and social skills (Neuhaus et al., 2014; Mazefsky, Pelphrey, & Dahl, 2012; Samson, Hardan, Lee, Phillips, & Gross, 2015). Delineating biological measures of externalizing behavior may identify autistic children who would benefit from intervention and promote better educational and health-related outcomes.

Physiological measures can be particularly useful for characterizing externalizing behaviors in autistic children. Difficulties with social-emotional reciprocity and communication are hallmarks of ASD (American Psychiatric Association, 2013; Wimpory et al. 2007; Joseph and Tager-Flusberg 2004; Klin et al. 2007). Therefore, young autistic children may have trouble identifying or articulating their emotions to another individual. Emotion regulation is also impacted in ASD (Fenning et al. 2019) and relates to externalizing behaviors in older children (Ting and Weiss 2017). Physiological variables derived from heart rate relate to parent reports of externalizing behavior (Baker et al., 2019; Neuhaus et al., 2014), and, importantly, provide an objective measurement regardless of age, verbal ability, and diagnostic status.

One physiological measure linked with self-regulatory behaviors is respiratory sinus arrhythmia (RSA), an index of cardiac activity in the parasympathetic nervous system (PNS). The PNS slows heart rate through vagal nerve input, and RSA captures the variability of heart rate across respiration. RSA reactivity (RSA-R), also known as RSA withdrawal, reflects the change in RSA from baseline to a stressor. High RSA and RSA-R are thought to reflect greater PNS control and thus greater regulatory control in response to environmental stressors (Porges, 1995, 2007; Beauchaine et al. 2007, Gyurak and Ayduk, 2008; Scarpa et al., 2010).

While RSA has been identified as a reliable physiological measure of emotion regulation (Beauchaine et. al 2007, Boyce et al. 2001, Graziano & Derefinko, 2013), RSA-R and its relationship to externalizing behaviors in autism are poorly understood. Previous research in autism is mixed, with some evidence of a negative relation and some reports of no relation (Cheng et al., 2020, Neuhaus et al., 2014) In one study, the relation between RSA-R and externalizing behavior was moderated by negative parent behavior (Baker et al. 2019), highlighting the complex role of social-emotional factors in the expression of externalizing behavior by autistic children. RSA-R accounted for dysregulated behaviors beyond the presence of autism features (Baker et al., 2022), and its relation to externalizing behavior depended on co-occurring sympathetic activity levels (Fenning et al., 2019). While these initial findings suggest that RSA-R is relevant for externalizing behaviors in autistic samples, more work is needed to characterize RSA in young autistic children.

To best characterize RSA-R, batteries require appropriately matched challenge and comparison tasks. Challenge tasks across different domains (e.g., cognitive versus social) may differentially probe PNS reactivity (Burt & Obradović, 2013). Further, measuring RSA change from a challenge task in one modality to an unrelated baseline task does not account for domain-specific baseline responses (Bush et al., 2011; Kamarck, Jennings, and Mannick 1993) or differences in task-related artifacts (e.g., movement), which is particularly important when sampling from autistic children who may have poorer motor control (Licari et al. 2020; Liu & Breslin, 2013). While reactivity paradigms with challenge and matched comparison tasks across modalities are available (Bush et al., 2011; Kamarck and Lovallo, 2003), we could find no published studies using this design in young autistic children.

The current pilot study addressed these limitations by evaluating parasympathetic reactivity and externalizing behaviors in autistic children. Our aims included the following:

Aim 1: Evaluate the feasibility and stability of RSA measured via challenge and matched comparison tasks across four distinct domains: cognitive, social, emotional, and sensory.

Aim 2: Explore whether RSA-R elicited by tasks with feasible and stable RSA response relates to parent reported externalizing behavior.

Given the primary goal was evaluating a physiological battery for the first time in ASD, we had no hypotheses regarding specific task domains. Broadly, we hypothesized that for any tasks with stable RSA, lower RSA-R, indicative of greater RSA withdrawal and reduced emotion regulation, would relate to increased externalizing behavior.

