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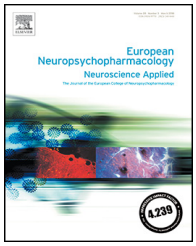

Electroencephalographic biomarkers as predictors of methylphenidate response in attention-deficit/hyperactivity disorder

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Abstract
EEG biomarkers have shown promise in predicting non-response to stimulant medication in ADHD and could serve as translational biomarkers. This study aimed to replicate and extend previous EEG biomarkers. The international Study to Predict Optimized Treatment for ADHD (iSPOT-A), a multi-center, international, prospective open-label trial, enrolled 336 children and adolescents with ADHD (11.9 yrs; 245 males; prescribed methylphenidate) and 158 healthy children. Treatment response was established after six weeks using the clinician rated ADHD-Rating Scale-IV. Theta/Beta ratio (TBR) and alpha peak frequency (APF) were assessed at baseline as predictors for treatment outcome. No differences between ADHD and controls were found for

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TBR and APF. 62% of the ADHD group was classified as a responder. Responders did not differ from non-responders in age, medication dosage, and baseline severity of ADHD symptoms. Male-adolescent non-responders exhibited a low frontal APF (Fz: R = 9.2 Hz vs. NR = 8.1 Hz; ES = 0.83), whereas no effects were found for TBR. A low APF in male adolescents was associated with non-response to methylphenidate, replicating earlier work. Our data suggest that the typical maturational EEG changes observed in ADHD responders and controls are absent in non-responders to methylphenidate and these typical changes start emerging in adolescence.
Clinical trials registration: www.clinicaltrials.gov; NCT00863499 (https://clinicaltrials.gov/ct2/show/NCT00863499).

1. Introduction

Many studies have compared resting state brain activity, especially electro-encephalography (EEG), of children with ADHD with that of typically developing children. Ever since the first description of deviant fronto-central slow-wave EEG activity (…*at frequencies of 5-6/s*…'), later so called 'theta activity' (Walter and Dovey, 1944), in 'behavioral problem children' (Jasper et al., 1938; p. 644), excess theta EEG power is an often reported finding in patients with ADHD (see: Arns et al., (2013) for review). Others have proposed the ratio of theta and beta, in short the Theta/Beta Ratio (TBR), to be a better differentiator of children with ADHD and healthy controls (Monastra et al., 2001). However, a recent meta-analysis could not confirm this measure to be a reliable diagnostic metric in ADHD (Arns et al., 2013), see (Arns et al., 2016b) for further discussion.

Another usage of EEG activity is its ability to predict treatment response, or a more prognostic rather then a pure 'diagnostic' usage (Arns et al., 2013; Arns and Gordon, 2014). Previous studies have demonstrated that an excess of slow (theta) activity and an elevated TBR were most consistently associated to a favorable treatment response to stimulant medication (Arns et al., 2008; Clarke et al., 2002b; Ogrim et al., 2014; Satterfield et al., 1971; Suffin and Emory, 1995) and EEG-neurofeedback (Arns et al., 2012a; Gevensleben et al., 2009; Monastra et al., 2002). Conceptually this can be understood as representative of a hypoarousal subgroup (with excess theta as a signature of drowsiness), hence psychostimulant medication to be most effective for this subgroup by its psychostimulant nature (Arns and Kenemans, 2014; Clarke et al., 2002a). Another EEG metric that has shown promise in predicting treatment outcome is the alpha peak frequency (APF), i.e. the individual frequency at which alpha activity oscillates. This low APF was previously found a biomarker associated with non-response to stimulant medication in male ADHD patients (Arns et al., 2008), but also to antidepressant treatments (Arns et al., 2012b; Arns et al., 2010; Ulrich et al., 1984) suggesting this could be considered a more generic biomarker for non-response and could serve as a translational biomarker to investigate the exact underlying etiology and potentially develop new treatments for such subgroups.

Resting-state EEG studies to date often consisted of small sample sizes with a large diversity in demographics and employed a large variety of methods such as different resting-state conditions (eyes-open [EO] or eyes-closed [EC]) etc. Therefore, studies are needed that prospectively test these differences under standardized conditions with appropriate sample size and the use of a multi-site approach to obtain more generalizable results. To this end, the aims of the current study were twofold. First, to investigate ADHD specific differences in brain function compared to typically developing children. Second, to investigate predictors of treatment response to methylphenidate (MPH) using EEG data from the multisite International Study to Predict Optimized Treatment for ADHD (iSPOT-A), collected from 158 healthy children and 336 children and adolescents with ADHD. This is the first and largest multisite study to investigate EEG treatment predictors to MPH using a standardized methodology. Its sample-size and multisite design ensure accurate and generalizable results that allow for investigating interactions with gender and age-group (children vs. adolescents).

Based on the previous literature we hypothesized that there would be no difference between ADHD patients and controls for the TBR and APF on the group level, but there would be main effects of age-group (well-known maturational EEG changes). Furthermore, we predict that non-responders to stimulant medication would have a low TBR compared to non-responders. In addition, we hypothesize in line with our earlier study (Arns et al., 2008) that male ADHD non-responders (NR) would have a lower APF compared to responders (R).

2. Experimental procedures

2.1. Design

This study was a phase-IV, multi-site, international, open-label effectiveness trial in which ADHD patients were prescribed with MPH, including 7 international research sites. Full details of the study protocol have been published elsewhere (Elliott et al., 2014). This study was registered at the clinicaltrials register at www.clinicaltrials.gov with identifier NCT00863499 and IRB approval was obtained at all clinic sites. Parents and/or children provided written informed consent.

2.2. Study participants

The iSPOT studies have been explicitly apriori designed to use a two-step analysis procedure, where the first half of

Table 1 Demographic features of ADHD patients and controls, as well as responders and non-responders to treatment (per protocol sample).

