SUMMARY

Recent advances in neuroscience have allowed researchers to introduce functional neuromarkers of autism spectrum disorders (ASD) into clinical practice. Most promising are studies of brain work in milliseconds, such as quantitative EEGs, event-related potentials (ERPs), low resolution electromagnetic tomography (sLORETA), especially when it is possible to use Human Brain Index (HBI) methodology to compare test results obtained by a child with ASD with the normative database. The use of HBI methodology makes possible to determine functional neuromarkers of ASD and comorbidities, such as epilepsy, impaired cognitive and emotional control or executive function (FF) disorders. Such procedure can contribute to more effective diagnosis, as well as provide personalized care for patients with ASD. In this case study we use the HBI methodology for more effective diagnosis of the child with ASD (level 2) with focal epilepsy and EF dysfunction in hope that we can provide more effective therapies which might improve her quality of life.

We present the case of a 11-years-old girl with an initial diagnosis of ASD with focal epilepsy and EF dysfunction. The patient was sent to the Teaching-Reintegration Centre in Kraków with remarks that she simply could not function in everyday life. A neuropsychological examination using standardized batteries to assess the girl's cognitive, emotional and social functioning confirmed the presence of ASD (level 2), and co-occurring executive function disorders (EF). She was also analyzed from the clinical EEG perspective as well as from the QEEG/ERPs perspective and current coexistence of 3 Hz Spike-Wave Discharges on EEG and focal epilepsy was found. In the examination several episodes of 3 Hz paroxysms were found in the ventro-lateral prefrontal cortex (BA 11) in 40 min EEG recording in Eyes open, Eyes closed as well as in task conditions. The patient's ERPs showed that to the first stimulus (continue cue) temporal P2 wave with larger duration and longer latency was elicited in comparison to the average of the norm for her age from the Human Brain Index (HBI) normative database at Chur, Switzerland. The P30 cue was smaller in the patient than in the healthy controls from HBI and statistically significant deviations from the average were found. Also in response to NOGO stimulus no positive parietal-central P3 was observed, which appears in the ERPs of healthy controls from HBI. The ERPs differences in the VCPT show statistically significant deviations in the patient's brain function when compared to the healthy controls from HBI. It should be stressed that the number of trials for the first stimulus presentation and NOGO condition were large enough for computing reliable ERPs, as it was indicated by Kropotov (2009; 2016).

HBI methodology was helpful in finding functional neuromarkers of co-existence of 3 Hz Spike-Wave Discharges on EEG with focal epilepsy and EF disorder in girl with ASD (level 2). Therefore, it was possible to offer more effective rehabilitation of the disorders, which will contribute to the integration of the self system and better quality of her life.

Key words: epilepsy, executive dysfunctions, qEEG, ERPs, sLoreta
INTRODUCTION

Autism spectrum disorders (ASD) is categorized in the Diagnostic and Statistical Manual of Mental Disorders (DSM-5) as a neurodevelopmental disorder (a subcategory of mental disorders). The new diagnostic criteria of ASD focuses on three criterion: (a) social communication impairment, (b) fixated interests/repetitive behaviors and (c) symptoms existing in early childhood (Sharma, Gonda, Tarazi 2018). Current Classification of Autism Spectrum Disorder devide these neurodevelopmental disorder due to the need for support for:

• ASD Level 1 – the lowest-range of the spectrum, which requires a little support,
• ASD Level 2 – the mid-range of the spectrum, which requires significant support,
• ASD Level 3 – the most severe end of the spectrum, which requires very substantial support (Pąchalska, Kaczmarek, Kropotov 2014).