Materials & methods

Participants

Participants were recruited from a larger, ongoing study of cognitive control development in early childhood at [details omitted for double-anonymized peer review]. Participants were originally recruited using research registries, community events and advocacy groups, clinics, early intervention programs, and word of mouth. Exclusionary criteria included medical disorders or medications impacting the central nervous system, a history of seizures or seizure medication, inability to complete parent questionnaires or testing in English, prolonged prenatal substance exposure, and significant sensory or motor impairments or major physical abnormalities which limited ability to complete the testing battery.

Participants included 19 verbal autistic children (16 male) aged 3.9 to 6.8 years (M_{age} = 5.6, SD = 0.82 years) at their initial visit. Sixteen children returned for the follow-up visit aged 4.0 to 6.9 years (M_{age} = 5.6

SD = 0.89). All participants used flexible phrase speech and had an age equivalent of at least 18 months as assessed via the Visual Reception domain of the Mullen Scales of Early Learning (MSEL; Mullen, 1995) as part of their participation in the larger study within the previous two years. Diagnosis of an autism spectrum disorder was determined using the ADOS-2 (Lord, Luyster, et al., 2012, Lord, Rutter et al., 2012) and the ADI-R (Rutter et al., 2003; Kim & Lord, 2012) and DSM-5 criteria (American Psychiatric Association, 2013) based on expert judgment of a licensed clinical psychologist11During the COVID-19 pandemic, the CARS-2 was added to the protocol to further confirm that children met standardized criteria for an autism spectrum disorder.. Sample characteristics are reported in Table 1.

Procedure

The Institutional Review Board at [details omitted for double-anonymized peer review] approved all study procedures and all parents provided written consent for their children to participate. Table 1 provides demographic and measure characteristics, respectively. Prior to study visits, families were provided with a social story and habituation protocol to familiarize children with the physiological recording equipment. Caregivers and their children completed two visits to the lab separated by 4-6 weeks. During both visits children completed the MacArthur Reactivity protocol (MRP; Bush et al. 2011). If children were comfortable, ECG was recorded during the MRP. Parents completed the Behavior Assessment System for Children (BASC) Parent Rating Scale – Preschool or Child (PRS-P or PRS-C, respectively).

Measures

Externalizing Behavior - Behavior Assessment System for Children-Second Edition (BASC-2)

Externalizing Behavior (EB) was measured via parent report using the Externalizing Problems composite on the Behavior Assessment System for Children (BASC-2, Reynolds & Kamphaus, 2004). The preschool form used with 2- to 5-year-olds (PRS-P) yields an externalizing composite comprised of the Hyperactivity and Aggression subscales. The child form used with 6- to 11-year-olds (PRS-C) yields an externalizing comprised of the Hyperactivity, Aggression, and Conduct subscales.

Emotion Regulation Paradigm - MacArthur Reactivity Protocol (MRP)

The MacArthur Reactivity Protocol (Bush et al. 2011) includes challenges across four different task domains and paired baseline tasks. Challenges include a social task (talking with an experimenter); a cognitive task (digit span); a sensory task (receiving two drops of lemon juice on the tongue); and an emotional task (watching an age-appropriate video clip intended to elicit fear). Baseline tasks include talking about pictures in a book for the social domain, an easier digit span task in the cognitive domain, identifying a drop of water for the sensory domain, and watching a neutral video clip for the emotional domain. Baseline tasks were designed to account for the psychomotor demands of the corresponding challenge task and each task lasted up to two minutes.

ECG collection and RSA-R quantification

RSA-R was extracted from each participant's electrocardiogram (ECG) using the following steps. ECG was acquired continuously at 2000 Hz using a BIOPAC MP150 system with ECG 100C amplifiers and analyzed offline using the AcqKnowledge 5.0.6 software (Biopac Systems, 2018). The ECG signal was recorded during the MacArthur Reactivity Protocol using the ECG100C amplifier with three pre-gelled Ag/AgCl electrodes with a lead II configuration.

