The impact of anxiety and attention-deficit/hyperactivity symptoms on the pupillary light reflex among children with ASD: Results from the ABC-CT Interim Analysis

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Background

- Pupillary light reflex (PLR) is an involuntary constriction response to a flash of light mediated by activity of the neuromodulators, acetylcholine and norepinephrine (NE), primarily projected from the locus-coeruleus (LC).
- Previous research indicates differences in the PLR in children with autism spectrum disorder (ASD) compared to typically-developing (TD) individuals (Daluwatte et al., 2013).[1]
- Limited research has explored the impact of common co-occurring symptomology, such as anxiety or difficulties with attention/hyperactivity on the PLR in children with ASD.
- These symptoms may affect the PLR, as LC-NE activity are implicated in both attentional control and anxiety (Redmond & Huang, 1979).[2]

Objective: To examine PLR among children with and without ASD in relation to anxiety and hyperactive symptomology.

Methods

ABC-CT Study Details:

- Methodologically rigorous, multi-site evaluation of potential biomarkers in a large sample of children with and without ASD.
- Longitudinal study evaluating children across 6 months, including clinical assessment, electroencephalogram (EEG), and eye-tracking.
- Current results from interim dataset of the larger ongoing study.

Inclusion/Exclusion Criteria:

- ASD Group: Age 6-11; met criteria for ASD based on ADOS-II, ADI-R, and DSM-5; IQ 60-150; stable medication for 8 weeks; children without sensory or motor impairments, epilepsy, and genetic or neurological conditions.
- TD Group: Age 6-11; IQ 80-150; stable medication for 8 weeks; no sibling with ASD; children without sensory or motor impairments, epilepsy, genetic or neurological conditions, or clinically significant scores on the *Child and Adolescent Symptom Inventory, 5th Edition* (CASI-5).

Participant Demographics:

	n (Female)	Age (*SD*)	IQ (*SD*)
TD	58 (21)	8.93 (1.72)	115.02 (13.96)
ASD	126 (23)	8.74 (1.64)	96.78(19.03)

**Diagnostic groups did not differ on age, [$t(182)=.726$, $p=.89$]. However, diagnostic groups significantly differed on full scale IQ, as measured by the *Differential Ability Scales, Second Edition* (DAS-II), [$t(182)=2.69$, $p<.01$] and sex [$t(182)=6.53$, $p=.01$].

Measures:

- *Autism Diagnostic Observation Schedule, 2nd Edition (ADOS-II)*
  - Clinician-administered, semi-structured, play-based task to assess ASD symptomology.
- *Child and Adolescent Symptom Inventory, 5th Edition* (CASI-5)
  - Parent-reported behavior rating scale for DSM-5 emotional and behavioral disorders in children.
  - Generalized Anxiety *T*-score and Hyperactivity *T*-score

Methods, cont.

Eye-tracking Acquisition:

- Binocular eye-tracking data were collected at 500 Hz using a SR Eyelink 1000 Plus.

Eye-tracking Experiment:

- Children were presented with 16 trials of 6-second stimuli (Figure 1) .

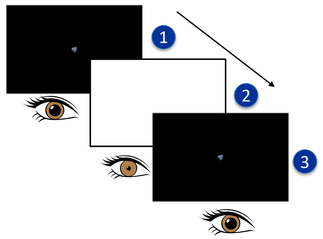

Figure 1 (left). Stimuli included a screen with (1) a central fixation point on a black background that (2) flashed white for 75 milliseconds. The flash onset occurred randomly between 1,600 and 2,400 milliseconds before (3) returning to black screen.

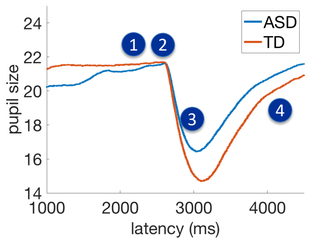

Figure 2 (right). The average PLR waveform comparing relative baseline of pupil size in each diagnostic group. The waveform reflects (1) onset of flash, (2) latency to constrict, (3) constriction velocity, and (4) redilation time.