	Controls	ADHD	Responders	Non-responders
Number	158	336	171	107
Males (%)	112 (71%)	245 (73%)	132 (77%)	70 (65%)
Average Age yrs. (SD)	12.2 (3.2)	11.9 (3.3)	12.2 (3.2)	11.5 (3.1)
Dosage, mg/kg (SD)			0.54 (0.36)	0.49 (0.30)
ADHD-RS-IV Total				
Baseline (SD)	3.59 (4.1)	36.72 (10.2)	36.89 (10.3)	37.22 (10.2)
Week 6 (SD)	3.99 (4.5)	24.2 (12.7)	17.27 (8.5)	35.72 (9.4)
Percentage Impr.		33.9%	53.1%	1.7%
ADHD subtype				
Combined		67%	62%	74%
Inattentive		32%	37%	25%
Hyperactive		1%	1%	1%

Abbreviations: RS = ADHD Rating Scale DSM-IV; SD = Standard Deviation; Impr. = improvement

the sample is used to identify potential predictors and moderators, whereas the second half will be used to replicate and confirm the results from the first half (Williams et al., 2011; Elliott et al., 2014). This study thus included 336 ADHD patients and 158 healthy controls, recruited between September 2009 and April 2012 (see Table 1 for demographics) and comprised the first cohort of children and adolescents (50%, N = 336) of iSPOT-A. In summary, the primary clinical diagnosis of ADHD was confirmed at baseline, preceding treatment, using the Mini International Neuropsychiatric Interview for Children and Adolescents (MINI kid: Sheehan et al., (2010) and clinician rated ADHD-Rating Scale-IV (ADHD-RS; Score of $\geq$ 6 items on the Inattentive or Hyperactive/Impulsive subscales (Zhang et al., 2005) and be unmedicated for 7 days prior to testing. No other primary diagnoses were allowed. Diagnostic interviews were conducted by well-trained research assistants/clinicians. Inter-rater reliability training for ADHD-RS-IV administration was provided and Inter-rater reliability for ADHD-RS-IV was assessed using a one-way, consistency, single-measures Intra-class Correlation Coefficient (ICC; McGraw and Wong, 1996) to assess the degree that coders provided consistency in their ratings of the ADHD-RS items. The resulting ICC was in the excellent range, ICC = 0.994 (Cicchetti, 1994), indicating that coders had a high degree of agreement and suggesting that the ADHD-RS items were rated similarly across coders.

2.3. Procedure

ADHD subjects were either treatment naïve or medication was washed out before baseline assessment (week 0), following recommendations on the package insert, and prescribed open-label MPH by their treating physician (ADHD subjects were submitted to MPH treatment for 6 weeks (=post-treatment) and were required to have a minimum duration of MPH treatment for 4 weeks; while refraining from other ADHD treatments, including other stimulants, non-stimulant ADHD drugs and non-pharmacological ADHD therapies during the first 6 weeks).

2.4. Pre-treatment assessments

The EEG recordings have been performed using a standardized methodology and platform (Brain Resource Ltd., Australia) for which full details have been published elsewhere (Arns et al., 2008; 2016; Williams et al., 2011), as have the results of the across-site consistency and reliability of this methodology (Paul et al., 2007; Williams et al., 2005). Summarized, children and adolescents were seated in a light and sound attenuated room and EEG data were collected from 26 channels (Quikcap; NuAmps; 10-20 electrode international system) from two minutes with EO and two minutes with EC and recordings took place during office hours and the operator did not intervene when drowsiness patterns were observed in the EEG. EEG signals were referenced to averaged mastoids and a ground at AFz. Vertical eye movements were recorded with electrodes placed 3 mm above the middle of the left eyebrow and 1.5 cm below the middle of the left bottom eyelid. Horizontal eye movements were recorded with electrodes placed 1.5 cm lateral to the outer canthus of each eye. Skin impedance was <5 KiloOhm for all electrodes (Sampling rate = 500 Hz; Low-pass filter of 100 Hz with attenuation of 40 dB per decade and no high-pass filter (DC)). In addition, subjects also took part in a broader neuropsychological testing battery, detailed in Elliott et al. (2014).

2.5. Analysis

2.5.1. EEG analysis

A detailed overview of the exact data-analysis procedure and validation against manual processing and deartifacting, can be found in Arns et al. (2016). "In summary, data were (1) filtered (0.3-100 Hz and notch); (2) EOG-corrected using a regression-based technique similar to that used by Gratton et al., (1983); (3) segmented in 4-second epochs (50% overlapping) and an automatic deartifacting method was applied (Arns et al., 2016).

The following EEG metrics were extracted from EO and EC resting states: TBR (4-8 Hz/13-21 Hz) and APF. APF was

assessed using a method similar to that used in Arns et al. (2012) (1) Fast Fourier Transform was applied to both EO and EC using 8192 millisecond segment epochs with 50% overlap to get a power spectrum for each site (with a Hamming window applied to each segment). (2) The difference between EO and EC power spectrum data was calculated in order to ensure alpha was quantified by its known suppression from EC to EO. (3) The APF for each site was scored by searching for the maximum value between 6-13 Hz in the power spectrum difference, found in step 2. For Theta/Beta ratio we specifically tested sites Fz, FCz, and Cz, because the Theta/Beta ratio is most often reported for these sites (for review see Arns et al., 2013) and for APF we specifically looked at Fz, FCz, Pz and Oz, since the alpha rhythm is most dominant at posterior sites (Pz and Oz), while prior studies have specifically implicated frontal APF to be associated to treatment response (Arns et al., 2012; Jin et al., 2006).

2.5.2. Statistics

The a priori defined primary outcome measure was clinical response, (>25% improvement on clinician rated ADHD-RS between baseline and post-treatment, rated by non-prescribing clinician).

Differences between groups (Group: ADHD vs. Controls and Response: Responders vs. Non-Responders) were tested using One-Way ANOVA's or non-parametric Chi-Square (Gender). For the comparison between ADHD and Controls, as well as analyses between responders and non-responders a repeated measures ANOVA with within-subject factor Condition (EO and EC) and Electrode Site (for TBR: 3 levels: Fz, FCz, Cz, and for APF: 4 levels: Fz, FCz, Pz, and Oz) and between-subject factors Group or Response, Age-group (children [6-11 yrs.] vs. adolescents [12-18 yrs.]) and Gender (males vs. females) was conducted. In order to further evaluate Group or Response differences dimensionally rather than categorically, partial correlations were calculated (controlling for age and gender) between the obtained EEG biomarker and symptom severity/symptom severity change. Effect sizes (ES) reported are Cohen's d (d).

2.5.2.1. Discriminant analysis

A discriminant analysis was performed to test whether the APF or TBR could predict treatment non-response or treatment response in ADHD male adolescents. In discriminant analysis, classification of groups is determined by predefined variables. If the model is significant, the predictor variables can accurately discriminate between the groups. Here, the first model included the APF and the second model included TBR. Receiver operating characteristic (ROC) curves and the area under the curve (AUC) were computed for both models.

