Spectral analysis of Electroretinography to differentiate autism spectrum disorder and attention deficit hyperactivity disorder

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***Abstract*— autism spectrum disorder (ASD) and attention deficit hyperactivity disorder (ADHD) are both neurodevelopmental conditions that produce social interaction and executive functioning challenges but require very different therapeutic strategies. For that reason, early and accurate differentiation is important. However, their heterogeneity and overlap in symptoms make ASD and ADHD difficult to differentiate. The current diagnostic procedure to detect and distinguish ASD and ADHD is lengthy as it involves a comprehensive medical, developmental, and behavioral assessment. A more accessible and faster screening tool is needed to avoid delays in treatment. There is evidence that some retinal responses captured by the electroretinogram (ERG) are reduced in ASD subjects compared to neurotypicals whereas an opposite trend has been reported in ADHD, making ERG a promising tool for differentiating ASD and ADHD. However, previous ERG analyses based on amplitude and timing of ERG waves have exhibited limited success in differentiating ASD and ADHD. Recently, it has been found that time-varying spectral analysis of ERG allows for more accurate ASD detection compared to time-domain analysis. In this study, we evaluated the feasibility of differentiation of ASD and ADHD using features obtained by decomposing ERG using variable frequency complex demodulation (VFCDM). We used VFCDM features to train machine learning models and evaluated them using a subject independent validation approach. We achieved a maximum accuracy of 84% (87% sensitivity, 79% specificity), outperforming previous studies using ERG. Features from higher frequencies were found to be more important than features from lower frequencies.**

***Clinical Relevance*—This study establishes high frequency ERG information as a potential biomarker to differentiate ASD and ADHD.**

I. Introduction

Autism spectrum disorder (ASD) and attention deficit hyperactivity disorder (ADHD) are the most prevalent neurodevelopmental disorders, affecting approximately 1% and 5% of children worldwide, respectively [1], [2]. Detecting neurodevelopmental disorders at an early stage can ameliorate the progress and reduce the long-term effect by enhancing their cognitive and executive functioning skills [3]. Despite their distinct neurological conditions, ASD and ADHD may present as a co-occurring condition and for ADHD the diagnosis may be delayed owing to there not being an objective clinical test as there is for ASD [4] to formalize the diagnosis. Thus there is a need to provide a clinical measure that can identify the ASD and ADHD phenotype [5]. Given there is no specific biological or genetic test available that can definitively diagnose either condition novel biological signals derived from the electroretinogram (ERG) may offer new biomarkers to support the diagnosis and management of ASD and ADHD [6]. Previous studies have limited the ERG analysis to measures of amplitudes and time to peaks of the main features of the ERG [7], [8]. In this study, we used a high-resolution spectral decomposition technique named variable frequency complex demodulation (VFCDM) to extract highly sensitive time-frequency features and deployed machine learning (ML) algorithms to differentiate ASD and ADHD.

Impaired social and communication skills along with restrictive and repetitive patterns of behavior characterizes ASD [9], whereas impulsivity, hyperactivity and struggling to sustain attention on tasks are associated with ADHD [10]. Currently available diagnosis for these disorders involves a comprehensive assessment of an individual's developmental history, behavioral and cognitive performance in various settings which includes multiple screening by qualified healthcare professionals such as psychiatrist, developmental pediatrician with expertise in diagnosing neurological disorders. This makes the diagnosis lengthy and expensive obstructing access to early intervention and support.

To establish a reliable basis and objective diagnosis of ASD and ADHD previous studies have analyzed different biosignals such as electroencephalogram (EEG) [11], [12], heart rate variability (HRV) [13], and eye tracking [14]. Shephard et al. has reported reduced power in alpha and theta bands in ASD subjects whereas ADHD subjects exhibited decreased power in delta bands of the EEG [12]. Bellato et al. showed atypical autonomous response obtained from HRV as an indicator to separate ASD and ADHD with reduced parasympathetic activation in ASD during stress and reduced sympathetic functioning in ADHD during resting [13]. These studies based on EEG and HRV did not report any quantification of classification performance and were limited to statistical analysis. Duda et al. proposed a ML model based on feature from behavioral measures to classify ASD and ADHD with an area under the ROC curve (AUC) of 0.96 [15]. However, behavioral assessments are subjective and can be influenced by external factors which fail to provide an objective assessment.

