ORIGINAL CONTRIBUTION



Beneath the surface: hyper-connectivity between caudate and salience regions in ADHD fMRI at rest

Stefano Damiani¹ · Livio Tarchi¹ · Andrea Scalabrini² · Simone Marini³ · Umberto Provenzani¹ · Matteo Rocchetti¹ · Francesco Oliva⁴ · Pierluigi Politi¹

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Abstract

Attention-Deficit/Hyperactivity Disorder (ADHD) comprises disturbances in attention, emotional regulation, and reward-related processes. In spite of the active efforts in researching neurofunctional correlates of these symptoms, how the activity of subcortical regions—such as basal ganglia—is related to ADHD has yet to be clarified. More specifically, how age may influence the critical changes observed in functional dynamics from childhood to adulthood remains relatively unexplored. We hence selected five core subcortical regions (amygdala, caudate, putamen, pallidum and hippocampus) as regions of interest from the previous literature, measuring their whole-brain voxel-wise rsFC in a sample of 95 ADHD and 90 neuro-typical children and adolescents aged from 7 to 18. The only subcortical structure showing significant differences in rsFC was the caudate nucleus. Specifically, we measured increased rsFC with anterior cingulate and right insula, two mesolimbic regions pertaining to the Salience Network. The degree of hyper-rsFC positively correlated with ADHD symptomatology, and showed different patterns of evolution in ADHD vs neurotypical subjects. Finally, the rsFC scores allowed a fair discrimination of the ADHD group (Area Under the Curve \geq 0.7). These findings shed further light on the fundamental role covered by subcortical structures in ADHD pathogenesis and neurodevelopment, providing new evidence to fill the gap between neurofunctional and clinical expressions of ADHD.

AUC

 $\textbf{Keywords} \ \ \text{Attention deficit hyperactivity disorder} \cdot fMRI \cdot Caudate \ nucleus \cdot Salience \ network \cdot Reward$

| Abbrevi ADHD AFNI | iations Attention deficit hyperactivity disorder Analysis of Functional NeuroImages |
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| Stefano D work. | amiani and Livio Tarchi have contributed equally to the |
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| ☐ Livio | Tarchi archi01@universitadipavia.it |
| | rtment of Brain and Behavioral Sciences, University via, Via Bassi 21, 27100 Pavia, PV, Italy |
| | rtment of Psychological, Health and Territorial Sciences PuTer), G. D'Annunzio University of Chieti-Pescara, |

Via dei Vestini 33, 66100 Chieti, CH, Italy

Mowry Road, Gainesville, FL 32610, USA

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Department of Epidemiology, University of Florida, 2004

Department of Clinical and Biological Sciences, University of Turin, Regione Gonzole 10, 10043 Orbassano, TO, Italy

| Bcaud | Bilateral caudate |
|--------------|--|
| DVARS | Spatial standard deviation from volume N to |
| | volume N+1 |
| FDR | False Discovery Rate |
| fMRI | Functional magnetic resonance imaging |
| GSR | Global Signal Regression |
| rsFC | Resting-state functional connectivity |
| FSIQ | Estimates of Full-Scale Intelligent Quotient |
| MNI | Montreal Neurological Institute |
| rACC | Right Anterior Cingulate Gyrus |
| rINS | Right anterior insula |
| ROI | Region/regions of interest |
| SN | Salience network |
| TYP | Neurotypicals |
| vmPFC | Ventro-medial Pre-Frontal Cortex |

Area under the curve



Introduction

Attention-Deficit/Hyperactivity Disorder (ADHD) is a common condition affecting approximately 5% of schoolaged children and 2.5% of adults [1]. It is clinically characterized by a reduced ability to focus or sustain attention and excessive motor and emotional activity, which in turn leads to impulsivity and disorganization [1].

Structural imaging suggests a central role of subcortical regions such as caudate and amygdala in ADHD (see "The role of subcortical structures in ADHD pathogenesis"). In recent years, functional alterations in subcortical structures started to be explored [2]. However, these studies often yielded inconsistent findings that require further corroborations [3].

The role of subcortical structures in ADHD pathogenesis

Basal ganglia dimensions were found generally reduced or altered in shape in ADHD [4, 5].

The caudate nucleus is a core subcortical, bilateral structure regulating not only motor but also emotional, cognitive and perceptual functions [6]. Caudate serves as entry point to both the basal ganglia and the dopaminergic reward systems. In ADHD, caudate's architecture shows volumetric reductions and increased asymmetry [7]. Together with putamen and nucleus accumbens, caudate forms the striatum nucleus, which connections have been demonstrated to be altered in ADHD. An altered cortico-striatal connectivity has been linked to behavioral features characterizing ADHD, such as the inability to tolerate delays in receiving a reward (delay-discounting, see Patros et al. [8]).

Recently, mesolimbic structures such as amygdala also started to be considered among the key regions laying at the core of ADHD pathophysiology. The presence of structural changes in the amygdala of ADHD subjects has been confirmed by an increasing number of findings [9]. Intriguingly, these alterations share a strong bond with the individual behavioral phenotype: it has been demonstrated how volumetric reductions in amygdala have been connected to higher hyperactivity [10] and impulsivity [11] scores. If on one hand the involvement of deep subcortical structures in ADHD is strongly supported by the morphological evidences, on the other the studies trying to describe their role from a functional perspective often showed mixed results.

fMRI correlates of subcortical spontaneous activity in ADHD

In functional neuroimaging, current scientific literature suggests the presence of major alterations in motor, cognitive, and emotional tasks for ADHD [12, 13]. A second line of research focused instead on the spontaneous activity of the brain (also known as resting state, see references for a systematic review [2]). The degree of co-activation between different regions at rest (restingstate functional connectivity, or rsFC) may deeply influence stimuli-processing through complex and yet unclear dynamics [14–17]. Yang et al. [18] measured both hyperand hypo-rsFC between dorsal-caudate and several brain regions in subjects between the age of 7 and 13. Rosch and colleagues [19] found that dorso-lateral prefrontal cortex showed increased rsFC with striatum and decreased hypo-rsFC with the amygdala and hippocampus in children between the age of 8 and 12. Hulvershon et al. [20] focused on amygdala rsFC in relation with the emotional liability of the subject, finding a positive correlation between emotional liability and amygdala-anterior cingulate rsFC and a negative correlation between emotional liability and amygdala-insula rsFC. In a smaller sample, Cao et al. [21] narrowed their analyses on the putamen nucleus, finding altered rsFC patterns without any association to clinical symptoms. Another study showed increased rsFC between thalamus and putamen/caudate [22]. Oldehinkel et al. considered the rsFC of the reward system in adolescents and young adults, which showed no differences between ADHD and controls [23], but a positive relationship between symptoms and the increase of rsFC for caudate and putamen [24]. Altogether, the studies on ADHD subcortical structures' rsFC have often conducted region-based analyses, leaving open questions about the rsFC dynamics between subcortical structures and the whole-brain. In addition, the crucial diachronic component of age has been overlooked and a characterization of the maturational processes of the brain on observed results requires further exploration. As Zhou et al. [25] highlighted, not considering the age of the subject fMRI when measuring rsFC differences may lead to controversial results.