Log-transformation was applied to not normally distributed data. For curve fitting procedures Prism 6 was used, all other statistics were computed in SPSS.

3. Results

See Table 1 for demographic features of all groups. There were no differences between ADHD and controls in Age ($p = .361$; average 12.0yrs.) and Gender ($p = .638$). Groups also did not differ on Age on the subgroup level (when examining only males, females, children, adolescents)). There were no differences between responders and non-responders with respect to Age ($p = .071$), baseline ADHD-RS (ADHD-RS Total, Inattention, and Hyperactive/Impulsive; all $p > .126$), prior treatment history (R: 40% vs. NR: 33% treatment naïve subjects; $p = .249$) and MPH dosage (mg/kg; $p = .197$). No significant differences were found between the per-protocol and intention to treat sample for baseline ADHD severity and age (all $p > .378$). Among responders there were significantly more males (77%) as compared to non-responders (65%: $p = .032$; Chi-square = 4.592) suggesting that females have a lower likelihood of responding to MPH. Furthermore, among responders there were significantly less patients with the combined subtype ($p = .047$; Chi-square = 3.946) and more with the inattentive subtype ($p = .033$, Chi-Square = 4.550). Log-transformation was applied to EEG TBR to yield normal distributions of the data.

See Fig. 1 for the power spectral plots for Fz and Pz, comparing ADHD patients with controls.

3.1. ADHD vs. Controls: TBR

Repeated measures ANOVA yielded main effects of Condition ($F(1,336) = 26.6$; $p < .001$), Electrode ($F(2335) = 47.6$; $p < .001$), Electrode X Age-group ($F(2335) = 5.2$; $p = .006$), Electrode X Group X Age-group ($F(2335) = 4.4$; $p = .012$) and Condition X Electrode ($F(2335) = 16.8$; $p < .001$) and Age-group ($F(1,336) = 14.9$; $p < .001$). Conducting this analysis separately per Age-group, did not reveal any main effects of Group or significant interactions involving Group, confirming no differences existed between ADHD and controls on TBR.

Partial correlations within the ADHD group, corrected for Age and Gender, yielded small and weakly significant correlations between TBR at electrode Fz and ADHD-RS total score (RsTotal) (EO: $r(232) = 0.138$; $p = .035$; EC: $r(232) = 0.174$, $p = .008$, $R^2 = 1.9$-3.0%) and inattention (EO: $r(232) = 0.132$; $p = .044$; EC: $r(232) = 0.135$, $p = .038$, $R^2 = 1.7$-1.8%) but no significant correlations for Cz. Given the large sample-size and repeated tests, these correlations, barring Fz_{EC}-RsTotal, would not have met significance using Bonferroni corrected-values.

3.2. ADHD vs. Controls: APF

A main effect of Electrode ($F(3,269) = 17.6$), $p < .001$) and Age-group ($F(1,271) = 11.9$, $p < .001$), but no interactions involving Group or main effect of Group ($p = .857$) were found. No differences between the ADHD and the control group were found for APF.

3.3. Treatment prediction

From the 332 CEHD patients included in the study, 278 patients (83%) attended for the week 6 visit (treatment outcome could be established) and adhered to the protocol, and were part of the per protocol sample. See Table 1 for demographics of the responder groups.

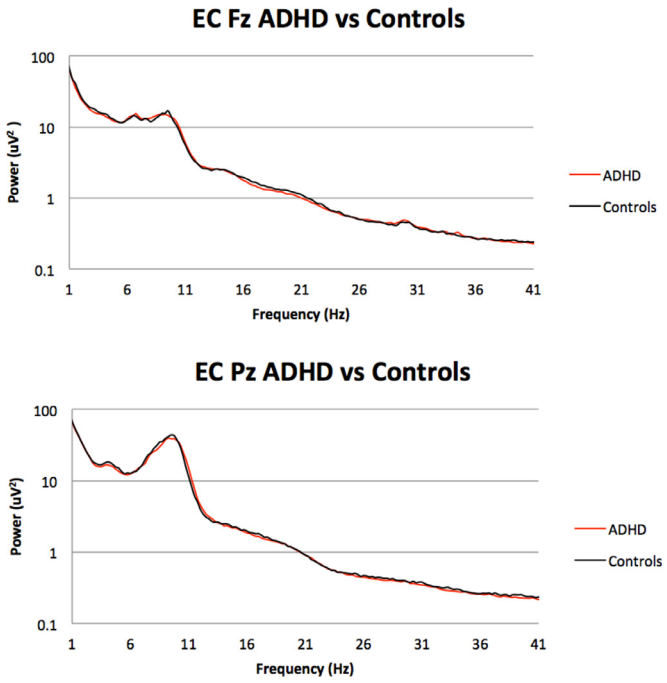

Fig. 1 The EEG power spectra for ADHD (red) and Controls (black) for EC EEG at electrode Fz (top) and Pz (bottom). Note that the power spectra are almost identical, suggesting no differences in resting state EEG between ADHD and controls at group level. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

3.3.1. Responders vs. non-responders: TBR

Due to interactions involving Condition, the analysis for TBR was repeated for EO and EC separately. For EC, a main effect of Electrode ($F(2,191) = 16.2$, $p < .001$) and Age-group ($F(1192) = 30.0$, $p < .001$), but no interactions with, or main effect of Response ($p = .857$) were found. Similar results were found for EO. Repeating the analysis for Males and Females separately and for Children and Adolescents separately, yielded no significant results, illustrating no difference on TBR between responders and non-responders. No significant correlations between baseline TBR and percentage improvement on ADHD-RS were found.

3.3.2. Responders vs. non-responders: APF

Repeated measures ANOVA yielded a main effect of Electrode ($F(3,137) = 10.8$, $p < .001$) and a Response X Gender ($F(1,139) = 6.6$, $p = .011$), and an Age-group X Gender interaction ($F(1,139) = 4.4$, $p = .037$).

Repeating this analysis for girls only, yielded an effect of Electrode ($F(3,39) = 3.8$, $p = .017$) and Age-group ($F(1,41) = 4.9$, $p = .033$), but no main effect of Response ($p = .064$) nor Response X Age-group interaction ($p = .548$). The trend effect for response for girls was in the opposite direction as compared to the findings for male adolescents.