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The retina contains a dense array of photosensitive cells providing a window to the brain and there is growing evidence suggesting that analyzing the underlying neurochemistry can help better understand brain disorders [16]. The ERG is a fast and non-invasive diagnostic test providing a superimposed electrical activity of retinal cells and their synaptic activity in response to light stimulation. Previous studies have identified atypical ERG responses in neurological disorders such as schizophrenia [17], bipolar disorder [18] and ASD [19]. Lee et al. has analyzed several time-domain indices of ERG reporting lower b-wave amplitude in ASD compared to neurotypicals and increased b-wave voltage in ADHD [8]. Based on this the authors were able to differentiate ASD and ADHD subjects with an AUC score of 0.88 (sensitivity: 84%, Specificity: 57%) although the ADHD sample size was small. Recently, time-varying spectral analysis of ERG have been used to identify more sensitive biomarkers of neurodevelopmental disorders [19], [20]. We hypothesize that detailed analysis of ERG using spectral characteristics and applying ML techniques will help improve the ASD vs ADHD classification performance.

II. Data Collection

A total of 46 ADHD (age mean ± SD, 13.2 ± 3.4) and 94 ASD (11 ± 4.5) individuals were recruited for this study at two different sites: London (UK) and Adelaide (Australia) where pediatric psychiatrists and psychologists performed diagnostic assessments. Participants met the diagnostic and statistical manual of mental disorder (DSM-IV or DSM-V) criteria after comprehensive evaluation supported by the Developmental, Dimensional and Diagnostic Interview (3Di) [21] and Autism Diagnostic Observation Schedule (ADOS) [4]. We did not include any subjects with any history of traumatic brain injury, inherited retinal or ocular disease, epileptic seizure and unable to follow verbal instructions. For participants under the age of 16 written consent from parents/legal guardian was collected. Ethical approval to collect data from human subjects was taken from Flinders University Human Research Ethics Committee and the Southeast Scotland Research Ethics Committee, UK. Customized light adapted (LA) full field ERG (LA-ERG) series was carried out by following the International Society for Clinical Electrophysiology of Vision (ISCEV) standards [22]. Based on our previous analysis we selected two flash strengths: 113 Trolands seconds (Td.s) and 446 Td.s. to evoke the LA-ERG responses starting from right eye and then left eye. Table 1 shows the total ERG samples for flash strength/ eye configurations. Flashes were presented on a 40 cd.m^-2 white background at 2 Hz and 30-60 responses were averaged to generate the final ERG waveform. ERG data was collected by placing self-adhesive electrodes 2-3 mm below the lower eyelid according to the manufacturer's recommendation. ERG waveforms were collected with a sampling frequency of 2000 Hz with electrode impedance < 5 kΩ. Data collection was conducted in seated condition and the subjects were asked not to blink during the procedure. ERG data was discarded from the study if either the a-wave amplitude was < 1μV or the electrode was placed > 4mm below the lower eyelid. We performed visual inspection of the data to remove highly corrupted samples which may affect the analysis. For further detailed information see [8], [20].

TABLE I. Total ERG Samples (ADHD/ASD)

	Right Eye	Left Eye
113 Td.s	112 / 191	107 / 172
446 Td.s	112 / 193	114 / 167

III. ERG Processing and Machine Learning

The ERG waveform is the summated response of the underlying neural generators. A typical LA-ERG waveform is shown in fig. 1 (Top) where the negative trough after the onset of the stimulus is caused by the hyperpolarization of cone photoreceptor cells and the positive b-wave is produced by depolarization of on and off bipolar cells. There are some high frequency wavelets seen in between the a-wave and the b-wave of the ERG waveform which are known as oscillatory potentials which are generated by the interaction between amacrine and bipolar cells of the retina [7], [22].