Brain maturation in ADHD

The discrepancies in basal ganglia morphology in ADHD tend to progressively reduce through adolescence [5].



As it has been shown morphologically for caudate, the functional relationship between amygdala and cortical structures also goes through important changes during the maturational processes of the individual [26]. In a similar way, ADHD's local connectivity appears to be altered in several areas (including subcortical regions [27]), and its topology changes from childhood to adolescence and adult age. Recent findings in healthy individuals showed that cortico-striatal rsFC progressively decreased through age, and a persistent hyper-rsFC was related to the presence of ADHD-like symptoms among others [28].

According to the aforementioned evidences, we may hypothesize that changes in rsFC are age-dependent and that these different developmental pathways on the neuronal side may subtend to clinical manifestations of ADHD.

Aims

We explored the rsFC of five subcortical regions (caudate, amygdala, putamen, pallidum and hippocampus), choosing them for their anatomical adjacency and their plausible involvement in the pathogenesis of ADHD (see "The role of subcortical structures in ADHD pathogenesis" and "fMRI correlates of subcortical spontaneous activity in ADHD"). The primary endpoint of the present study was to determine the presence of altered rsFC in ADHD when compared to neurotypical control subjects (TYP), expecting to find increased cortico-striatal rsFC and altered rsFC in amygdala. Specifically, we evaluated whether the strength of the functional connections between subcortical regions is differently influenced by age in these two groups. We predicted to find a delayed maturational process in ADHD with respect to the TYP trajectory according to the structural and preliminary functional evidence.

The secondary endpoints aim to explore (1) the relationship between the rsFC of subcortical structures and ADHD symptoms, possibly confirming previous literature and (2) the possibility to detect the presence of the disorder based on the fMRI findings.

Materials and methods

Sample

Our sample was obtained from the New York University dataset of the ADHD200 repository, specifically from the International Neuroimaging Data-Sharing Initiative. Scanning and sample characteristics are reported in Tables 1 and 2.

All the recordings were collected in accordance with the Declaration of Helsinki.

Estimates of Full-Scale Intelligent Quotient (FSIQ) above 80, right-handedness and absence of other chronic medical or psychiatric conditions were required for all subjects. A quality check for each subject was present in the phenotypic key provided with the dataset, and those subjects that did not pass were discarded preventively. Additional information can be found on the website (https://fcon_1000.projects.nitrc.org/indi/adhd200/).

Procedures

fMRI data preprocessing steps were implemented in AFNI (https://afni.nimh.nih.gov/afni, Cox 1996). First, the structural volume was corrected for signal intensity bias using AFNI's 3dUnifize at standard settings. The structural and functional reference images were then co-registered [29].

Table 1 Image acquisition

| Sample MR characteristics | | Functional imaging | | | | | |
|---------------------------|--|-----------------------|---------------|---------|---------|------------|------------------|
| | Scanner/software | Anatomical resolution | Scan duration | TR (ms) | TE (ms) | Flip angle | Resolution |
| ADHD200 | Siemens Magnetom Allegra syngo MR 2004A | 1.3×1.0×1.3 [mm] | 360 s | 2000 | 15 | 90 | 3.0×3.0×4.0 [mm] |

Table 2 Sample data

| Sample | Subj (n) | Subj (n) after motion correction | Mean age (years ± 1 SD) | Sex (M/F) | Medications |
|---------|-----------|----------------------------------|-------------------------|----------------------------|--|
| ADHD200 | ADHD: 123 | ADHD: 95 | ADHD: 11.42 ± 2.74 | ADHD: 72♂/23♀ 76% males | ADHD: 23 naïve 20 medicated 52 unknown |
| | TYP: 99 | TYP: 90 | TYP: 12.38 ± 3.12 | TYP: 44♂/46♀ 49% males | TYP: 90 naïve |

SD standard deviation



The first three frames of each fMRI run were removed to discard the transient effects in amplitude observed until magnetization achieves steady state [30]. Slice timing correction [31] and despike methods [32] were applied. Rigid-body alignment of the structural and functional image was performed. The anatomical image was then warped using the Montreal Neurological Institute standard space (MNI152_ T1 2009c) template provided with the AFNI binaries. Volume registration was then used to align the functional data to the base volume, warping it to the stereotactic space of choice. Spatial blurring was performed, with a kernel of full width at half maximum of 6 mm. Bandpass (0.01–0.25 Hz) was performed [33], to include the low-frequency bands from slow-5 to slow-2 [34]. Each of the voxel time series was then scaled to have a mean of 100. To control for nonneural noise, regression based on the six rigid body motion parameters and their derivates was applied, as well as mean time series from white matter and cerebro-spinal fluid masks [35] eroded by one voxel [36]. Whether preprocessing should include or not Global Signal Regression (GSR) is controversial [37]: to account for possible artifacts due to the exclusion of GSR, results were computed with and without the additional regression of the grey matter averaged timeseries. To further improve motion correction, censoring/scrubbing such as DVARS of two [38] and Framewise Displacement of 0.5 mm was applied [39] to the timeseries: two subjects (one ADHD-one TYP) were excluded from the fMRI analysis, as they exceeded the cut-off of 5% censored timepoints.

Primary aim

Resting-state functional connectivity—voxel-wise analysis

Based on previous literature (see "The role of subcortical structures in ADHD pathogenesis" to "Aims"), we selected caudate, amygdala, hippocampus, pallidum and putamen as seeds from the MNI atlas provided with the AFNI binaries. We extracted Left and Right regions from the atlas and combined them in a single bilateral region. An illustration of the ROIs used in this study can be found in the Supplementary Materials as Figure S1. Voxel-wise rsFC analysis was performed to investigate the differences between neurotypicals and ADHD patients during resting state. Individual rsFC maps were generated by calculating the Pearson's correlation coefficients between the mean time series of the seed and the time series of each voxel in the whole brain. In the t test between TYP and ADHD FC, clusters with more than 30 voxels at the threshold of p = 0.05 (corrected for False Discovery Rate, or FDR) were considered significant [40].

Resting-state functional connectivity—region-based analysis

To quantify the results' magnitude, single region significant voxel-wise masks were obtained and defined as regions of interest (ROIs). Mean values of correlations in those regions were extracted and compared between ADHD and TYP subjects. Statistical analyses included *t* test for the measure of rsFC mean value differences between ADHD subjects and neurotypicals. Pearson's coefficient was used to measure correlations/anticorrelations of the rsFC mean value with clinical scales. R-3.6.1 and R Studio-1.2.1335 were used to plot correlation figures.