For boys only, a main effect of Electrode ($F(3,96) = 10.1$, $p < .001$), and interaction effects for Electrode X Response ($F(3,96) = 4.4$, $p = .006$) and Electrode X Response X Age-group ($F(3,96) = 4.0$, $p = .011$) were found. Limiting the analysis to children, yielded an Electrode X Response interaction ($F(3,47) = 3.8$, $p = .017$) but no main effect for response ($p = .839$). Univariate analysis resulted in non-significant Response effects for Pz ($p = .070$), Oz ($p = .730$), FCz ($p = .482$), and Fz ($p = .554$).

Limiting the analysis to adolescents, yielded an effect of Electrode ($F(3,47) = 8.4$, $p < .001$), an Electrode X Response interaction ($F(3,47) = 3.8$, $p = .016$) and a trend toward significance for Response ($F(1,49) = 3.9$, $p = .053$). Univariate analysis resulted in a significant main effect for Response for Fz ($F(1,61) = 9.1$, $p = .004$; $d = 0.83$: $R = 9.2$ Hz; $NR = 8.1$ Hz) and FCz ($F(1,61) = 4.9$, $p = .031$; $d = 0.60$: $R = 9.3$ Hz; $NR = 8.5$ Hz), but not for Pz ($p = .579$) and Oz ($p = .828$).

For the APF measures in male adolescents there were no correlations with age ($p > .178$) and MPH dosage ($p > .328$). Male adolescent non-responders were adequately dosed and no differences in dosing were obvious ($p = .423$; NR: 0.53 mg/kg vs. $R = 0.46$ mg/kg). Within the subgroup of male adolescents, baseline APF was correlated to baseline Hyperactivity/Impulsivity (Fz: $r(64) = -0.332$, $p = .007$, $R^2 = 11\%$; FCz: $r(64) = -0.340$, $p = .006$, $R^2 = 11.6\%$) but not to baseline Inattention ($p > .630$) or RS-total ($p > .058$). Furthermore, these baseline APF measures correlated to percentage improvement on ADHD-RS (Fz: $r(62) = 0.279$, $p = .028$, $R^2 = 7.8\%$); Inattention (Fz: $r(62) = 0.304$, $p = .016$, $R^2 = 9.2\%$) but not Hyperactivity/Impulsivity (Fz: $p = .062$; FCz: $p = .129$). Partial correlations controlling for baseline Hyperactivity/Impulsivity still yielded significant correlations for percentage improvement on Inattention (Fz: $r(44) = 0.323$, $p = .029$), suggesting that baseline APF at Fz is an independent predictor for treatment outcome in male adolescents, and not mediated by baseline ADHD-RS severity. For the whole sample (including females and children) there were no significant correlations between APF and percentage improvement on ADHD-RS, neither when controlling for age. In the subgroup of male adolescents no significant differences were found

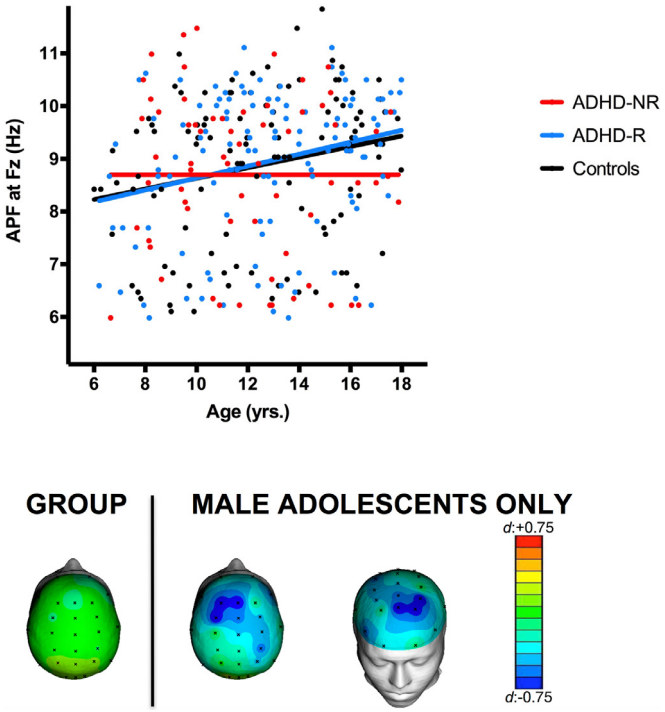

Fig. 2 Top: APF at electrode Fz for Controls, ADHD Responders and Non-Responders plotted against age with the significant fitted trend lines. For both Controls and Responders, a clear maturational effect can be seen where APF becomes faster with age. However, for Non-Responders the best fit was a horizontal line indicating a maturational stagnation for the non-responder group (because significant differences between groups only emerged in adolescents). Bottom: Topographical localization of the low APF for all adolescents combined (Group: left) and male adolescents only (right) (in effect size with a range of $d > -0.075$ to $d < 0.075$), further demonstrating the effect was specifically explained by male-adolescents.

between R and NR and various cognitive tasks that have previously been reported to be associated with APF e.g. spontaneous verbal memory recall (all $p > .587$); Digit span (all $p > .324$); Choice reaction time ($p = .831$) and WM/CPT (all $p > .352$), suggesting low APF is uniquely associated with MPH non-response.

A scatterplot for APF plotted against Age for Controls, responders and non-responders can be seen in figure. Both responders and Controls show the expected maturational change in APF, confirmed by curve fitting where a line with slope was a significantly better fit as compared to a horizontal line (R: $p = .004$, $R^2 = 6.6\%$; Controls: $p = .02$, $R^2 = 4.9\%$) whereas for the NR's no line with a slope could be identified with a better fit than a horizontal line ($p = .435$, $R^2 = 1.0\%$).

In order to further understand the strength of prediction for APF we calculated the response rate for different cut-points. Overall response rate for male adolescents was 71.8% (total $N = 85$). When using ≥ 9 Hz as a cut-off for APF at Fz, the response rate increased to 86.5% (total $N = 37$).

3.3.3. Discriminant analysis

When conducting a discriminant analysis on the male adolescent sample to predict NR, a model comprising APF (using the individual APF in Hz) yielded a significant model ($p = .004$; Wilks' Lambda $= 0.869$; Chi-square $= 8.376$; df $= 1$), while a model comprising TBR did not ($p = .974$; Wilks' Lambda $= 1.000$; Chi-square $= 0.001$; df $= 1$). Below in Fig. 4 also see the corresponding ROC curves for both models, where it is visualized that only the model comprising APF significantly predicts group membership.