A. Feature Extraction and Statistical Analysis

There are four main time-domain parameters of the ERG waveform: a- wave peak time T_a , a-wave peak amplitude V_a, b-wave peak time T_b and b-wave peak amplitudeV_b . These parameters were extracted from local minima and maxima of a and b waves using RFF extractor software followed by visual inspection to confirm. Atypical change of these parameters indicates abnormalities in retinal function. The power spectral density of the ERG (Fig. 1 (bottom)) shows that most of the

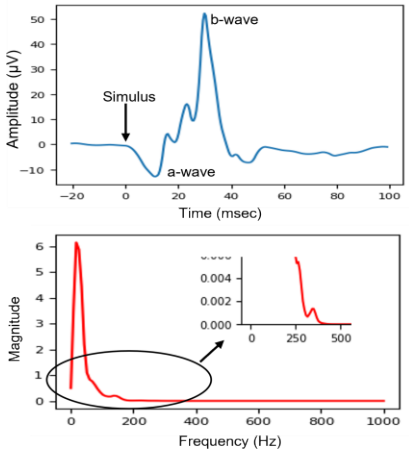

Figure 1: ERG signal (Top) and Power spectral density (Bottom)

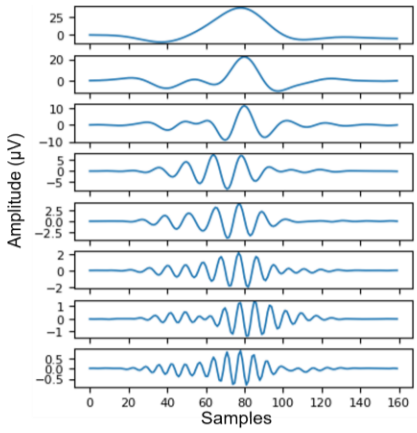

Figure 2: VFCDM decomposition of ERG

power is contained within 300 Hz which is in accordance with retinal physiology [22]. To extract informative features from different spectral regions we then decomposed the ERG signal into 24 equal width (1000/24=41.67 Hz) frequency bands using VFCDM [23]. However, since relevant information is found within 300 Hz, we utilized only the first 8 components in our analysis. Fig. 2 shows the first 8 VFCDM components extracted from a given ERG signal. Note that the first 20 and the last 50 points from the VFCDM components were removed to discard border effects. We extracted several statistical features from these components to analyze the dynamics of different frequency bands. Table 2 lists the time-domain features from raw ERG and statistical features computed from VFCDM components. Then we performed statistical analysis on these features to explore the features that provide more information for a more accurate separation between ASD and ADHD. We performed thresholding-based classification using these features and used Youden's J index to find optimum threshold.

TABLE II. FEATURES USED FOR MLMODELS

Time-domain features from ERG waveform	T_a, V_a, T_b, V_b
Statistical Features from VFCDM components	Mean, Max, Variance, Kurtosis, Skewness, Entropy, Inter-Quartile range

B. *Machine Learning Analysis*

After extracting features and statistical analysis we performed machine learning (ML) to classify ASD and ADHD. We applied different ML models such as random forest (RF), adaptive boosting (AdaBoost), gradient boosting (GradBoost), support vector machine (SVM), K-nearest neighbor (KNN), extreme gradient boosting (XGBoost) to find the best classifier. Since we had limited number of samples and multiple samples from the same subject, we performed 10-fold leave p groups out cross validation on the whole dataset which ensured that there was no data leakage. We extracted 60 features (4 time-domain, 56 statistical features from VFCDM components) in total and some of which may be redundant for classification. To discard redundant features, we used RF based feature selection process since RF has built-in feature selection method and is easily interpretable. After selecting features with higher separation strength, we performed hyperparameter optimization of the classifiers using 3-fold cross validation on the training dataset using GridSearchCV. Fig. 3 shows the workflow for the ML analysis.