Since 2012, 99 estimates from 71 studies were published indicating a global autism prevalence that ranges within and across regions, with a median prevalence of 100/10,000 (range: 1.09/10,000 to 436.0/10,000). The median male-to-female ratio was 4.2. The median percentage of autism cases with co-occurring intellectual disability was 33.0%. Estimates varied, likely reflecting complex and dynamic interactions between patterns of community awareness, service capacity, help seeking, and sociodemographic factors. Recent estimation showed that approximately 1/100 children are diagnosed with ASD around the world. Prevalence estimates increased over time and varied greatly within and across sociodemographic groups. These findings reflect changes in the definition of ASD and differences in the methodology and contexts of prevalence studies. (Zeidan, Fombonne, Scorah et al. 2022).

Hereditary factors, parental history of psychiatric disorders, pre-term births, and fetal exposure to psychotropic drugs or insecticides have all been linked to higher risk of ASD. Several scales such as the Childhood Autism Rating Scale (CARS), The Autism Spectrum Disorder-Observation for Children (ASD-OC), The Developmental, Dimensional, and Diagnostic Interview (3di), are available to aid in better assessing the behaviors and symptoms associated with ASD (Pąchalska, Kaczmarek, Kropotov 2014).

Nearly 75% of ASD patients suffer from comorbid psychiatric illnesses or conditions, which may include epilepsy, attention-deficit hyperactivity disorder (ADHD), anxiety, bipolar disorder, depression, Tourette syndrome, and other overlapping clinical manifestations, including developmental delays, cognitive problems as well as challenging behaviours such as difficulty sleeping, self-injury (Woolfenden, Sarkozy, Ridley et al. 2009; Khetrapal 2010; Russ, Larson, Halfon 2012; Close, Lee, Kaufmann et al. 2012) and executive function disorders (Pąchalska, Kaczmarek, Kropotov 2014).

Co-occurrence of epilepsy and ASD

Epilepsy is a group of non-communicable neurological disorders characterized by recurrent epileptic seizures which can vary from brief and nearly unde-
tectable periods to long periods of vigorous shaking due to abnormal electrical activity in the brain (Hammer & McPhee 2010; Kropotov 2016). Epilepsy is known to occur in a higher than expected proportion of individuals with autism spectrum disorders (ASDs) (Ewen, Marvin, Law et al. 2019). Some reports, suggesting that the two conditions share underlying biology. For example, both conditions are characterized by overly excitable brains (Danielsson, Gillberg, Billstedt et al. 2005; Hara 2007; Spence, Schneider 2009; Woolfenden, Sarkozy, Ridley et al. 2009; Gillberg, Billstedt, Sundh et al. 2010; Mouridsen, Rich, Isager 2011). The co-occurrence of epilepsy and ASD is associated with increasing child age, female gender, intellectual disability, speech problems and lower socioeconomic status (Thomas, Hovinga, Rai et al. 2017). As the extensive and complex web of co-occurrence between epilepsy and ASD is revealed, there is growing interest in the extent to which the two disorders are linked (whether epilepsy contributes to autism or is a consequence of the condition) (Pąchalska, Kaczmarek, Kropotov 2014).

The literature also points out that there is abnormal epileptiform activity in the EEG recordings of children with ASD. This means that these children are at high risk of developing epilepsy (Ekinci, Arman, Isik et al. 2007). Similarly, mounting evidence has indicated that children newly diagnosed with various epilepsies often exhibit comorbid ASD-like behavioral and neuropsychiatric problems (Kanner 2010), and that early-onset seizures (before 2 years) may predict that infants are at high risk of developing ASD (Tuchman, Moshe, Rapin 2009; Saemundsen, Ludvigsson, Hilmarsdottir et al. 2007).