4. Discussion

The primary aims of this study were (1) to investigate ADHD specific differences in brain function compared to typically developing children and adolescents and (2) to investigate predictors of treatment response to MPH within the ADHD sample. Results failed to show differences in TBR and APF between ADHD and controls. Furthermore, an age and gender specific effect was found for APF, where male adolescents with a low APF were more likely to be non-responders to MPH. TBR was not associated with treatment response.

4.1. Diagnostic EEG differences

The absence of a difference in the TBR between ADHD and controls is in line with our initial hypothesis and expectations based on our prior meta-analysis (Arns et al., 2013; 2016a), also visualized in Fig. 5 which is an updated figure from this meta-analysis including the ES from this study in black. These studies showed that the dichotomous difference in the TBR between ADHD and controls as found in older studies, lacked in recent studies. As can be observed from these data, results from this study are in line with other recent studies that do not find a difference between

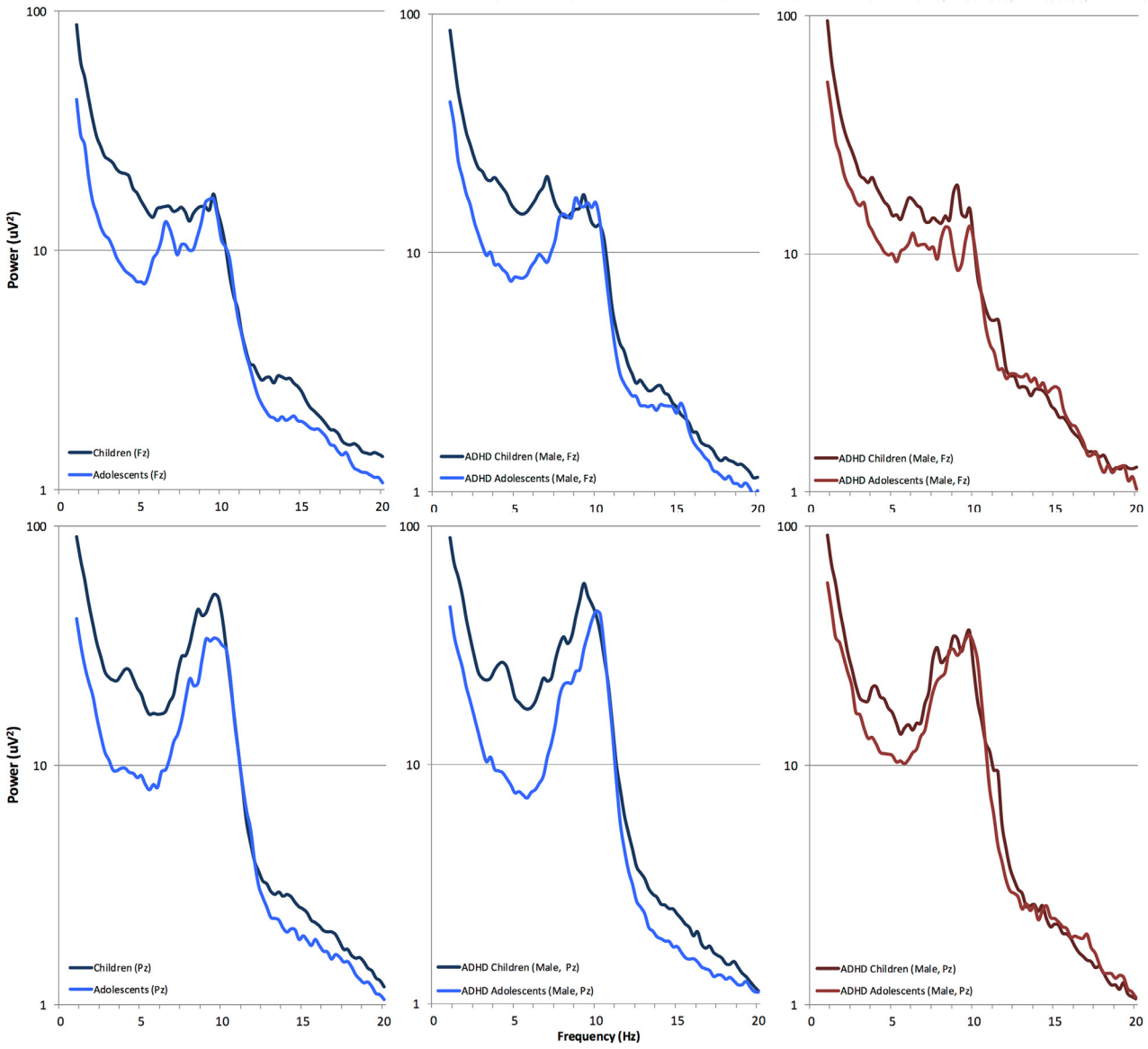

Fig. 3 The EC power spectra between 0–20 Hz for controls (left), male ADHD Responders (middle) and male ADHD Non-Responders (right) with children and adolescents overlaid to facilitate visualizing the developmental changes in the EEG from childhood to adolescence (children in darker color-gradient). Note the well-known maturational changes in the healthy controls, also visible in MPH responders, characterized by a decrease in theta activity, and a faster APF, most clearly visible at Pz. Note that this maturational change is almost absent for male non-responders, further emphasizing the maturational stagnation visualized in Fig. 2. (EC = Eyes Closed; Hz = Hertz; MPH = Methylphenidate).

ADHD and non-ADHD groups, expressed by the small effect sizes around 0.2–0.3 and thus confirm the trend that TBR can no longer be considered a reliable diagnostic marker for ADHD (Arns et al., 2013; 2016a). This is further confirmed by weak and mostly non-significant correlations with ADHD symptoms (only explaining 1.9–3.0% of the variance). Furthermore, inspection of Fig. 1 further confirms the lack of differences in EEG spectral power between the ADHD and controls.

For APF, neither differences were found between the ADHD and controls, nor interactions with gender or age-group. Most studies that were suggestive of such a difference, concerned older studies (Capute et al., 1968; Cohn and Nardini, 1958; Jasper et al., 1938; Stevens et al., 1968) that were conducted in children with diagnosis such as MBD rather than the current DSM-IV or DSM 5 definition of ADHD (Capute et al., 1968).

4.2. Treatment outcome

Current results did not replicate previous reports of high TBR in responders to MPH (Arns et al., 2008; Clarke et al., 2002b; Ogrim et al., 2014; Satterfield et al., 1971; Suffin and Emory, 1995). Since previous studies were generally small ($N < 100$), both the diagnostic as well as prognostic value of TBR in ADHD is questionable.