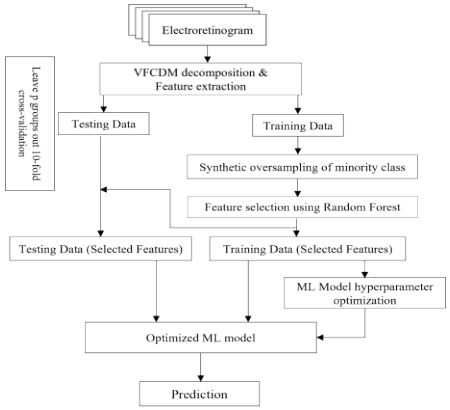

Figure 3: Machine learning analysis workflow

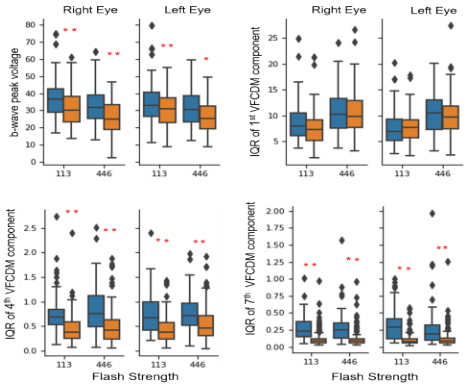

Figure 4: Statistical analysis of features

** and ** refer to statistically significantly different based on 0.01 and0.001 level of significance respectively*

IV. RESULTS

A. *Feature Analysis and thresholding*

One study identified the LA b-wave amplitude of ERG as the best time-domain feature to classify ASD and ADHD [8]. So, we analyzed features including V_b for comparison. We performed Shapiro-wilk normality test and found that most of the features are non-normal and that's why we performed Mann Whitney U test to determine whether they differed significantly between ASD and ADHD. Since comparing all the features from different frequency regions is beyond the scope of this report, and the inter-quartile range (IQR) was found to be typically the most important feature based on RF feature selection model, we chose IQR from 1st (0-41.67 Hz), 4th (125-166.6 Hz) and 7th (250-291.67 Hz) VFCDM component to compare with V_b as shown in Fig. 4. Table 3 shows the classification result using thresholding on these features and we report only (446/Right Eye) configuration. Here, sensitivity and specificity were used to assess the performance of the model to classify ASD and ADHD.

TABLE III. THRESHOLDING BASED CLASSIFICATION

Feature	Sensitivity (Sen.)	Specificity (Spe.)	Accuracy (Acc.)	AUC
V_b	0.42	0.86	0.58	0.69
IQR-1	0.72	0.38	0.59	0.52
IQR-4	0.83	0.56	0.73	0.74
IQR-7	0.81	0.75	0.79	0.82

A. *Classification Result*

Since we had ERG samples using 2 different flash strengths (113 Td.s and 446 Td.s) from both right and left eyes giving us 4 different combinations, we performed ML analysis for all the combinations separately. Out of all the ML models, the best classification performance was found using GradBoost and XGBoost. Table 4 reports classification performance of our proposed ML models.

TABLE IV. ML BASED CLASSIFICATION

Flash strength/ Eye	Classifier	Sen.	Spe.	Acc.	AUC
446 / Right	**GradBoost**	**0.87**	**0.79**	**0.84**	**0.87**
	XGBoost	0.86	0.78	0.83	0.90
446 / Left	GradBoost	0.78	0.60	0.70	0.77
	XGBoost	0.76	0.55	0.68	0.78

113 / Right	GradBoost	0.78	0.63	0.72	0.77
	XGBoost	0.83	0.60	0.75	0.79
113 / Left	GradBoost	0.85	0.64	0.76	0.80
	XGBoost	0.82	0.71	0.78	0.87

V. Discussion and Conclusion

In this study, we have presented a novel approach to classify ASD and ADHD. We have divided the overall spectral information into several equal width bands and showed that features computed from the high frequency bands of VFCDM decomposition are highly discriminative between ASD and ADHD subjects. We performed thresholding-based classification on both previously reported biomarker time domain parameter V_b [8] and statistical features from high frequency region. We found that IQR from the high frequency VFCDM component were more sensitive than low frequency components and able to separate ASD and ADHD subject with 79% accuracy compared to 58% accuracy with V_b . Finally, we performed machine learning based classification using all the features achieving 0.87 AUC score and 84% accuracy (Sen.= 0.87, Spe. = 0.79) which is a significant improvement over previously reported 0.88 AUC score (Sen.= 0.84, Spe.= 0.57) [8]since we achieved a balanced classification of both ASD and ADHD subjects where the previous study was highly biased towards one class. We have found that VFCDM decomposition of ERG provides suitable biomarkers to classify ASD and ADHD. However, ASD and ADHD are sometimes present in the same individual which has not been considered in this study. For our future studies we will investigate the potential of VFCDM decomposition and ML analysis to separate concomitant ASD and ADHD from ASD and ADHD alone. We found higher classification performance using 446/Right Eye configuration which might be the effect of collecting ERG data from right eye first. Further research is needed to explore the physiological significance of this finding and correlate with clinical severity. This work paves the way towards establishing a biomarker capable of detecting neurodevelopmental disorders to improve earlier diagnosis.