Subsequently, age was used as a covariate and its effect estimated through an ANCOVA analysis on voxel-wise results. Mean values of correlations, mentioned above, were also averaged through groups and age classes, then plotted to follow their evolution in time. Two polynomial fits, each for diagnostic class, were used to represent the differences observed through age. The "ggplot2" library, in R/R-Studio, was used to graphically represent 95% confidence intervals of the polynomial fits.

Secondary aims

- Total ADHD, Hyperactivity and Inattention scores were collected using the Conners' Parent Rating Scale-Revised, Long Version [41] for all the available subjects (93 ADHD and 88 TYP, 2 missing observations for each class). Those scores were correlated with the above-mentioned ROIs to test whether the presence of an altered rsFC may be related or not to the phenomenological level.
- 2. ROC curves were built based on components obtained from the significant ROI's rsFC values, and the diagnostic power of AUC was evaluated. To do so, the software MATLAB 2019b (version 9.7.01190202) and its app "Classification Learner" were used. Two logistic regressions were fit using either rACC or rINS results in order to classify TYP and ADHD samples. A fivefold cross-validation was used to estimate the test error rate. For this kind of test, the sample was split into five subgroups (20% of the sample each). Each group was taken as a test set versus the remaining groups, which were considered as the training set. The model was thus fit on the training set and subsequently evaluated on the test set. Finally, the skill of the model was computed using the summarized evaluation scores of the five models [42].



Confirmatory and control analyses

The present study included the following control analyses:

- Motion during scan was evaluated through measurements of Framewise Displacement (FD). Mean values of FD were averaged per subject, then its distribution evaluated for diagnostic class. The "ggplot2" library was used to graphically illustrate mean Framewise Displacement values through a histogram plot and density curves, divided by diagnostic class.
- To account for possible artifacts due to the exclusion of Global Signal Regression (GSR), results were computed with the additional regression of the grey matter averaged timeseries.
- 3. Some ADHD subjects were not medication free, with type and dose of the medication not known (see Table 2). For all these subjects though, psychostimulant drugs were withheld at least 24 h before scanning. A *t* test was performed to compare the medicated and non-medicated population in ADHD patients, to exclude confounding effects given by the pharmacological status.
- 4. ADHD patients were not homogenous for diagnostic criteria applied, different sub-groups of ADHD diagnosis from DSM-5 were used for the inclusion criteria. *T* tests were conducted on the two significant FC-ROI to investigate the possible differences between clinical ADHD sub-groups. Hyperactive subjects were excluded from this confirmatory analysis as only one patient was present in our sample.
- 5. As the distribution of males and females was unequal between the two populations (see Table 2 for exact percentages), a control analysis for rsFC differences possibly due to gender was necessary. Hence, we performed four ROI-based *t* tests between TYP and ADHD computing: (1) Beaud-rACC rsFC for male subjects. (2) Beaud-rACC rsFC for female subjects. (3) Beaud-rINS rsFC for male subjects. (4) Beaud-rINS rsFC for female subjects.

 To confirm the specificity of our findings, we performed whole brain, voxel-wise rsFC analyses using the result regions from rACC and rINS. Masks obtained from result regions were used as seeds.

Results

Primary aim—voxel-wise analysis

No statistically significant difference was observed in rsFC between TYP and ADHD for bilateral amygdala, hippocampus, pallidum and putamen (Table 3). Conversely, when bilateral caudate (Bcaud) was considered as a seed, two significant ROI with increased rsFC in ADHD were found—right anterior cingulate cortex (rACC) and right anterior insula (rINS). Results are shown in Fig. 1 and Table 3; overlaps with the MNI coordinates are reported in Supplementary Materials (Table S1).

Primary aim—region-based analysis

rACC and rINS masks were generated. Average values of correlation between Bcaud and statistically significant ROIs (rACC and rINS) were calculated and subsequently used for confirmatory analyses. Mean rsFC values between the regions are shown in Table 4.

When rsFC values of significant ROI were extracted, a strong difference for developmental trends was observed between ADHD and TYP, especially in the range between 9- and 12-years old (Fig. 2). The trendline of this difference was better represented by a second-order polynomial (i.e., U-shaped curve, $r^2 = 0.7788$) rather than by a linear relationship ($r^2 = 0.0527$), with a peak at the age between 11 and 13 and a progressive reduction after that time window. Similar trends were observed for rINS, though with a smaller age effect than in rACC. Again, the distribution of differences better fitted to a second-order polynomial than to a linear trend. Mean differences per year of age are reported

Table 3 Whole brain, voxelwise functional connectivity *t* test between TYP and ADHD

| Seed (bilateral) | Smallest FDR p | Significant ROI* | Voxels forming the cluster (n) | Center of mass MNI coordinates (x,y,z. RAI) |
|------------------|----------------|------------------|--------------------------------|---|
| Caudate | 0.003 | rACC | 183 | -10.5, -40.5, +19.5 |
| | | rINS | 38 | -34.5, -16.5, -4.5 |
| Amygdala | 0.354 | _ | _ | _ |
| Hippocampus | 0.331 | _ | _ | _ |
| Pallidum | 0.228 | _ | _ | _ |
| Putamen | 0.209 | _ | _ | _ |

FDR False Discovery Rate correction



^{*}at FDR p = 0.05

Fig. 1 Graphical representation of the voxel-wise, resting-state functional connectivity of the bilateral caudate (t map, t test between neurotypical and ADHD groups; significant clusters at FDR corrected p value = 0.05). Red indicates increased functional connectivity in ADHD. The first row represents the result region identified as right Insula, the second row the result region of the right Anterior Cingulate Cortex. rINS right Insula, rACC right Anterior Cingulate Cortex

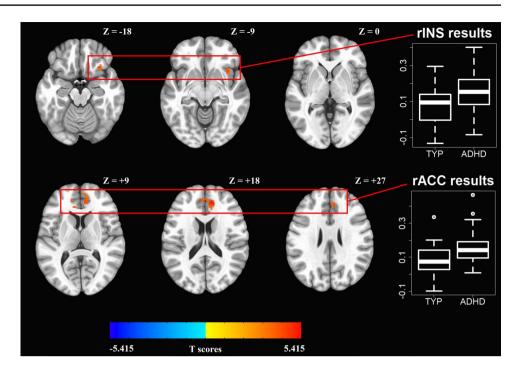


Table 4 Region-based analysis, comparison of Bcaud rsFC mean values between TYP and ADHD

| | Mean (variance) | | t stat | p value | |
|------|------------------------|------------------------|----------|----------|--|
| | TYP | ADHD | | | |
| rACC | 0.079286 (0.005897) | 0.153021 (0.006271) | -642.927 | 1.08E-09 | |
| rINS | 0.07335 (0.010524) | 0.149825 (0.010429) | -507.903 | 9.34E-07 | |

rACC right anterior cingulate, rINS right insula

for both samples in Table 5. 95% confidence intervals for polynomial fitted curves were plotted, to evaluate tail distributions, and can be found as Supplementary Figure S2. 95% confidence intervals did not overlap for the 9–15 years old groups, both for rACC and rINS results, whereas results for the <9 and >15 years old showed a convergence of the confidence intervals. To confirm statistical significance of the age effect, a series of t tests for mean differences in measured values between age sub-classes (<9, 9–15, >15) was calculated and can be found as Table S2a and b. Statistical significance was reached only for the 9–15 years old group, both for rACC and rINS results.

