Introduction: Previous literature demonstrates that youth with Autism Spectrum Disorder (ASD) can present with hyper- and hypo-aroused behavioral profiles; however, objective physiological measures of arousal are seldom used in these studies, especially in youth with ASD who are severely affected. The objective of this study was to record inter-beat interval (IBI), respiratory rate (RR), and respiratory sinus arrhythmia (RSA) responses to experimentally presented environmental stressors in youth with ASD and typically developing (TD) controls. We hypothesized there would be significantly greater cardiorespiratory reactivity to a greater number of environmental stressors in severely affected youth with ASD compared to age-matched TD youth.

Method: Two groups of youth with parental consent participated in this study. The first group consisted of 41 DSM-IV confirmed youth with ASD (75% male), aged 4 to 25 yrs (M = 13 yrs, SD = 5 yrs). Bayley Scales of Infant Development—Second Edition scores ranged from 24 to 38 (M = 31), which is below the cutoff for “moderately impaired or delayed,” and Childhood Autism Rating Scale scores spanned 31.5 to 43.5 (M = 39), placing the sample in the moderately to severely autistic range. The ASD group was subdivided into those taking one or more medications (ASDmed, n = 21, aged 4 to 25 yrs) versus those not prescribed medications (ASDnomed, n = 20, aged 6 to 20 yrs). Four male TD youth ranging in age from 8 to 18 yrs (M = 14.8 yrs, SD = 4.6 yrs) without documented comorbid disorders (e.g., attention-deficit/ hyperactivity disorder, obsessive-compulsive disorder, seizure disorder) served as a reference group.

All participants were seated in a comfortable chair in a sound-attenuated room with low-level incandescent lighting. A familiar person accompanied each participant to increase comfort with the setting and experimental conditions. A wide sample of potential stressors was selected to ensure multiple opportunities for participants to elicit cardiorespiratory responses. The observational design for each participant consisted of 14 phases. Each session began with a 5-minute baseline condition (sitting quietly with a familiar person). After the baseline condition, participants engaged in 7 potentially stressful tasks. These potentially stressful tasks alternated with 2-minute rest periods (sitting quietly with a familiar person). A physical exertion task (riding a stationary bicycle for 2 minutes) was included to ensure participants could demonstrate cardiorespiratory changes significantly greater than baseline. The protocol ended with this task. Tasks were intentionally not counterbalanced to permit direct comparisons between and within ASD and TD groups. The stress tasks were included in the study because they (a) included events that frequently occur naturally in the environment; (b) could be replicated in an experimental setting; (c) represented physical, social, and cognitive stimuli that overlap considerably with socialization, communication, and behavioral features of ASD; and (d) were shown to elicit increased cardiovascular activity in prior studies (Goodwin et al., 2006; Groden et al., 2005). The stress tasks were ordered as follows: (1) Loud Noise, a vacuum cleaner runs outside the room; (2) Robot, a remote control robot navigates around the room; (3) Unstructured Time, participant is left sitting in the room alone, given only the instruction “We will be back in 2 minutes.”; (4) Eating a Preferred Food, participant is given a preferred food to eat; (5) Difficult Task, participant is asked to fold a towel the same way the familiar person folds it. When the participant folds the towel incorrectly, the familiar person is instructed to say, “Try it this way.”; (6) Change in Staff, familiar person leaves the room and a person unfamiliar to the participant enters the room and sits; and (7) Physical Exercise, while seated on a stationary bike, the participant is instructed to ride as fast as he or she can for two minutes.

Ambulatory cardiorespiratory responses were recorded at 1kHz using the LifeShirt (Vivometrics, Inc.), a noninvasive telemetric recording device that stores continuous electrocardiograph data on a portable battery-powered electronic recorder worn on the body (Wilhelm, Roth, & Sackner, 2003). All data stored in the electronic recorder were downloaded to a PC computer as ASCII files and subsequently analyzed in MindWare (HRV version 3.0.22, MindWareTech, Inc., Columbus, Ohio) to produce estimates of inter-beat interval (IBI), respiratory rate (RR), and respiratory sinus arrhythmia (RSA).

Resulting data for IBI, RR, and RSA were averaged in order to contrast pooled measures of the following three domain-related conditions: (1) Rest (i.e., mean of the seven resting epochs); (2) Psychological Challenge (i.e., mean of five epochs: Loud Noise, Robot, Unstructured Time, Difficult Task, Change of Staff); and Physical Challenge (i.e., mean of two epochs: Eating, Exercise). We used a mixed-model ANOVA framework, with type of condition constituting a repeated factor and ASD status (i.e., ASDmed, ASDnomed, TD) constituting a between-groups factor. Given sizeable differences in the number of participants in the TD versus ASD groups, we also analyzed data using a multilevel model (MLM) with type of situation nested within individuals. This approach served as a check on the robustness of that offered by the more familiar ANOVA framework. In all cases, results from MLM
analyses supported findings identified using the mixed-model ANOVA approach. Thus, we present findings using that more familiar interpretative framework.