References

[1] "Zeidan, J., Fombonne, E., Scorah, J., Ibrahim, A., Durkin, M. S., Saxena, S., Yusuf, A., Shih, A., & Elsabbagh, M. (2022). Global prevalence of autism: A systematic review update. Autism Research, 15(5), 778-790. https://doi.org/10.1002/aur.2696".

[2] "Polanczyk G, de Lima MS, Horta BL, Biederman J, Rohde LA. The worldwide prevalence of ADHD: a systematic review and metaregression analysis. Am J Psychiatry. 2007 Jun;164(6):942-8. doi: 10.1176/ajp.2007.164.6.942. PMID: 17541055.".

[3] "Elder JH, Kreider CM, Brasher SN, Ansell M. Clinical impact of early diagnosis of autism on the prognosis and parent-child relationships. Psychol Res Behav Manag. 2017;10:283-292. Published 2017 Aug 24. doi:10.2147/PRBM.S117499".

[4] "Lord C., M., R., DiLavore P. C., Risi S., Gotham K., and S., B. (2012). Autism diagnostic observation schedule, second edition. Torrance, CA:: Western Psychological Services.".

[5] "Krakowski, A.D., Cost, K.T., Szatmari, P., Anagnostou, E., Crosbie, J., Schachar, R., et al. (2022). Characterizing the ASD-ADHD phenotype: measurement structure and invariance in a clinical sample. J Child Psychol Psychiatry. doi: 10.1111/jcpp.13609.".

[6] "Mahroo, O.A. Visual electrophysiology and 'the potential of the potentials'. Eye (2023). https://doi.org/10.1038/s41433-023-02491-2\".

[7] "Constable, P.A., Ritvo, E.R., Ritvo, A.R. et al. Light-Adapted Electroretinogram Differences in Autism Spectrum Disorder. J Autism Dev Disord 50, 2874–2885 (2020). https://doi.org/10.1007/s10803-020-04396-5".

[8] "Lee, I. O., D. H. Skuse, P. A. Constable, F. Marmolejo-Ramos, L. R. Olsen and D. A. Thompson (2022). 'The electroretinogram b-wave amplitude: a differential physiological measure for Attention Deficit Hyperactivity Disorder and Autism Spectrum Disorder.' J Neurodev Disord 14(1): 30.".

[9] "Hodges H, Fealko C, Soares N. Autism spectrum disorder: definition, epidemiology, causes, and clinical evaluation. Transl Pediatr. 2020;9(Suppl 1):S55-S65. doi:10.21037/tp.2019.09.09".

[10] "Singh A, Yeh CJ, Verma N, Das AK. Overview of Attention Deficit Hyperactivity Disorder in Young Children. Health Psychol Res. 2015 Apr 13;3(2):2115. doi: 10.4081/hpr.2015.2115. PMID: 26973960; PMCID: PMC4768532.".

[11] "Bosl, W., Tierney, A., Tager-Flusberg, H. et al. EEG complexity as a biomarker for autism spectrum disorder risk. BMC Med 9, 18 (2011). https://doi.org/10.1186/1741-7015-9-18".

[12] "Shephard, E., Tye, C., Ashwood, K.L. et al. Resting-State Neurophysiological Activity Patterns in Young People with ASD, ADHD, and ASD + ADHD. J Autism Dev Disord 48, 110–122 (2018). https://doi.org/10.1007/s10803-017-3300-4".