Secondary aims

A low to moderate, significant association of the Bcaud/rACC and Bcaud/rINS rsFC with (1) the main ADHD index; (2) Inattentive score; (3) Hyperimpulsive score of

the Connor Parents Rating Scale has been reported for the whole sample. The relationship of the rsFC values with symptoms and clinical scales, the correlations matrices and the significance levels are shown in Fig. 3. An overview of the correlations measured not by the whole sample, but within the diagnostic class, can be found in the Supplementary Materials as Table S3a and b.

RsFC values between the Bcaud seed and rACC/rINS were used to build predictive models, to estimate their diagnostic value in ADHD. Estimates of predictive performances were obtained after fivefold cross validation. ROC curves were plotted, reporting their AUC values (see Fig. 4). The logistic regression model built on Bcaud-rACC rsFC findings showed accuracy of 67%, sensitivity of 64% and specificity of 71%. The logistic regression model built on Bcaud-rINS showed an accuracy of 64.3%, sensitivity of 62% and specificity of 64%.

Confirmatory and control analyses

Motion during the scan is one of the fundamental drawbacks in fMRI studies [43]. As our sample is particularly vulnerable to such an effect, the degree of motion during the scan was evaluated through mean individual FD values. A t test of mean FD value per subject between TYP and ADHD patients was calculated and did not show a statistically significant difference (mean FD for TYP: 0.105 ± 0.003 ; mean FD for ADHD= 0.117 ± 0.004 ; p value for independent samples t test=0.197, see Table S4). The distribution of mean FD values per subject, per diagnostic class, was plotted and can be found as Supplementary Figure S3.



Functional Connectivity changes through age

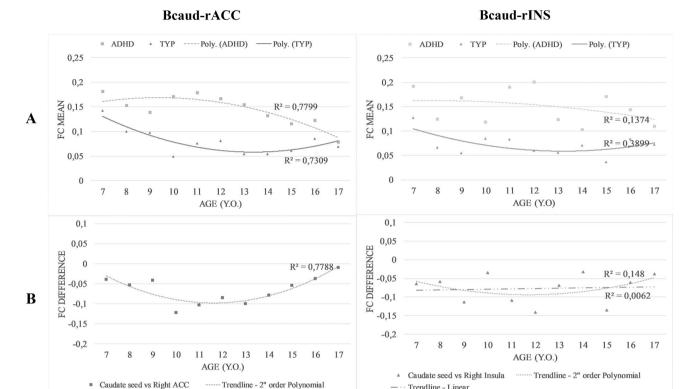


Fig. 2 Functional connectivity (rsFC) differences between neurotypicals and ADHD patients ordered by subjects' age. **a** *rsFC* mean value per age class, ADHD and TYP. **b** Differences and trendlines for rsFC

mean values and age class (TYP-ADHD). The trendline shows a better fit of the subjects' distribution to a U-shaped polynomial rather than to a linear trend

Table 5 Average rsFC values per age class

| rACC | | | rINS | | | |
|-------|--|--|---|---|--|--|
| TYP | ADHD | Difference | TYP | ADHD | Difference | |
| 0.143 | 0.182 | -0.039 | 0.127 | 0.192 | -0.065 | |
| 0.100 | 0.153 | -0.053 | 0.066 | 0.125 | -0.058 | |
| 0.098 | 0.140 | -0.042 | 0.055 | 0.169 | -0.113 | |
| 0.050 | 0.172 | -0.122 | 0.085 | 0.119 | -0.034 | |
| 0.076 | 0.179 | -0.103 | 0.083 | 0.191 | -0.108 | |
| 0.082 | 0.167 | -0.085 | 0.061 | 0.201 | -0.141 | |
| 0.054 | 0.155 | -0.100 | 0.056 | 0.124 | -0.068 | |
| 0.054 | 0.133 | -0.078 | 0.071 | 0.103 | -0.033 | |
| 0.061 | 0.116 | -0.055 | 0.036 | 0.171 | -0.135 | |
| 0.086 | 0.123 | -0.037 | 0.084 | 0.144 | -0.061 | |
| 0.069 | 0.079 | -0.010 | 0.072 | 0.110 | -0.038 | |
| | TYP 0.143 0.100 0.098 0.050 0.076 0.082 0.054 0.054 0.061 0.086 | TYP ADHD 0.143 0.182 0.100 0.153 0.098 0.140 0.050 0.172 0.076 0.179 0.082 0.167 0.054 0.155 0.054 0.133 0.061 0.116 0.086 0.123 | TYP ADHD Difference 0.143 0.182 -0.039 0.100 0.153 -0.053 0.098 0.140 -0.042 0.050 0.172 -0.122 0.076 0.179 -0.103 0.082 0.167 -0.085 0.054 0.155 -0.100 0.054 0.133 -0.078 0.061 0.116 -0.055 0.086 0.123 -0.037 | TYP ADHD Difference TYP 0.143 0.182 -0.039 0.127 0.100 0.153 -0.053 0.066 0.098 0.140 -0.042 0.055 0.050 0.172 -0.122 0.085 0.076 0.179 -0.103 0.083 0.082 0.167 -0.085 0.061 0.054 0.155 -0.100 0.056 0.054 0.133 -0.078 0.071 0.061 0.116 -0.055 0.036 0.086 0.123 -0.037 0.084 | TYP ADHD Difference TYP ADHD 0.143 0.182 -0.039 0.127 0.192 0.100 0.153 -0.053 0.066 0.125 0.098 0.140 -0.042 0.055 0.169 0.050 0.172 -0.122 0.085 0.119 0.076 0.179 -0.103 0.083 0.191 0.082 0.167 -0.085 0.061 0.201 0.054 0.155 -0.100 0.056 0.124 0.054 0.133 -0.078 0.071 0.103 0.061 0.116 -0.055 0.036 0.171 0.086 0.123 -0.037 0.084 0.144 | |

Results obtained including GSR showed high concordance when the analyses were repeated non including GSR, confirming significant rsFC differences in the two regions of interests (see Figure S4 and Table S5 in Supplementary Materials for Bcaud rsFC maps).