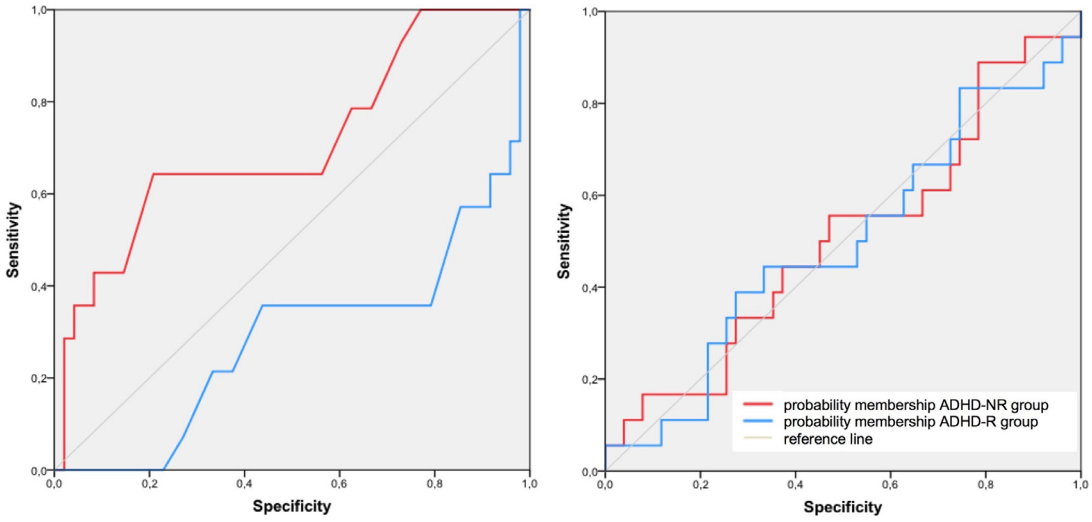

Fig. 4 ROC curves for two different models visualizing the probability of membership of the ADHD non-response group (red) and the probability of membership of the ADHD response group (blue) in male adolescents. Left: the ROC curve based on APF (AUC = 0.709/0.291). Right: the ROC curve based on TBR (AUC = 0.504/0.496), showing the clearest separation for APF at Fz. (ROC = Receiver Operator Curves; APF = Alpha Peak Frequency; AUC = Area Under the Curve). (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

However, for APF an age and gender specific effect was found, where male adolescents with a low APF were more likely to be non-responders to MPH with a large ES ($d = 0.83$). The APF in adolescent males was found to correlate to percentage improvement on the ADHD-Rating Scale-IV total and inattention after treatment with 7.8-9.2% explained variance. This correlation was not explained by baseline severity, because results remained significant when controlling for baseline severity and these effects were not mediated by impaired cognition. This result replicates our earlier study that only included male ADHD patients (Arns et al., 2008) and extends the finding to specifically apply to male adolescents.

As can be seen in Fig. 2, both controls and ADHD responders demonstrated the typically expected maturational speeding up of the APF with increasing age. However, for male non-responders the association was best explained by a horizontal line with slope zero. Results suggest that the APF difference emerges at adolescence onset because the non-responder group only significantly differed from responders in adolescents and not in children (also further visualized in Fig. 3). Although these data are cross-sectional, they suggest that in a subgroup of male ADHD patients a developmental stagnation occurs at the onset of adolescence, resulting in a stagnation of brain development and associated with non-response to MPH. This also suggests a different etiology for this subgroup of patients. Furthermore, these results replicate earlier studies where a low APF was associated with non-response to stimulant medication in male ADHD patients (Arns et al., 2008) and antidepressant treatments (Arns et al., 2012; 2010; Ulrich et al., 1984).

Relatively few studies have systematically investigated sex-specific effects, mostly due to a lack of statistical power. Recently, Loo et al. (2017) reported based on a large sample of 781 children with ADHD an overrepresentation of males in a Delta and Theta EEG clusters, further confirming sex-specific EEG differences in ADHD. Furthermore, we recently also reported several sex-specific EEG predictors and findings from a large multicenter depression study in 1008 depressed patients, such as right frontal alpha asymmetry related to SSRI response for females only (Arns et al., 2016), smaller N100 ERP amplitudes in male non-responders to venlafaxine-XR (van Dinteren et al., 2015), overall increased alpha and theta connectivity within the DLPFC-sgACC network for females relative to males and state related decreases in alpha connectivity for males only (Iseger et al., 2017). Therefore, at this stage we cannot fully interpret the sex-specific effects, but these findings do urge future studies to focus more on sex-specific effects in ADHD.

How can we explain a low APF in male adolescent non-responders to MPH? Oscillatory activity in the alpha band is thought to reflect functional inhibition, gating information by inhibiting task-irrelevant regions, thereby routing information to task-relevant regions (Jensen & Mazaheri, 2010). A deviant pattern in the modulation of alpha oscillations during covert attentional performance -thought to be a robust design to investigate the inhibitory role of alpha oscillations in healthy adults (e.g. Händel et al., 2011; ter Huurne et al., 2013) and children (Vollebregt et al., 2015) - has been observed in boys with ADHD (Vollebregt et al., 2016) and adults with ADHD (ter Huurne et al., 2013). Following the hypothesis that alpha plays a functional inhibitory role, the process of allowance and stopping of information transfer is slower in subjects with a low APF (Grandy et al., 2013). In healthy adults, APF is correlated with general cognitive abilities (Grandy et al., 2013). A low APF is thus expected to have broad consequences for behavior. As was proposed earlier in a review on APF and treatment response, APF is also associated with cerebral blood flow (Arns, 2012). Based on these findings taken together with the current results, we speculate that a different etiology may underlie the symptomatology in this subgroup of patients with