**Results:** Subgroup differences in demographic variables were assessed by one-way ANOVA (age) and chi square (sex distribution and verbal status). The three subgroups did not differ in mean age, \( F(2, 40) = 0.29, p > .70 \), and there were no significant subgroup differences in sex distribution (\( \chi^2 = 1.26, p > .50 \)) or verbal status (\( \chi^2 = 4.23, p > .35 \)).

Test-retest reliability of the autonomic measures was assessed in a subset of 5 participants with ASD (ASD_{med} = 3, ASD_{nomed} = 2; aged 9 to 20 yrs) who completed the full protocol on two occasions, ranging from 1 to 7 months apart. Paired t-tests indicated no significant differences for this subset between Time 1 and Time 2 in any of the measures, all ps > .50. Also, ANOVAs for each measure at Time 1 and Time 2 demonstrated sufficient statistical power within the subset to show significant change among conditions.

For IBI, a 3 (Group: ASD_{med}, ASD_{nomed}, TD) X 3 (Condition: Rest, Psychological Challenge, Physical Challenge) mixed ANOVA revealed a main effect for condition, \( F(1,18, 49.50) = 84.15, p < .001 \), partial \( \eta^2 = .67 \), a main effect for group \( F(2, 42) = 12.15, p < .001 \), partial \( \eta^2 = .37 \), and an interaction between the two, \( F(2.36, 49.50) = 4.89, p = .008 \), partial \( \eta^2 = .19 \). The interaction was ordinal in nature, allowing main effects to be interpreted. With respect to condition, a planned contrast showed that IBI significantly decreased when participants were experiencing a physical challenge compared to a psychological challenge or resting state, \( F(1, 44) = 82.24, p < .001 \). However, as documented by the interaction (and supported by MLM analyses), the steepness of the decline from IBI in rest and psychological challenge conditions to that of physical challenge was greater in the TD group than in either ASD group. With respect to ASD groups, Fisher’s LSD tests revealed that both ASD groups showed reduced IBI across conditions compared to TDs (\( p's < .001 \)). However, the two ASD groups did not differ in IBI from each other (\( p = .140 \)).

For RR, a 3 (Group: ASD_{med}, ASD_{nomed}, TD) X 3 (Condition: Rest, Psychological Challenge, Physical Challenge) mixed ANOVA again revealed two main effects. As anticipated, the main effect for condition, \( F(1.43, 57.37) = 42.59, p < .001 \), partial \( \eta^2 = .52 \), could again be explained by an expected increase in RR as a function of facing a physical challenge compared to a psychological one or a resting state, \( F(1, 42) = 84.59, p < .001 \). With respect to group differences, a main effect also emerged, \( F(2, 40) = 4.83, p = .013 \), partial \( \eta^2 = .20 \). Although Fisher LSD comparisons clearly showed that ASD participants who were taking medication showed significantly greater RR across conditions compared to TDs (\( p = .005 \)), ASD participants who were unmedicated showed marginal differences from both ASD participants who were medicated (\( p = .089 \)) and TD controls (\( p = .065 \)), suggesting a RR that fell in between these two.

For RSA, a 3 (Group: ASD_{med}, ASD_{nomed}, TD) X 3 (Condition: Rest, Psychological Challenge, Physical Challenge) mixed ANOVA revealed two main effects. As expected, the main effect for condition, \( F(1.19, 49.77) = 35.36, p < .001 \), partial \( \eta^2 = .46 \), could again be explained by an anticipated decrease in RSA as a function of facing a physical challenge compared to a psychological one or a resting state, \( F(1, 44) = 51.07, p < .001 \). Of greater interest, however, was a main effect for group, \( F(2, 42) = 5.76, p = .006 \), partial \( \eta^2 = .22 \). Here, Fisher LSD comparisons showed that ASD participants who were taking medication evidenced lower RSA than nonmedicated ASD participants or TD controls (\( p's < .02 \)), who do not significantly differ from each other (\( p = .165 \)).

**Discussion:** Cardiorespiratory measures are a feasible and useful objective measure of arousal in more severely affected youth with ASD who are unable to provide reliable self-reports about their reactions to environmental stressors. Such measures and identification of hyperarousal profiles could enhance future studies evaluating medication effects, temperament, emotion regulation, and emotion expression in this understudied and underrepresented segment of the autism population.

**References:**


1 Northeastern University  2 McMaster University