Offline, ECG signals were bandpass filtered using an FIR filter from 0.5 to 35 Hertz. Next, R-R intervals were extracted for the ECG signal using a modified Pan-Tompkins QRS detector (Pan & Tompkins, 1985) and resampled to 8hz after cubic spline interpolation. Data were then visually inspected for movement artifacts or incorrect identification of the QRS complex by the scoring algorithm.

RSA values were computed using a fast Fourier transform with a hamming window within the respiratory frequency band that corresponded to the participant's age (0.2-1.0Hz for children under six, 0.15-0.5Hz for children six and older; Boyce et al., 2001, Gentzler et al. 2009). RSA values were computed in 60 second

epochs and log transformed. RSA during the first and last 60 seconds was averaged for each baseline and challenge task, respectively. RSA reactivity (RSA-R) reflected the difference score for each challenge task minus its corresponding comparison task11The psychometric properties of using a regression residual are preferable, but more difficult to interpret. We computed all analyses with both difference scores and with the regression residual and all results were identical. Difference scores are reported to increase ease of interpretation..

Analytic plan

We characterized the feasibility of ECG collection in this sample (Aim 1) by computing the percent of collected, and subsequently usable, ECG data for each MRP task across both visits. For participants with usable ECG data at both visits, we ran intraclass correlations with RSA values for corresponding tasks at visits 1 and 2 to assess the test-retest stability of each RSA measure (Aim 1). ICC values were interpreted as poor (< 0.4), fair (0.4-0.59), good (0.6-0.74), and excellent (0.75-1.0; Cicchetti, 1994). Finally, for tasks with adequate ICCs, we tested whether RSA-R related with concurrent externalizing behavior (Aim 2) at visit 1 via bivariate Pearson's correlations. Before conducting analyses, we first confirmed that age and developmental level were not related to RSA or externalizing scores.

According to Bujang & Baharum (2017) with 80% power, 15 subjects with data at both timepoints are required to detect an ICC of 0.6 (i.e., good). Seven subjects with data at both timepoints are required to detect an ICC of 0.8 (i.e., excellent reliability). For Aim 2, data from 25 children would enable detection of a large effect size ($f2 = 0.35$) with power of 0.8 and $\alpha = 0.05$. Fewer children with required language levels were available to participate given the COVID pandemic, so we may not detect all large effects for concurrent validity estimates. Given the study was designed to pilot the initial feasibility and stability, we believe the data reported could still be useful for planning future studies.

Results

Feasibility and Stability of MRP

Feasibility (% acquired data) and Stability (ICC) for each MRP task are reported in Table 2. At each visit, most of our sample tolerated application of the ECG equipment and provided data. In addition, RSA values during three of the four challenge tasks had good or excellent stability. Specifically, RSA stability during the social (.694, 95% CI [.197 .907]) and emotional (.693, 95% CI [.157 .940]) challenges fell in the good range, and the cognitive challenge had excellent stability (.817 [.424 .951]). In contrast, the sensory challenge did not yield adequate feasibility with only 10 participants completing both visits and stability in the fair range (.490, $p > .05$).

Convergent Validity with Externalizing Behavior

Externalizing behavior was negatively correlated with RSA-R recorded during the social condition (r = -.673, $p = .008$). As RSA-R decreased, indicating withdrawal, externalizing symptomatology increased (see Figure 1). Externalizing behavior was not significantly related to RSA-R measured during the cognitive or emotional conditions (r s < 0.468, p s $> .05$). Descriptive statistics of RSA-R across domains and correlations with externalizing behavior can be found in Table 3.

Discussion

This study is the first to evaluate the feasibility of the MRP and examine domain-specific RSA-R in relation to externalizing behaviors in young autistic children. Physiological data collection was feasible for young children, with adequate completion and good to excellent stability of the cognitive, emotional, and social tasks, and lower completion rate and stability of the sensory task (Aim 1). Increased RSA withdrawal during a social challenge correlated with higher levels of externalizing behavior (Aim 2). Taken together, our findings highlight the feasibility of eliciting a reliable RSA response and underscore a potential relationship between PNS reactivity in social contexts and externalizing behaviors.