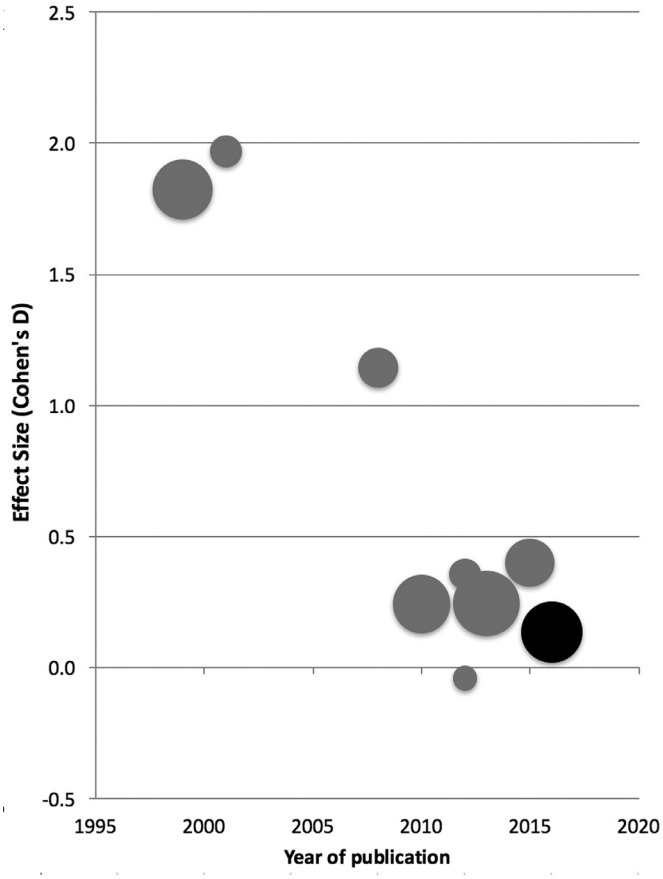

Fig. 5 Updated data from meta-analysis by Arns et al. (2013) depicting the ES (difference in TBR at Cz taken from EO condition between ADHD and controls (6–18 yrs.), expressed in Cohen's d; Y-axis) and year of publication (X-axis) with size of the sphere reflecting sample size. The black sphere toward the right depicts the ES from the current study, demonstrating the obtained results are in line with other recent studies.

a low APF compared to patients with an APF in the normal range. Interestingly, in a previous report on the iSPOT trial in depression in 1008 depressed patients, we found that a low APF was specifically associated with *favorable* response to Sertraline (but no association for venlafaxine-XR and escitalopram), hypothesized to be related to its higher DAT inhibitory activity and higher D2 receptor binding activity (Arns et al., 2015a), offering a possible new lead for a biomarker driven drug-development for this subgroup.

Summarizing, this study failed to find clear differences in EEG measures between ADHD and controls, further questioning the psychiatric diagnostic use of EEG (Arns et al., 2016b). For treatment prediction however, clear gender and age-group (child vs. adolescent) differences were found, where a low APF in male adolescents with ADHD was associated with a smaller likelihood of responding to MPH with large effect sizes, robustly replicating earlier work (Arns et al., 2008).

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Author contributions

MA initiated the proposal for this manuscript, conducted the statistical analyses and the EEG processing. DP, CS and MAV were involved in data processing and data analysis, and development of the manuscript. MA, MAV, DP, CS, EG, MK, SC, GE and JKB read and contributed to the manuscript.

References

Arns, M., 2012. EEG-based personalized medicine in ADHD: Individual alpha peak frequency as an endophenotype associated with nonresponse. J. Neurother. 16, 123–141.

Arns, M, Bruder, G, Hegerl, U, Spooner, C, Palmer, DM, Etkin, A, et al., 2015. EEG alpha asymmetry as a gender-specific predictor of outcome to acute treatment with different antidepressant medications in the randomized iSPOT-D study. Clin. Neurophysiol. 509–519.

Arns, M, Conners, CK, Kraemer, HC, 2013. A decade of EEG theta/beta ratio research in ADHD: a meta-analysis. J. Atten. Disord. 17, 374–383.

Arns, M, Drinkenburg, P, Fitzgerald, P, Kenemans, L, 2012. Neurophysiological predictors of non-response to rTMS in depression. Brain Stimul. 5, 569–576.

Arns, M, Drinkenburg, W, Kenemans, JL, 2012. The effects of QEEG-informed neurofeedback in ADHD: an open-label pilot study. Appl. Psychophysiol. Biofeedback 37, 171–180.

Arns, M, Gordon, E, 2014. Quantitative EEG (QEEG) in psychiatry: diagnostic or prognostic use? Clin. Neurophysiol. 125, 1504–1506.

Arns, M, Gunkelman, J, Breteler, M, Spronk, D, 2008. EEG phenotypes predict treatment outcome to stimulants in children with ADHD. J. Integr. Neurosci. 7, 421–438.

Arns, M, Kenemans, JL, 2014. Neurofeedback in ADHD and insomnia: Vigilance stabilization through sleep spindles and circadian networks. Neurosci. Biobehav. Rev. 4, 183–194.

Arns, M, Loo, SK, Sterman, MB, Heinrich, H, Kuntsi, J, Asherson, P, et al., 2016. Editorial perspective: how should child psychologists and psychiatrists interpret FDA device approval? Caveat emptor. J. Child Psychol. Psychiatry 57, 656–658.

Arns, M, Spronk, D, Fitzgerald, PB, 2010. Potential differential effects of 9 hz rTMS and 10 hz rTMS in the treatment of depression. Brain Stimul. 3, 124–126.

Capute, AJ, Niedermeyer, EFL, Richardson, F, 1968. The electroencephalogram in children with minimal cerebral dysfunction. Pediatrics 41, 1104.

Cicchetti, DV, 1994. Guidelines, criteria, and rules of thumb for evaluating normed and standardized assessment instruments in psychology. Psychol. Assess. 6, 284.

Clarke, AR, Barry, RJ, McCarthy, R, Selikowitz, M, 2002. EEG differences between good and poor responders to methylphenidate and dexamphetamine in children with attention-deficit/hyperactivity disorder. Clin. Neurophysiol. 113, 194–205.

Clarke, AR, Barry, RJ, McCarthy, R, Selikowitz, M, Croft, RJ, 2002. EEG differences between good and poor responders to methylphenidate in boys with the inattentive type of attention-deficit/hyperactivity disorder. Clin. Neurophysiol. 113, 1191–1198.

Cohn, R, Nardini, JE, 1958. The correlation of bilateral occipital slow activity in the human EEG with certain disorders of behavior. Am. J. Psychiatry 115, 44–54.

Elliott, GR, Blasey, C, Rekshan, W, Rush, AJ, Palmer, DM, Clarke, S, et al., 2014. Cognitive testing to identify children with ADHD who do and do not respond to methylphenidate. J. Atten. Disord. doi:10.1177/1087054714543924.

Gevensleben, H, Holl, B, Albrecht, B, Schlamp, D, Kratz, O, Studer, P, et al., 2009. Distinct EEG effects related to neurofeedback training in children with ADHD: A randomized controlled trial. Int. J. Psychophysiol 74, 149–157.