Co-occurrence of executive functions disorder and ASD

It is well known that children with ASD also manifest impairments in executive functions (EF), especially the planning and control of goal-oriented action (Russo, Flanagan, Iarocci, et al. 2009; Robinson, Goddard, Dritschel et al. 2009; Lynch, Breeden, You et al. 2017). EF impairment co-occurring with other ASD-specific symptoms is observed in 41% to 78% of cases (Murray 2010), increases with age (Sikora et al. 2012; Rosenthal et al. 2013), and persists despite amelioration of ASD symptoms (Troyb et al. 2013) through the treatment process (Pąchalska, Kaczmarek, Kropotov 2014). EF disorders in ASD involves the component processes of these functions (e.g. monitoring of action, initiation, inhibition and mode change of action, working memory and planning/organization (Granader, Wallace, Hardy et al. 2014). These disorders moderate ASD symptoms (Yerys, Wallace, Sokoloff et al. 2009), cause poorer adaptive functioning in children with ASD (Szatmari, Bartolucci, Bremner et al. 1989; Pugliese, Anthony, Strang et al. 2015), and reduce their quality of life (Sikora et al. 2012).

Research is being conducted on explaining the neural mechanisms of various deficits in ASD, including EF disorders. Jeste & Nelson (2009) published a critical review of the literature on the use of event-related potentials (ERPs), in the process of explaining the neural mechanisms of primary auditory and visual deficits and EF disorders in children with ASD. The authors conclude that these
children are likely to have disorders at both low and higher levels of auditory and visual processing, with marked impairments in the processing of social stimuli. They also present putative neuronal circuits underlying all these deficits. Subsequent studies devoted to the neural mechanisms of common EF disorders in children with ASD have shown that they may result from dysfunction of the control network of fronto-parietal neurons to interact extensively with other networks or to act as a convergence zone when necessary to adapt to new behavioral demands (van den Heuvel, Sporns 2013). Therefore, adaptive integration of information from the whole brain is hindered or impossible, resulting in EF disorders (Kropotov 2016).

Co-occurrence of focal epilepsy, executive functions disorders and ASD

In our previous work, we found that co-occurrence of focal epilepsy or seizure activity recorded in EEG, and EF disorders in children with ASD is high and ranging from 38 to 82%. Recurrent epileptic seizures might affect the depth of EF impairment found in a children with ASD (Pąchalska, Kaczmarek & Kropotov 2014; Kropotov 2016).

Therefore, research is needed to understand the mechanisms underlying such a configuration of disorders. The most promising are studies of brain work in milliseconds, such as quantitative EEGs, event-related potentials (ERPs) and sLoreta tomography, especially when it is possible to use Human Brain Index (HBI) methodology to compare test results obtained by a child with ASD with the normative database (Kropotov 2009; 2016). The use of HBI methodology make possible to determine functional neuromarkers of ASD and comorbidities, such as epilepsy, impaired cognitive and emotional control or executive function (FF) disorders (Pąchalska, Kaczmarek & Kropotov 2014). Such procedure can contribute to more effective diagnosis, as well as provide personalized care for patients with ASD. In this case study we use the HBI methodology for more effective diagnosis of the child with ASD (level 2) with focal epilepsy and EF dysfunction in hope that we can provide more effective therapies which might improve her quality of life.

CASE STUDY

We present the case of a 11-years-old girl with an initial diagnosis of ASD with focal epilepsy and EF dysfunction. The patient was sent to the Teaching-Reintegration Centre in Kraków with remarks that she simply could not function in everyday life. A neuropsychological examination using standardized batteries to assess the girl's cognitive, emotional and social functioning confirmed the presence of ASD (level 2), and co-occurring executive function disorders (EF). She was also analyzed from the clinical EEG perspective as well as from the QEEG/ERPs perspective and current coexistence of 3 Hz Spike-Wave Discharges on EEG and focal epilepsy was found.
Neuropsychological testing

The Clinical Test of Executive Functions (CTEF) (Pąchalska 2008) was conducted to assess EF disorders in our patient. This test assesses the ability to perform basic tasks of daily life activities in the following six subtests:

- Finding milk in the refrigerator
- Pouring a ¼ cup of drink from a bottle
- Buying a favorite drink
- Writing the sentence "Mummy is cooking dinner"
- Finding a hidden favorite toy
- Playing a game on a smartphone

The performance of the test is evaluated by 3 competent judges: a neuropsychologist, a neurologist and the child's parent. A maximum of 10 points can be scored for the correct independent completion of the task, and a maximum of 5 points for completion with the assistance of the examiner.