No statistically significant difference was found between medicated and non-medicated ADHD patients, nor when the sub-type of ADHD diagnosis was considered. An overview of the minimal FDR-adjusted p value associated with this control analyses can be found in the Supplementary Materials as Table S6.



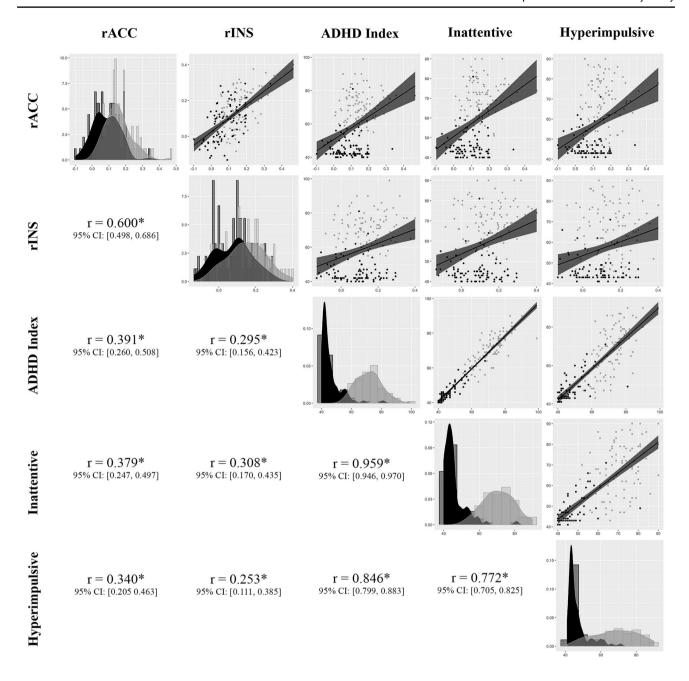


Fig. 3 Correlation matrices between fMRI findings and clinical scales. All the subjects (TYP and ADHD) are considered in the analysis. *p < 0.01 (Bonferroni corrected). ADHD patients are repre-

sented by the color black in the distribution curves and scatter plots. TYP patients are in light gray

Statistically significant differences between-groups of rsFC values were maintained when considering only males or only females, in all the four *t* tests performed. A detailed overview of the control analyses for sex can be found in the Supplementary Materials as Table S7, with a report of means, standard deviations and standard errors.

Taking rACC and rINS as seeds, higher rsFC observed in ADHD remained specific for the caudate, as no other region emerged from the voxel-wise analysis (see Figures S5 and S6 in the Supplementary Materials).

Discussion

The present study finds a significantly higher rsFC in ADHD subjects between Bcaud, rACC and rINS, which are part of the so-called salience network (SN). The rsFC evolutional pattern shows age-dependent dynamics and is mildly linked to core ADHD clinical constructs such as hyperactivity-impulsivity and inattention. Finally, when rsFC between caudate and mesolimbic areas of the SN is taken as a single



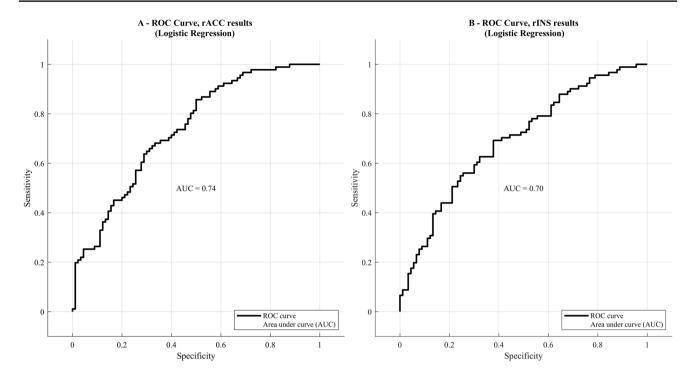


Fig. 4 AUC Area Under the Curve. Values above 0.700 are suggestive of a fair diagnostic discrimination between TYP and ADHD. **a** ROC curve for the logistic regression model built on rACC results. **b** ROC curve for the logistic regression model built on rINS results

parameter to differentiate ADHD from TYP, a fair diagnostic discrimination is reached.

Connectivity topography and ADHD functioning

The caudate nucleus shares an increased rsFC (Fig. 1) with rACC and rINS. These regions pertain to the SN, a well-studied functional network playing a pivotal role in attentional switches, internal-external integration, and tolerance to reward delays [44]. The possibility of a stimulus to emerge with respect to the other ones is directly connected with the attentional valence attributed to it by the brain throughout a phasic process [45]. Clinically, alterations in this process such as heightened phasic attention at the expenses of the tonic/sustained one reflect ADHD distractibility [46]. Neuronally, our study shows a positive correlation (i.e., increased interdependence) of the rsFC strength between the caudate and SN region with impulsivity and inattention scores. A strengthened bond between subcortical and mesolimbic salience regions may thus relate clinical and neurobiological findings to a fundamental alteration of salience processes in ADHD. More specifically, dACC and rINS are crucial regions for attentional switching such as the one between rest-oriented versus task-oriented modalities [47]. The rsFC differences we observed in both of them may thus be related to the attentional lapses and increased reaction-time variability that may be due to an impaired switching in ADHD [48].

The involvement of rINS in both attentive and emotional processes [49] highlights the deep intertwinement between salience and emotion: the appetitive-aversive (emotional) quality attributed to a stimulus is directly connected to its strength, i.e., its possibility to emerge from the "environmental noise" [50].

The evaluation of a stimulus' emotional quality appears to be quantitative and given by the level of its appetitive-aversive strength registered by deep structures of the brain such as caudate [51]. Other structures such as SN subregions are then recruited to actively bestow a refined qualitative and cognitive valence to the emotional content [52]. An increased interdependence between subcortical and limbic-cognitive areas may characterize ADHD as a disorder of emotional salience regulation. The hyper-connection between caudate and SN regions showed by our findings, coherently with existing literature, might allow us to consider ADHD as a disorder of emotional salience attribution.

Salience and brain maturation

We showed how caudate rsFC is characterized by an increased and reciprocal influence with the meso-limbic districts. Additionally, we characterized distinct progressions through the age in TYP vs ADHD subjects according to the areas that are considered. Furthermore, the between-groups rsFC shows the maximum difference



around the age of 10. After this peak, discrepancies between TYP and ADHD tend to regress, a process similar to the one observed in structural caudate related findings. Behaviorally, the rsFC in TYP vs ADHD may indicate: (1) a delay in brain maturation [53]; (2) an excessive interdependence between reward/emotional and salience/ attentional processes in ADHD, resulting in a reduced ability to involve cognition as a mediator for a more finegrained processing of information and decision making (which is instead characteristic of the adult neurotypical brain). Of note, our findings are in line with the ones from Szekely et al. [54] suggesting an age-dependent corticalization of the inhibition-related disfunctions in ADHD: while they are predominantly related to caudate's activity during childhood, this link is not evident in adults, where modulations of other areas such as vmPFC and precuneus are instead shown.