Statistical Analyses:

- Relationships between ADOS calibrated severity scores (CSS), anxiety, and hyperactivity symptoms and relative pupil constriction (RPC) and constriction velocity in the PLR were analyzed using correlations and multiple regressions.

Results

- The ASD group showed higher scores than the TD group on both Anxiety [$t(181)=-9.8$, $p<.01$)] and Hyperactivity *T*-scores [$t(181)=-11.2$, $p<.01$)].
- CASI-5 Anxiety and Hyperactivity *T*-scores significantly predicted RPC when controlling for ADOS CSS (Figure 3; Tables 1 and 2).

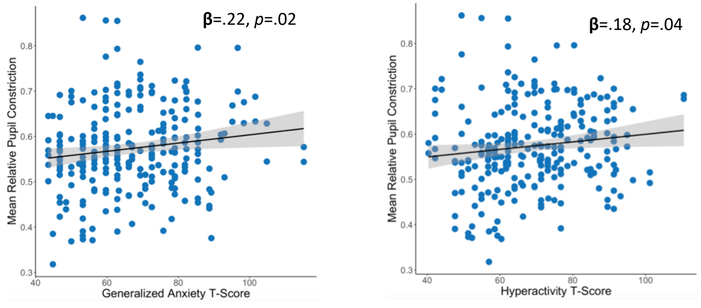

Figure 3. The relationship between anxiety and hyperactivity symptomology and relative pupil constriction when controlling for ADOS CSS n the ASD group.

Results, cont.

Table 1. Anxiety and relative pupil constriction in ASD group

	RPC			
Variables	**B**	**SE B**	**β**	**t**
ADOS CSS	.01	.004	.19*	2.19
Generalized Anxiety	.01	.01	.22*	2.42

Note. *$p<.05$ **$p<.01$

Table 2. Hyperactivity and relative pupil constriction in ASD group

	RPC			
Variables	**B**	**SE B**	**β**	**t**
ADOS CSS	.01	.004	.17	1.89
Hyperactivity	.01	.01	.18*	2.09

Note. *$p<.05$ **$p<.01$

- Higher Generalized Anxiety *T*-scores were correlated with mean constriction velocity in children with ASD who met symptom cutoff on CASI-5 (Figure 4).

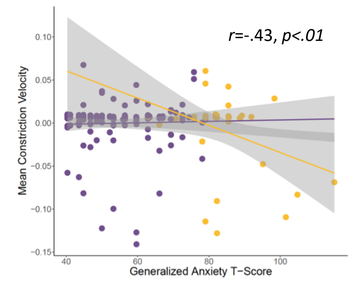

Figure 4 (left). The relationship between Generalized Anxiety *T*-scores and mean constriction velocity (pupil size over time) in the ASD group by symptom cutoff on the CASI-5.

Discussion

- Anxiety and hyperactive symptomology predicted unique variance in pupillary dynamics.
- Collinearity between anxiety and hyperactivity scores precluded dissociation between these constructs and their relation to the PLR.
- Neither relationship was found in the TD group, though elevated levels of anxiety and hyperactivity were exclusionary.
- These findings suggest PLR in individuals with ASD may index anxiety and hyperactivity and associated neural systems mediated by LC-NE activity.
- Future directions include examining the PLR across multiple time points and as a potential biomarker to target treatment for different clinical presentations among those with ASD.

References

1. Daluwatte, C., Miles, J. H., Christ, S. E., Beversdorf, D. Q., Takahashi, T. N., & Yao, G. (2013). Atypical pupillary light reflex and heart rate variability in children with autism spectrum disorder. *Journal of Autism and Developmental Disorders*, *43*(8), 1910-1925.
2. Redmond Jr, D. E., & Huang, Y. H. (1979). II. New evidence for a locus coeruleus-norepinephrine connection with anxiety. *Life Sciences*, *25*(26), 2149-2162.

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