The results obtained by the patient under study show a significantly lower ability to perform all basic activities of daily living assessed in the CTEF than the average results obtained in a control group (100 children with ASD level 2, without epilepsy). The differences in all subtests are statistically significant at p=0.001. These results indicate that the patient has a significant EF impairment.

Neurophysiological testing

Procedures of EEG recording and analysis

EEG was recorded by means of the Mitsar (Mitsar, Ltd.) amplifier from 19 electrodes (Fp1, Fp2, F7, F3, Fz, F4, F8, T3, C3, Cz, C4, T4, T5, P3, Pz, P4, T6, O1, O2 sites in the International 10-20 system) with a 250 Hz sampling rate in the 0.3 – 70 Hz frequency range in the following conditions: 1) Eyes opened (EO) – at least 3 minutes, 2) Eyes closed (EC) – at least 3 minutes, and 3) a modification of GO/NOO task (20 minutes). The task consisted of 400 trials presenting
to a subject every 3 seconds (Fig.2). In the task we selected four categories of stimuli: 1) 20 different images of animals — referred to later as A, 2) 20 different images of plants — P, 3) 20 different images of humans presented together with an artificial sound — HS. Each sound was composed of four pure tones of 500, 800, 1100, 1400 Hz of 20 ms duration. Trials consisted of the presentation of a pair of stimuli with interstimulus interval of 1.1 sec. Four categories of trials were selected: A-A, A-P, P-P, and P-HS. The trials were grouped into four sessions with one hundred trials each. In each session a unique set of five A stimuli, five P and five HS stimuli was selected. Each session consisted of a pseudorandom presentation of 100 pairs of stimuli with an equal probability for each category and each stimulus.

Absolute and relative magnitude spectra and coherences in all conditions computed and compared with the parameters of a corresponding age group from the Human Brain Index (HBI) reference database.

The analysis consists of the following steps:

1. **Eye movement artifact correction** and elimination: a) using a spatial filtration technique based on zeroing the activation curves of individual Independent
Component Analysis (ICA) components corresponding to horizontal and vertical eye movements, as well as b) excluding epochs with excessive amplitude of EEG and excessive faster and slower frequency activity;
2. Fast-Fourier Transformation (FFT) of the corrected EEG for extracting EEG power and coherence for all 0.25 Hz bins in the frequency band from 0.5 to 30 Hz;
3. Computation of event related potentials by averaging EEG over trials for each category of trial and each channel with time resolution of 4 ms;
4. Decomposition of an individual ERPs into independent components by applying spatial filters extracted by means of the ICA from the collection of ERPs computed for the corresponding group of healthy subjects;
5. Comparison of each extracted electrophysiological and behavioral variable against the corresponding variable computed for a carefully constructed and statistically controlled age-regressed, normative database in which the variables have been transformed and confirmed for their Gaussian distribution.

Automated spike detection

Visual inspection of raw EEG was made in order to search for paroxysmal patterns that pop out of the background EEG. Besides the visual inspection, an automated spike detection was performed. The method of automated spike detection is based on temporal parameters of spikes as well on spatial location of the corresponding spike dipole (Ktonas 1987). The amplitude-temporal parameters are defined on the basis of comparison spike detection by the program and by experienced experts on the data base of more than 300 EEG recordings in epileptic patients. There are three characteristics that define a spike or a sharp wave in EEG. They are paroxysmal character, high degree of sharpness and short duration. These parameters are presented in Fig 3.