Reward and salience: two sides of a coin?

Caudate contributes to initiate and shape complex human behaviors which comprise not only emotion and attention [6] but also the evaluation of appropriate goal-directed behaviors, including motivation and cognition [55]. Reward and salience are in fact reciprocally modulating processes [56]. Cortico-striatal connections hence deeply relate constructs such as reward, emotion and salience [57], all of which show a certain degree of impairment in ADHD [58].

From fMRI findings further support this hypothesis: being caudate and insula neural determinants of human habitual action control and avoidance motivation [59], their activity shows major differences in ADHD and is related to the severity of impulsivity [60]. The high correspondence of salience and reward networks [61] in ADHD is coherent with the association of the caudate-SN hyper-connectivity to both inattentive and hyper-impulsive scores. While inattention and impulsivity pertain to two different neurobiological domains (salience and reward respectively), these domains are closely interacting through dopaminergic pathways involving especially in caudate, insula, and anterior cingulate [62, 63]. An increased rsFC may thus be a common neural phenomenon reflecting the expression of both symptoms.

Diagnostic prediction of rsFC scores

Both Bcaud/rACC and Bcaud/rINS rsFC were useful to discriminate ADHD and TYP subjects with a fair rate of success (AUC≥0.7). These results are promising as although they use a single parameter, they achieve similar predictive

scores when compared with more complex and integrated models [64, 65]. Such a measure is experimental and needs further validation, as a larger cohort with replication samples can increase the model precision or disconfirm it. A cohort including adults as well as adolescents may also help to differentiate the patterns related to each life stage, possibly helping to integrate the clinical observations throughout the diagnostic process.

Limitations

- (1) Even though the findings are solid, follow-ups of the same subject across different time-points should strongly improve the accuracy when measuring the developmental evolution in brain rsFC, possibly locating neuroimaging as a useful diagnostic tool for ADHD.
- (2) The increased caudate/salience rsFC does not explain per se the persistency of ADHD symptoms in adulthood, as the difference between TYP and ADHD subjects progressively reduces through age. Nevertheless, our functional findings are in line with the progressive reduction of the structural discrepancies observed for caudate in ADHD subjects.
- (3) Our findings are only partially consistent with previous studies such as Yang et al. [18], which firstly explored caudate FC. These differences may be due to the different age range of the sample studied and to methodological differences (in Yang et al. [18] age was not regressed as a possible factor influencing caudate rsFC, scrubbing of the fMRI signal was not performed and only dorsal caudate was considered, using one voxel masks).
- (4) The lack of effect of medication on the results, which contradicts most of the literature available, is most likely due to the fact that drugs were suspended at least 24 h before the scan was performed, and although medicated, most patients retained high levels of clinical severity.
- (5) rsFC is an effect determined by the combined action of factors not analyzed by this study, as they pertain to structural and biochemical domains going beyond the purpose of our work. Hence, our work does not allow to suppose causative effects of the subcortical functioning over the mesolimbic one or vice versa. Parallelly, the present findings encourage future research combining structural, functional and biochemical data to further qualify and quantify the contribution of subcortical structures to both ADHD symptomatology and the overall brain activity.



Conclusions

In our paper, we confirmed how in ADHD subcortical-mesolimbic rsFC is markedly increased and involves specific areas pertaining to the SN. Caudate, which structurally and functionally pertains to circuits involved in salience and reward, has specific areas of increased rsFC associated with the SN, confirming the involvement of key pathways theorized by previous authors. Therefore, our results allow to contextualize neuronal, biological, and behavioral findings into an enriched framework in line with ADHD literature adding new conceptual and clinical implications.

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Compliance with ethical standards

Conflict of interest The authors declare they have no conflict of interest.

References

- American Psychiatric Association (2013) Diagnostic and statistical manual of mental disorders: DSM-5, 5th edn. American Psychiatric Association, Washington, D.C
- Rubia K, Alegría AA, Brinson H (2014) Brain abnormalities in attention-deficit hyperactivity disorder: a review. Rev Neurol 58(Suppl 1):S3–S16
- Samea F, Soluki S, Nejati V et al (2019) Brain alterations in children/adolescents with ADHD revisited: a neuroimaging meta-analysis of 96 structural and functional studies. Neurosci Biobehav Rev 100:1–8. https://doi.org/10.1016/j.neubiorev.2019.02.011
- Qiu A, Crocetti D, Adler M et al (2009) Basal ganglia volume and shape in children with attention deficit hyperactivity disorder. Am J Psychiatry 166:74–82. https://doi.org/10.1176/appi. ajp.2008.08030426
- Hoogman M, Bralten J, Hibar DP et al (2017) Subcortical brain volume differences in participants with attention deficit hyperactivity disorder in children and adults: a cross-sectional megaanalysis. Lancet Psychiatry 4:310–319. https://doi.org/10.1016/ S2215-0366(17)30049-4
- Robinson JL, Laird AR, Glahn DC et al (2012) The functional connectivity of the human caudate: an application of meta-analytic connectivity modeling with behavioral filtering. Neuroimage 60:117–129. https://doi.org/10.1016/j.neuroimage.2011.12.010
- 7. Castellanos FX, Lee PP, Sharp W et al (2002) Developmental trajectories of brain volume abnormalities in children and