[13] "Bellato A, Arora I, Kochhar P, Ropar D, Hollis C, Groom MJ. Heart Rate Variability in Children and Adolescents with Autism, ADHD and Co-occurring Autism and ADHD, During Passive and Active Experimental Conditions. J Autism Dev Disord. 2022 Nov;52(11):4679-4691. doi: 10.1007/s10803-021-05244-w. Epub 2021 Oct 30. PMID: 34716841; PMCID: PMC9556357.".

[14] "Muszkat M, de Mello CB, Muñoz Pde O, Lucci TK, David VF, Siqueira Jde O, Otta E. Face Scanning in Autism Spectrum Disorder and Attention Deficit/Hyperactivity Disorder: Human Versus Dog Face Scanning. Front Psychiatry. 2015 Oct 23;6:150. doi: 10.3389/fpsyt.2015.00150. PMID: 26557097; PMCID: PMC4615933.".

[15] "Duda, M et al. 'Use of machine learning for behavioral distinction of autism and ADHD.' Translational psychiatry vol. 6,2 e732. 9 Feb. 2016, doi:10.1038/tp.2015.221".

[16] "Yap, Timothy E et al. 'Retinal correlates of neurological disorders.' Therapeutic advances in chronic disease vol. 10 2040622319882205. 2 Dec. 2019, doi:10.1177/2040622319882205".

[17] "Hébert M, Mérette C, Gagné AM, Paccalet T, Moreau I, Lavoie J, Maziade M. The Electroretinogram May Differentiate Schizophrenia From Bipolar Disorder. Biol Psychiatry. 2020 Feb 1;87(3):263-270. doi: 10.1016/j.biopsych.2019.06.014. Epub 2019 Jun 27. PMID: 31443935.".

[18] "Gross G, Tursini K, Albuisson E, Angioi-Duprez K, Conart JB, Louis Dorr V, Schwan R, Schwitzer T. Bipolar disorders and retinal electrophysiological markers (BiMAR): Study protocol for a comparison of electroretinogram measurements between subjects with bipolar disorder and a healthy control group. Front Psychiatry. 2022 Sep 8;13:960512. doi: 10.3389/fpsyt.2022.960512. PMID: 36159928; PMCID: PMC9492949.".

[19] "S. M. Manjur et al., 'Detecting Autism Spectrum Disorder Using Spectral Analysis of Electroretinogram and Machine Learning: Preliminary results,' 2022 44th Annual International Conference of the IEEE Engineering in Medicine & Biology Society (EMBC), 2022, pp. 3435-3438, doi: 10.1109/EMBC48229.2022.9871173.".

[20] "Constable PA, Marmolejo-Ramos F, Gauthier M, Lee IO, Skuse DH, Thompson DA. Discrete Wavelet Transform Analysis of the Electroretinogram in Autism Spectrum Disorder and Attention Deficit Hyperactivity Disorder. Front Neurosci. 2022 Jun 6;16:890461. doi: 10.3389/fnins.2022.890461. PMID: 35733935; PMCID: PMC9207322.".

[21] "Skuse D, Warrington R, Bishop D, Chowdhury U, Lau J, Mandy W, Place M. The developmental, dimensional and diagnostic interview (3di): a novel computerized assessment for autism spectrum disorders. J Am Acad Child Adolesc Psychiatry. 2004 May;43(5):548-58. doi: 10.1097/00004583-200405000-00008. PMID: 15100561.".

[22] "Robson AG, Frishman LJ, Grigg J, Hamilton R, Jeffrey BG, Kondo M, Li S, McCulloch DL. ISCEV Standard for full-field clinical electroretinography (2022 update). Doc Ophthalmol. 2022 Jun;144(3):165-177. doi: 10.1007/s10633-022-09872-0. Epub 2022 May 5. PMID: 35511377; PMCID: PMC9192408.".

[23] "Wang H, Siu K, Ju K, Chon KH. A high resolution approach to estimating time-frequency spectra and their amplitudes. Ann Biomed Eng. 2006 Feb;34(2):326-38. doi: 10.1007/s10439-005-9035-y. Epub 2006 Feb 7. PMID: 16463086.".