Our results support the use of the MRP. Acquisition rates suggestthat the social, cognitive, and emotional tasks are feasible in young autistic children with phrase speech or fluent language. Notably, fewer children wore sensors for the full sensory condition (Visit 1: 68%; Visit 2: 63%) compared with the other three tasks (79-84%). Given heightened sensory sensitivities in this population (Chistol et al., 2018), gustatory sensory tasks may not be sufficiently feasible to elicit a stable RSA signal. Nonetheless, additional task refinement to increase acquisition rates may provide essential information about sensory reactivity for a subgroup of young autistic children.

The negative relationship between externalizing behaviors and socially elicited RSA-R highlights the value of measuring domain-specific reactivity. Decreased RSA-R, or increased withdrawal, may reflect physiological dysregulation and limited coping responses to challenging situations (Graziano & Derefinko, 2013; Mezzacappa et al., 1997; Beauchaine, 2001). However, both RSA-withdrawal and reactivity may be adaptive depending on the context (Fenning et al. 2019). Indeed, 3.5-year-olds with elevated internalizing behaviors and moderate baseline RSA levels had fewer longitudinal externalizing problems than those with high or low baseline RSA (Ugarte et al. 2021). Characterizing physiological reactivity across domains could determine optimal domain-specific or domain-independent responses.

The specificity of the relation between externalizing behavior and RSA-R in the social domain replicates work in neurotypical individuals and may highlight the battery's sensitivity to autism-specific regulatory demands. In a meta-analysis, only tasks using negative emotional reactivity RSA for non-autistic adults (Beauchaine et al., 2019). Although cognitive and emotional differences may be impacted by the heterogeneity of ASD, reduced social functioning is a core feature of autism. Consequently, engaging in face-to-face interactions may place the greatest self-regulatory demands on autistic children and best probe reactivity. Indeed, difficulty adapting to social norms and reading social cues has been linked to externalizing behaviors among autistic individuals (Shea et al., 2018). Therefore, socially induced RSA may reflect mechanisms linked with externalizing behaviors or better elicit negative emotional reactivity amongst autistic children.

Limitations/Future Directions

The current study has several limitations. Our pilot sample was recruited from an ongoing longitudinal study, so feasibility may be influenced by the familiar research setting or increased adherence to habituation protocols at home. Still an unfamiliar team member administered all procedures to ensure the social challenges were comparable. The convenience sample also prevented concurrent measurement of developmental level. Additionally, MRP task demands excluded children using single or no words. Taken together, experimental design modifications and larger sample sizes may elucidate how RSA relates to externalizing behavior in a more representative sample of autistic children.

Future studies may also benefit from collecting additional physiological measures. While there is no "gold-standard" method for computing RSA, methods are differentially impacted by artifact (Larsen et al., 2010; Lewis et al., 2012) and by measuring RSA using obtained respiration versus normative respiration levels (Graziano & Derefinko, 2013). Studies collecting both ECG and respiration could directly test the impact of different RSA computation methods. Furthermore, given RSA and behavioral outcomes may be dynamically impacted by co-occurring sympathetic activity (Fenning et al., 2019), obtaining measures of PEP or galvanic skin response could further elucidate relations between physiological response and externalizing behavior.

Conclusion

The current study provides preliminary support for the use of a domain-specific approach to examining RSA-R in autistic children. By evaluating physiological responses during social, cognitive, emotional, and sensory challenges, we can more precisely characterize how autistic children self-regulate across contexts and identify what kinds of physiological reactivity are most relevant for externalizing behavior for autistic children. The current study is the first to find a relationship between socially induced RSA-R and externalizing behavior among young autistic children. Further, the feasibility of the current protocol is encouraging for replication in a larger sample. Understanding the physiological mechanisms underlying externalizing behaviors supports the development of objective methods to identify which children may respond best to behavioral support or

intervention to optimize behavioral outcomes.