- adolescents with attention-deficit/hyperactivity disorder. JAMA 288:1740–1748. https://doi.org/10.1001/jama.288.14.1740
- Patros CHG, Alderson RM, Kasper LJ et al (2016) Choice-impulsivity in children and adolescents with attention-deficit/hyperactivity disorder (ADHD): a meta-analytic review. Clin Psychol Rev 43:162–174. https://doi.org/10.1016/j.cpr.2015.11.001
- Plessen KJ, Bansal R, Zhu H et al (2006) Hippocampus and amygdala morphology in attention-deficit/hyperactivity disorder. Arch Gen Psychiatry 63:795. https://doi.org/10.1001/archpsyc.63.7.795
- Nickel K, Tebartz van Elst L, Perlov E et al (2017) Manual morphometry of hippocampus and amygdala in adults with attention-deficit hyperactivity disorder. Psychiatry Res Neuroimaging 267:32–35. https://doi.org/10.1016/j.pscychresns.2017.07.001
- 11. Tajima-Pozo K, Ruiz-Manrique G, Yus M et al (2015) Correlation between amygdala volume and impulsivity in adults with attention-deficit hyperactivity disorder. Acta Neuropsychiatr 27:362–367. https://doi.org/10.1017/neu.2015.34
- Rapport MD, Alderson RM, Kofler MJ et al (2008) Working memory deficits in boys with attention-deficit/hyperactivity disorder (ADHD): the contribution of central executive and subsystem processes. J Abnorm Child Psychol 36:825–837. https://doi. org/10.1007/s10802-008-9215-y
- Hart H, Radua J, Nakao T et al (2013) Meta-analysis of functional magnetic resonance imaging studies of inhibition and attention in attention-deficit/hyperactivity disorder: exploring task-specific, stimulant medication, and age effects. JAMA Psychiatry 70:185– 198. https://doi.org/10.1001/jamapsychiatry.2013.277
- Huang Z, Obara N, Davis HH et al (2016) The temporal structure of resting-state brain activity in the medial prefrontal cortex predicts self-consciousness. Neuropsychologia 82:161–170. https:// doi.org/10.1016/j.neuropsychologia.2016.01.025
- Scalabrini A, Ebisch SJH, Huang Z et al (2019) Spontaneous brain activity predicts task-evoked activity during animate versus inanimate touch. Cereb Cortex. https://doi.org/10.1093/cercor/bhy340
- Damiani S, Scalabrini A, Gomez-Pilar J et al (2019) Increased scale-free dynamics in salience network in adult high-functioning autism. NeuroImage Clin 21:101634. https://doi.org/10.1016/j. nicl.2018.101634
- Scalabrini A, Huang Z, Mucci C et al (2017) How spontaneous brain activity and narcissistic features shape social interaction. Sci Rep 7:9986. https://doi.org/10.1038/s41598-017-10389-9
- Yang Z, Li H, Tu W et al (2018) Altered patterns of restingstate functional connectivity between the caudate and other brain regions in medication-naïve children with attention deficit hyperactivity disorder. Clin Imaging 47:47–51. https://doi. org/10.1016/j.clinimag.2017.07.009
- Rosch KS, Mostofsky SH, Nebel MB (2018) ADHD-related sex differences in fronto-subcortical intrinsic functional connectivity and associations with delay discounting. J Neurodev Disord 10:34. https://doi.org/10.1186/s11689-018-9254-9
- Hulvershorn LA, Mennes M, Castellanos FX et al (2014) Abnormal amygdala functional connectivity associated with emotional lability in children with attention-deficit/hyperactivity disorder. J Am Acad Child Adolesc Psychiatry 53:351–361.e1. https://doi.org/10.1016/j.jaac.2013.11.012
- Cao X, Cao Q, Long X et al (2009) Abnormal resting-state functional connectivity patterns of the putamen in medication-naïve children with attention deficit hyperactivity disorder. Brain Res 1303:195–206. https://doi.org/10.1016/j.brainres.2009.08.029
- Mills KL, Bathula D, Dias TGC et al (2012) Altered cortico-striatal-thalamic connectivity in relation to spatial working memory capacity in children with ADHD. Front Psychiatry 3:2. https://doi. org/10.3389/fpsyt.2012.00002
- Oldehinkel M, Beckmann CF, Franke B et al (2016) Functional connectivity in cortico-subcortical brain networks underlying reward processing in attention-deficit/



- hyperactivity disorder. Neuroimage Clin 12:796–805. https://doi.org/10.1016/j.nicl.2016.10.006
- Oldehinkel M, Beckmann CF, Pruim RHR et al (2016) Attention-deficit/hyperactivity disorder symptoms coincide with altered striatal connectivity. Biol Psychiatry Cogn Neurosci Neuroimaging 1:353–363. https://doi.org/10.1016/j. bpsc.2016.03.008
- Zhou Z-W, Sun L, Fang Y-T et al (2019) Inconsistency in abnormal functional connectivity across datasets of ADHD-200 in children with attention deficit hyperactivity disorder. Front Psychiatry 10:692. https://doi.org/10.3389/fpsyt.2019.00692
- Gabard-Durnam LJ, Flannery J, Goff B et al (2014) The development of human amygdala functional connectivity at rest from 4 to 23 years: a cross-sectional study. Neuroimage 95:193–207. https://doi.org/10.1016/j.neuroimage.2014.03.038
- Tang C, Wei Y, Zhao J, Nie J (2018) Different developmental pattern of brain activities in ADHD: a study of resting-state fMRI. Dev Neurosci 40:246–257. https://doi.org/10.1159/00049 0289
- 28. Barber AD, Sarpal DK, John M et al (2019) Age-normative pathways of striatal connectivity related to clinical symptoms in the general population. Biol Psychiat 85:966–976. https://doi.org/10.1016/j.biopsych.2019.01.024
- Neufeld J, Kuja-Halkola R, Mevel K et al (2018) Alterations in resting state connectivity along the autism trait continuum: a twin study. Mol Psychiatry 23:1659–1665. https://doi.org/10.1038/ mp.2017.160
- Caballero-Gaudes C, Reynolds RC (2017) Methods for cleaning the BOLD fMRI signal. NeuroImage 154:128–149. https://doi. org/10.1016/j.neuroimage.2016.12.018
- 31. Konstantareas MM, Hewitt T (2001) Autistic disorder and schizophrenia: diagnostic overlaps. J Autism Dev Disord 31:19–28
- Satterthwaite TD, Elliott MA, Gerraty RT et al (2013) An improved framework for confound regression and filtering for control of motion artifact in the preprocessing of resting-state functional connectivity data. NeuroImage 64:240–256. https://doi.org/10.1016/j.neuroimage.2012.08.052
- Kim J-Y, Kim S-H, Seo J et al (2013) Increased power spectral density in resting-state pain-related brain networks in fibromyalgia. PAIN® 154:1792–1797. https://doi.org/10.1016/j. pain.2013.05.040
- Giménez M, Guinea-Izquierdo A, Villalta-Gil V et al (2017) Brain alterations in low-frequency fluctuations across multiple bands in obsessive compulsive disorder. Brain Imaging Behav 11:1690–1706. https://doi.org/10.1007/s11682-016-9601-y
- Fox MD, Snyder AZ, Vincent JL et al (2005) The human brain is intrinsically organized into dynamic, anticorrelated functional networks. Proc Natl Acad Sci USA 102:9673–9678. https://doi. org/10.1073/pnas.0504136102
- Chai XJ, Castañón AN, Ongür D, Whitfield-Gabrieli S (2012) Anticorrelations in resting state networks without global signal regression. Neuroimage 59:1420–1428. https://doi.org/10.1016/j. neuroimage.2011.08.048
- Murphy K, Fox MD (2017) Towards a consensus regarding global signal regression for resting state functional connectivity MRI. NeuroImage 154:169–173. https://doi.org/10.1016/j.neuroimage .2016.11.052
- Afyouni S, Nichols TE (2018) Insight and inference for DVARS. NeuroImage 172:291–312. https://doi.org/10.1016/j.neuroimage .2017.12.098
- Power JD, Mitra A, Laumann TO et al (2014) Methods to detect, characterize, and remove motion artifact in resting state fMRI. NeuroImage 84:320–341. https://doi.org/10.1016/j.neuroimage .2013.08.048