Grandy, TH, Werkle-Bergner, M, Chicherio, C, Lövdén, M, Schmiedek, F, Lindenberger, U, 2013. Individual alpha peak frequency is related to latent factors of general cognitive abilities. Neuroimage 79, 10–18.

Gratton, G, Coles, MG, Donchin, E, 1983. A new method for off-line removal of ocular artifact. Electroencephalogr Clin. Neurophysiol. 55, 468–484.

Händel, BF, Haarmeier, T, Jensen, O, 2011. Alpha oscillations correlate with the successful inhibition of unattended stimuli. J. Cogn. Neurosci. 23, 2494–2502.

Iseger, T.A., Korgaonkar, M.S., Kenemans, J.L., Grieve, S.M., Baeken, C., Fitzgerald, P.B., Arns, M., 2017. EEG connectivity between the subgenual anterior cingulate and prefrontal cortices in response to antidepressant medication. Eur. Neuropsychopharmacol. doi:10.1016/j.euroneuro.2017.02.002.

Jasper, HH, Solomon, P, Bradley, C, 1938. Electroencephalographic analyses of behavior problem children. Am. J. Psychiatry 95, 641.

Jensen, O, Mazaheri, A, 2010. Shaping functional architecture by oscillatory alpha activity: gating by inhibition. Front Hum. Neurosci. 4, 186.

Jin, Y., Potkin, S.G., Kemp, A.S., Huerta, S.T., Alva, G., Thai, T.M., Bunney, W.E., 2006. Therapeutic effects of individualized alpha frequency transcranial magnetic stimulation (alphatms) on the negative symptoms of schizophrenia. Schizophrenia Bull. 32 (3), 556–561. doi:10.1093/schbul/sbj020.

Loo, S.K., McGough, J.J., McCracken, J.T., Smalley, S.L., 2017. Parsing heterogeneity in attention-deficit hyperactivity disorder using EEG-based subgroups. J. Child Psychol. Psychiatry, Allied Discip. doi:10.1111/jcpp.12814.

McGraw, KO, Wong, SP, 1996. Forming inferences about some intraclass correlation coefficients. Psychol. Methods 1, 30.

Monastra, VJ, Lubar, JF, Linden, M, 2001. The development of a quantitative electroencephalographic scanning process for attention deficit-hyperactivity disorder: Reliability and validity studies. Neuropsychology 15, 136–144.

Monastra, VJ, Monastra, DM, George, S, 2002. The effects of stimulant therapy, EEG biofeedback, and parenting style on the primary symptoms of attention-deficit/hyperactivity disorder. Appl. Psychophysiol. Biofeedback 27, 231–249.

Ogrim, G, Hestad, KA, Kropotov, J, Sandvik, L, Candrian, G, Brunner, JF, 2014. Predicting the clinical outcome of stimulant medication in pediatric attention-deficit/hyperactivity disorder: data from quantitative electroencephalography, event-related potentials, and a go/no-go test. Neuropsychiatr. Dis. Treat 231.

Paul, RH, Gunstad, J, Cooper, N, Williams, LM, Clark, CR, Cohen, RA, et al., 2007. Cross-cultural assessment of neuropsychological performance and electrical brain function measures: additional validation of an international brain database. Int. J. Neurosci. 117, 549–568.

Satterfield, JH, Lesser, LI, Podosin, RL, 1971. Evoked cortical potentials in hyperkinetic children. Calif. Med. 115, 48.

Sheehan, DV, Sheehan, KH, Shytle, RD, Janavs, J, Bannon, Y, Rogers, JE, et al., 2010. Reliability and validity of the mini inter-

national neuropsychiatric interview for children and adolescents (MINI-KID). J. Clin. Psychiatry 71, 313–326.
Stevens, JR, Sachdev, K, Milstein, V, 1968. Behavior disorders of childhood and the electroencephalogram. Arch. Neurol 18, 160.
Suffin, SC, Emory, WH, 1995. Neurometric subgroups in attentional and affective disorders and their association with pharmacotherapeutic outcome. Clin. Electroencephalogr. 26, 76–83.
ter Huurne, N, Onnink, M, Kan, C, Franke, B, Buitelaar, J, Jensen, O, 2013. Behavioral consequences of aberrant alpha lateralization in attention-deficit/hyperactivity disorder. Biol. Psychiatry 74, 227–233.
Ulrich, G, Renfordt, E, Zeller, G, Frick, K, 1984. Interrelation between changes in the EEG and psychopathology under pharmacotherapy for endogenous depression. A contribution to the predictor question. Pharmacopsychiatry 17, 178–183.
van Dinteren, R., Arns, M., Kenemans, L., Jongsma, M.L., Kessels, R.P., Fitzgerald, P., Williams, L.M., 2015. Utility of event-related potentials in predicting antidepressant treatment response: an iSPOT-D report. Eur. Neuropsychopharmacol. doi:10.1016/j.euroneuro.2015.07.022.
Vollebregt, MA, Zumer, JM, Ter Huurne, N, Buitelaar, JK, Jensen, O, 2016. Posterior alpha oscillations reflect attentional problems in boys with attention deficit hyperactivity disorder. Clin. Neurophysiol. 127, 2182–2191.
Vollebregt, MA, Zumer, JM, Ter Huurne, N, Castricum, J, Buitelaar, JK, Jensen, O, 2015. Lateralized modulation of posterior alpha oscillations in children. Neuroimage 123, 245–252.
Walter, W, Dovey, VJ, 1944. Electro-encephalography in cases of sub-cortical tumour. J. Neurol. Neurosurg. Psychiatry 7, 57.
Williams, LM, Rush, AJ, Koslow, SH, Wisniewski, SR, Cooper, NJ, Nemeroff, CB, et al., 2011. International study to predict optimized treatment for depression (iSPOT-D), a randomized clinical trial: rationale and protocol. Trials 12, 4.
Williams, LM, Simms, E, Clark, CR, Paul, RH, Rowe, D, Gordon, E, 2005. The test-retest reliability of a standardized neurocognitive and neurophysiological test battery: "Neuromarker. Int. J. Neurosci. 115, 1605–1630.
Zhang, S, Faries, DE, Vowles, M, Michelson, D, 2005. ADHD rating scale IV: psychometric properties from a multinational study as a clinician-administered instrument. Int. J. Methods Psychiatr. Res. 14, 186–201.