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Table 1. Sample Characteristics

	N (%)
Assigned Sex Male	16 (84.2%)
Female	3 (15.8%)
Race American Indian/Alaskan Native	0 (0.0%)
Asian	2 (10.5%)
Hawaiian/Pacific Islander	0 (0.0%)
Black/African American	2 (10.5%)
White/Caucasian	14 (73.7%)
More than one Race	0 (0.0%)
Unknown/Unreported	0 (0.0%)
Other	1 (5.3%)
Ethnicity Non-Hispanic/Latine	18 (94.7%)
Hispanic/Latine	1 (5.3%)

Table 2. Feasibility and Stability of MRP

RSA by Task	V1 Wore sensor for full task N (%)	V1 Data usable N (%)	V2 Wore sensor for full task N (%)
Cognitive Challenge	15 (79%)	14 (74%)	13 (81%)
Cognitive Comparison	15 (79%)	14 (74%)	13 (81%)
Social Challenge	16 (84%)	15 (79%)	13 (81%)
Social Comparison	16 (84%)	15 (79%)	13 (81%)
Sensory Challenge	13 (68%)	11 (58%)	10 (63%)
Sensory Comparison	15 (79%)	14 (74%)	13 (81%)
Emotional Challenge	16 (84%)	14 (74%)	13 (81%)
Emotional Comparison	16 (84%)	14 (74%)	13 (81%)

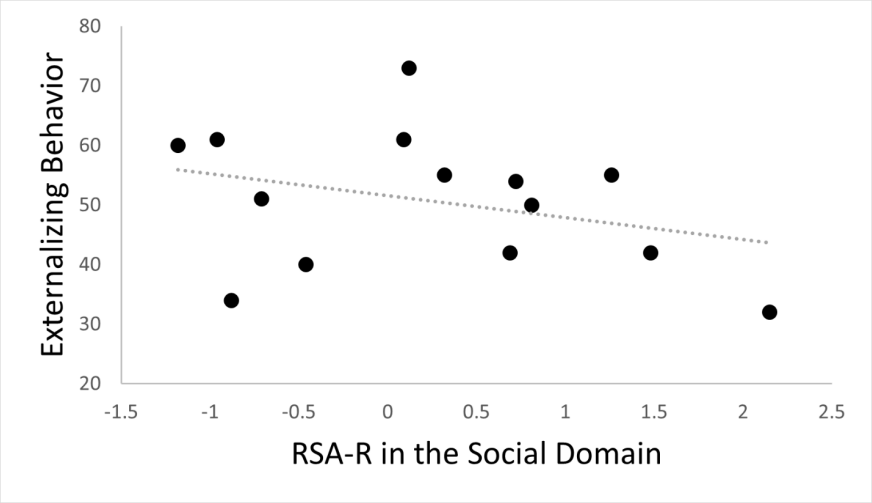
Note: Feasibility is defined as the % of children who tolerated the sensor and provided usable data; Reliability is defined by the ICC computed as single measure; T1 = Time 1; T2 = Time 2; * reflects a p -value less than .01, ** reflects a p -value less than .001. During the sensory task, participants were counted if they completed both lemon juice tastes.

Table 3. RSA-R Descriptives and Correlations with Externalizing Behavior

RSA-R Domain	Mean (Standard Deviation), Range	Correlation with BASC Externalizing
Social	.105 (.922), -1.40 to 2.00	-.673**
Emotional	-.21 (.021), -2.06 to 0.64	.458
Cognitive	.100 (.504), -0.72 to 1.22	.468

Note: ** reflects a *p-value* less than .001

Figure 1. Externalizing Behavior Decreases with Increased Social RSA-R



Notes: RSA-R in the Social Domain computed as the difference between the Social challenge and Talking Baseline tasks, Externalizing Behavior drawn from the BASC Externalizing Behavior T-score.