- Thomason ME, Dennis EL, Joshi AA et al (2011) Resting-state fMRI can reliably map neural networks in children. NeuroImage 55:165–175. https://doi.org/10.1016/j.neuroimage.2010.11.080
- Conners CK, Sitarenios G, Parker JD, Epstein JN (1998) The revised Conners' Parent Rating Scale (CPRS-R): factor structure, reliability, and criterion validity. J Abnorm Child Psychol 26:257–268. https://doi.org/10.1023/a:1022602400621
- 42. Kriegeskorte N (2015) Crossvalidation in Brain Imaging Analysis. BioRxiv 017418. https://doi.org/10.1101/01741
- Liu TT (2016) Noise contributions to the fMRI signal: an overview. NeuroImage 143:141–151. https://doi.org/10.1016/j.neuroimage.2016.09.008
- Steimke R, Nomi JS, Calhoun VD et al (2017) Salience network dynamics underlying successful resistance of temptation. Soc Cogn Affect Neurosci 12:1928–1939. https://doi.org/10.1093/ scan/nsx123
- Asanowicz D, Marzecová A (2017) Differential effects of phasic and tonic alerting on the efficiency of executive attention. Acta Psychol 176:58–70. https://doi.org/10.1016/j.actps y.2017.03.004
- Badgaiyan RD, Sinha S, Sajjad M, Wack DS (2015) Attenuated tonic and enhanced phasic release of dopamine in attention deficit hyperactivity disorder. PLoS ONE 10:e0137326. https://doi. org/10.1371/journal.pone.0137326
- Sidlauskaite J, Sonuga-Barke E, Roeyers H, Wiersema JR (2016)
 Default mode network abnormalities during state switching in attention deficit hyperactivity disorder. Psychol Med 46:519–528. https://doi.org/10.1017/S0033291715002019
- Karalunas SL, Geurts HM, Konrad K et al (2014) Annual research review: reaction time variability in ADHD and autism spectrum disorders: measurement and mechanisms of a proposed transdiagnostic phenotype. J Child Psychol Psychiatry 55:685–710. https://doi.org/10.1111/jcpp.12217
- Uddin LQ, Nomi JS, Hébert-Seropian B et al (2017) Structure and function of the human insula. J Clin Neurophysiol 34:300–306. https://doi.org/10.1097/WNP.000000000000377
- Janak PH, Tye KM (2015) From circuits to behaviour in the amygdala. Nature 517:284–292. https://doi.org/10.1038/nature14188
- Carretié L, Ríos M, de la Gándara BS et al (2009) The striatum beyond reward: caudate responds intensely to unpleasant pictures. Neuroscience 164:1615–1622. https://doi.org/10.1016/j.neuro science.2009.09.031
- Phan KL, Taylor SF, Welsh RC et al (2004) Neural correlates of individual ratings of emotional salience: a trial-related fMRI study. NeuroImage 21:768–780. https://doi.org/10.1016/j.neuro image.2003.09.072
- Shaw P, Eckstrand K, Sharp W et al (2007) Attention-deficit/ hyperactivity disorder is characterized by a delay in cortical maturation. Proc Natl Acad Sci 104:19649–19654. https://doi. org/10.1073/pnas.0707741104
- Szekely E, Sudre GP, Sharp W et al (2017) Defining the neural substrate of the adult outcome of childhood ADHD: a multimodal neuroimaging study of response inhibition. Am J Psychiatry 174:867–876. https://doi.org/10.1176/appi.ajp.2017.16111313
- Grahn JA, Parkinson JA, Owen AM (2008) The cognitive functions of the caudate nucleus. Prog Neurobiol 86:141–155. https:// doi.org/10.1016/j.pneurobio.2008.09.004
- Gong M, Liu T (2018) Reward differentially interacts with physical salience in feature-based attention. J Vis 18:12. https://doi.org/10.1167/18.11.12
- 57. Soder HE, de Dios C, Potts GF (2016) The role of the neural reward system in attention selection. NeuroReport 27:787–790. https://doi.org/10.1097/WNR.0000000000000617
- Plichta MM, Vasic N, Wolf RC et al (2009) Neural hyporesponsiveness and hyperresponsiveness during immediate and delayed reward processing in adult attention-deficit/hyperactivity



- disorder. Biol Psychiatry 65:7–14. https://doi.org/10.1016/j.biopsych.2008.07.008
- Eryilmaz H, Rodriguez-Thompson A, Tanner AS et al (2017) Neural determinants of human goal-directed vs. habitual action control and their relation to trait motivation. Sci Rep 7:6002. https://doi.org/10.1038/s41598-017-06284-y
- Plichta MM, Scheres A (2014) Ventral-striatal responsiveness during reward anticipation in ADHD and its relation to trait impulsivity in the healthy population: a meta-analytic review of the fMRI literature. Neurosci Biobehav Rev 38:125–134. https:// doi.org/10.1016/j.neubiorev.2013.07.012
- von Rhein D, Beckmann CF, Franke B et al (2017) Network-level assessment of reward-related activation in patients with ADHD and healthy individuals: network-level assessment of rewardrelated activation. Hum Brain Mapp 38:2359–2369. https://doi. org/10.1002/hbm.23522
- 62. Fusar-Poli P, Rubia K, Rossi G et al (2012) Striatal dopamine transporter alterations in ADHD: pathophysiology or adaptation to psychostimulants? A meta-analysis. Am J Psychiatry 169:264–272. https://doi.org/10.1176/appi.ajp.2011.11060940
- Conio B, Martino M, Magioncalda P et al (2020) Opposite effects of dopamine and serotonin on resting-state networks: review and implications for psychiatric disorders. Mol Psychiatry 25:82–93. https://doi.org/10.1038/s41380-019-0406-4
- 64. Brown MRG, Sidhu GS, Greiner R et al (2012) ADHD-200 global competition: diagnosing ADHD using personal characteristic data can outperform resting state fMRI measurements. Front Syst Neurosci 6:69. https://doi.org/10.3389/fnsys.2012.00069
- 65. Riaz A, Asad M, Alonso E, Slabaugh G (2018) Fusion of fMRI and non-imaging data for ADHD classification. Comput Med Imaging Graph 65:115–128. https://doi.org/10.1016/j.compmedimag.2017.10.002