Fig. 3. The relative residual energy for dipole approximation of the detected spike is chosen less than 0.2.
Source: Pąchalska, Kaczmarek, Kropotov 2014.
RESULTS

Raw EEG
In the examination several episodes of 3 Hz paroxysms were found in 40 min EEG recoding in Eyes open, Eyes closed as well as in task conditions. Fig. 4 depicts one of these episodes. The EEG fragment is shown after eye blinks and lateral eye movement artifacts were excluded. The current density generators of the 3 Hz paroxysm are found in the ventro-lateral prefrontal cortex (BA 11.)

EEG spectra in VCPT
The difference “patient-norm” for the relative EEG spectra at Cz are presented in Fig.5. The maps of the difference at alpha and beta frequency bands are presented at the bottom. One can see decrease of the relative EEG spectra in the alpha band. The decrease is widely distributed over posterior central electrodes. In contrast, the increase of beta activity is quite local and is present only at Cz.

Behavioral data in VCPT
Table 1 shows omission, commission errors, reaction time and error in the variance of response for the patient in comparison to the average data of 63 healthy subjects of the same age. It is observed that the patient had strong difficulties in performing the task correctly.

ERPs
The number of trials for the first stimulus presentation and NOGO condition were large enough for computing reliable ERPs. The patient’s ERP are presented in Fig. 6 in comparison with the ERPs of healthy controls for her age from the Human

Fig. 4. Depiction of 3 Hz paroxysmal activity in the patient. A. Raw EEG fragment in Eyes open condition. B. Map of EEG spectra of the 3Hz activity fragment. C.and D. sLORETA images of the paroxysmal activity
Brain Index (HBI) normative database at Chur, Switzerland. One can see that in response to the first stimulus (continue cue) a temporal P2 waves with larger durations and longer latencies in comparison to norms from HBI is elicited (Fig. 6 A). The P30 cue is smaller in the subject than in norms from HBI (Fig 6 A). In response to the NOGO stimulus no positive parietal-central P3 is observed (Fig 6 B).

Fig. 5. The difference of the relative EEG spectra in the VCPT the patient minus the grand average of EEG spectra in the group of healthy subjects. Top – statistically significant differences are marked by vertical bars at the bottom of the EEG spectra difference (small bar – p<0.05, medium bar – p<0.01 and large bar – p<0.001). Maps at the 10 and 16 Hz frequencies for the difference spectra

Table 1. Behavioral data in the VCPT

<table>
<thead>
<tr>
<th></th>
<th>Omission</th>
<th>Commission</th>
<th>RT1</th>
<th>var(RT1)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patient</td>
<td>0.00</td>
<td>0.00</td>
<td>0.11</td>
<td>0.00</td>
</tr>
<tr>
<td>Norms</td>
<td>80%</td>
<td>13%</td>
<td>558ms</td>
<td>49.8ms</td>
</tr>
<tr>
<td></td>
<td>2.5%</td>
<td>0.7%</td>
<td>420ms</td>
<td>9.8ms</td>
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Brain Index (HBI) normative database at Chur, Switzerland. One can see that in response to the first stimulus (continue cue) a temporal P2 waves with larger durations and longer latencies in comparison to norms from HBI is elicited (Fig. 6 A). The P30 cue is smaller in the subject than in norms from HBI (Fig 6 A). In response to the NOGO stimulus no positive parietal-central P3 is observed (Fig 6 B).

Fig. 6. The ERP differences in the VCPT. A: ERPs to the first “continue” stimulus – an image of an animal in the pair. Green- patient. Red – the grand average for the healthy subjects. Blue – the difference wave. Statistically significant deviations from the average are marked by blue vertical bars on the bottom. B: ERPs to the second NOGO stimulus. Green–patient. Red– the grand average for the healthy subjects. Blue – the difference wave. Statistically significant deviations from the average are marked by blue vertical bars on the bottom.
The ERP differences in the VCPT show statistically significant deviations in the patient’s brain function compared to the average for her age from the Human Brain Index (HBI) normative database at Chur, Switzerland.

**DISCUSSION**

Relatively recent studies have addressed the topic of co-existence of Rolandic and 3 Hz Spike-Wave Discharges on EEG in Children with focal epilepsy (Saha, Proix, Cash et al. 2019; Datta, Wallbank, Mak et al. 2019). Such co-occurrence of disorders has also been noted in children with ASD (Russ, Larson, Halfon 2012; Close, Lee, Kaufmann et al. 2012; Pąchalska, Kaczmarek, Kropotov 2014; Pąchalska, Kaczmarek, Bednarek 2020).

This research revealed that in our patient with an initial diagnosis of ASD (level 2) co-existence of 3 Hz Spike-Wave Discharges on EEG with focal epilepsy is overlapping with EF disorders. From the clinical EEG perspective as well as from the QEEG/ERP perspective several episodes of 3 Hz paroxysms were found in the ventro-lateral prefrontal cortex (BA 11) in 40 min EEG recoding in Eyes open, Eyes closed as well as in the task conditions. The patient’s ERPs showed that for the first stimulus (continue cue) temporal P2 wave with larger duration and longer latency was elicited in comparison to the average of the norm for her age from the Human Brain Index (HBI) normative database at Chur, Switzerland. The P30 cue was smaller in the patient than in the healthy controls from normative database HBI and statistically significant deviations from the average were found. Also in response to NOGO stimulus no positive parietal-central P3 was observed, which appears in the ERPs of healthy controls from HBI. The ERPs differences in the VCPT show statistically significant deviations in the patient’s brain function when compared to the healthy controls from normative database HBI. It should be stressed that the number of trials for the first stimulus presentation and NOGO condition were large enough for computing reliable ERPs, as it was indicated by Kropotov (2009; 2016).

As we wrote before a child with ASD with focal epilepsy and EF disorder has a disturbed self system, mainly because he does not make appropriate contacts and does not form strong links with society (Pąchalska, Kaczmarek, Kropotov 2014; Pąchalska, Kaczmarek, Bednarek 2020). As a result, the minimal (working) self and therefore the longitudinal (comprehensive, autobiographical) self is not developed or even disintegrates (Fig. 7). These disorders do not support the development of the child with ASD and require rehabilitation.

To sum up, studies of brain work in milliseconds, such as quantitative electroencephalography (qEEG) and event-related potentials (ERPs) are promising for diagnosis of ASD (Pąchalska, Kaczmarek & Kropotov 2014; Pąchalska, Kaczmarek & Bednarek 2020). The simplicity of the task during qEEG recording, the automatic spike detection, the diagnostic power of ERPs enhanced by the recent emergence of new methods of analysis, such as independent component analysis (ICA) and low resolution electromagnetic tomography (sLORETA) as well as
the ability to compare results with the Human Brain Index (HBI) database at Chur, Switzerland maximizes the accuracy of the diagnosis of the disorder while inducing a goal-oriented state requiring subjects to monitor and ignore relevant and irrelevant stimuli, respectively (Kropotov 2009, 2016).

It should be pointed out that the use of HBI methodology to compare test results obtained by a child with ASD with the normative database make possible to determine functional neuromarkers of ASD and comorbidities, such as epilepsy, impaired cognitive and emotional control or executive function (FF) disorders (Pąchalska, Kaczmarek, Kropotov 2014). Such procedure can contribute to more effective diagnosis, as well as provide personalized care for patients with ASD, and will lead to more effective integration of the child’s self system and therefore improve the quality of their life.

It should be add that a more cognitively demanding EF task would likely produce the observed pattern of results, perhaps stronger rather than qualitatively different. This prediction can be tested in future work.

CONCLUSIONS

HBI methodology was helpful in finding functional neuromarkers of co-existence of 3 Hz Spike-Wave Discharges on EEG with focal epilepsy and EF disorder in girl with ASD (level 2). Therefore, it was possible to offer more effective rehabilitation of the disorders, which will contribute to the integration of the self system and better quality of her life.
Acknowledgments

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