Aggression Subtypes Relate to Distinct Resting State Functional Connectivity in Children and Adolescents with Disruptive Behavior

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ABSTRACT

Objective: There is increasing evidence to suggest altered resting state functional connectivity (rsFC) in adolescents with conduct problems. However, no rsFC studies have addressed the effects of reactive and proactive aggression. Herein, we examined the associations between these aggression subtypes along with subdimensions of callous-unemotional (CU) traits and rsFC using predefined seeds in aggression-related salience network (SN) and default mode network (DMN).

Method: Aggression subtype-specific whole-brain rsFC of SN and DMN seeds was investigated in a resting state sequence (mean acquisition time = 8 min 25 sec) acquired from 207 children and adolescents of both sexes aged 8 - 18 years (mean age (SD) = 13.30 (2.60) years; range = 8.02 – 18.35) in a multi-center study. One hundred eighteen individuals exhibited disruptive behavior (conduct disorder/oppositional defiant disorder) with different levels of comorbid ADHD symptoms, 89 were healthy.

Results: Compared to healthy controls, cases demonstrated reduced DMN and – after controlling for ADHD scores – SN seed-based rsFC with left hemispheric frontal clusters. We found increased and distinct aggression-subtype specific rsFC patterns in brain regions linked to processes like emotion, empathy, moral, cognitive control, and decision-making. Specifically, reactive and proactive aggression correlated with distinct SN and DMN seed-based rsFC patterns. CU dimensions led to different DMN and SN rsFC with clusters including frontal, parietal, and cingulate areas, with uncaring-related clusters extended to cerebellar regions.

Conclusions: This first study investigating reactive and proactive aggression along with CU dimensions reveals new subtype-specific whole-brain rsFC patterns, extending the knowledge of neural networks to further distinct forms of disruptive behavior.

INTRODUCTION

Oppositional defiant disorder (ODD) and conduct disorder (CD) are among the most common psychiatric disorders in childhood and adolescence [1]. ODD is characterized by angry, argumentative, and vindictive behaviors, CD indicates a pattern of violating rules, norms, and rights [2]. Comorbid symptoms of attention-deficit/hyperactivity disorder (ADHD) are present in 36% of boys and 57% of girls with disruptive behavior [3] with overlapping neural deficits in prefrontal and limbic areas [4]. Callous-unemotional (CU) traits are reported in 25%-30% of children with an early onset of CD symptoms [5]. On a behavioral level, there is evidence that CU traits are associated with impaired emotion recognition skills, particularly for fear[6]. Recent studies point to the importance of differentiating distinct CU dimensions [7,8]. Additionally, reactive and proactive aggression [9] can characterize disruptive behavior. Reactive aggression is described as impulsive, "hot-tempered" behavior in response to perceived provocation or threat, while proactive aggression is characterized by instrumental and sometimes manipulating behavior. Despite behavioral correlations between reactive and proactive aggression[10], there might be differences. For instance, reactive aggression correlated positively with impulsivity, while proactive aggressive behavior has been related to high CU traits[11].

Recent studies of resting state fMRI (rs-fMRI) in adolescents with CD have mainly reported reduced resting state functional connectivity (rsFC) or activity. Affected brain regions include the amygdala and insula as parts of the salience network (SN) [12–14], also implicated in emotion-related processes[15], and areas of the default mode network (DMN) [16–20]. The anterior medial prefrontal cortex (amPFC) and the posterior cingulate cortex (PCC), as the core regions of the DMN, were found to be associated with (affective) self-referential processes [21]. Connectivity between those DMN regions was reduced in male adolescents with CD compared to healthy controls after controlling for ADHD symptoms, as ADHD symptoms correlated positively with DMN rsFC [18]. Other recent studies have also reported reduced rsFC in male

adolescents with CD compared to healthy controls within DMN regions [17], along with other resting state networks (RSNs) [16]. Furthermore, neural activity during rest in SN [14] and DMN areas was reduced. Higher ADHD scores, however, related to increased rsFC density [19] and alterations [20,22,23] in DMN areas. In sum, rs-fMRI studies have mainly reported reduced connectivity and activity particularly in DMN and SN regions in youths with disruptive behavior. ADHD symptoms in those youths were linked to increases in such regions. Critically, the existing studies were mostly limited to male adolescents with CD, whereas ODD, ADHD, CU traits, and reactive and proactive aggression were largely neglected.

To date, five rsFC studies have evaluated CU subdimensions in children and adolescents. While the anticorrelation between the DMN and fronto-parietal network decreased with higher CU-related traits [23], anterior DMN rsFC increased with higher scores on the CU dimension [24], and DMN rs-fMRI parameters increased with higher CU-related scores in incarcerated adolescents (mean age 17 years) [25]. In male youths with CD, CU-related traits correlated with distinct amygdala subregional rsFC with clusters including SN and DMN regions [12]. Compared to male youths with CD and lower CU traits or healthy controls, juveniles with CD and higher CU scores showed increased amygdala subregion rsFC with a cluster including frontal and DMN regions [13]. Psychopathic adults demonstrated reduced rsFC between dorsal parts of the DMN [26] and increased and diminished rsFC in three RSNs including anterior insula and PCC depending on distinct CU-related traits [27]. In a large sample of incarcerated adults, the interpersonal CU-related dimension correlated with rsFC in the DMN, executive control network (ECN), and SN along with paralimbic regions [28]. Other recent studies have reported reduced [29] and increased [30] DMN rsFC in addition to rsFC alterations in the central gyrus in adults [27,28,30,31] and also in adolescents [25]. Taken together, there is growing evidence for CU dimension-specific rsFC predominantly increasing in brain regions including SN and DMN areas. Regarding CU dimensions, all but one fMRI resting state study [24] restricted analyses to

males. However, healthy controls were not included, children were underrepresented (mean age 17.8 years), and further aggression dimensions were not considered. To our knowledge, not a single rsFC analysis has addressed the role of reactive and proactive aggression in children and adolescents with disruptive behavior to date. Literature on task-based fMRI suggests that amygdala functioning is negatively related to proactive aggression [32] and positively to reactive aggression [33]. Insula reactivity is mostly positively related to reactive aggression [34].

This is the first study of fMRI rsFC in boys and girls with disruptive behavior including diagnoses of CD and ODD that takes CU dimensions and reactive and proactive aggression into account. Based on the findings of altered rsFC of the DMN [16–19], SN [12,13], and other RSNs [16] in youth with CD, we defined regions of interest (ROIs) in these brain areas and expected to find reduced rsFC in cases compared to healthy controls. We applied a seed-based approach, frequently used for analyzing rsFC [12,13,22,29]. As reactive and proactive aggression have not been addressed yet, we analysed seed-based rsFC patterns of amygdala and anterior insula[32–34]. Further, we expected to find distinct rsFC patterns for different CU dimensions [12,13,24] and increased rsFC with higher ADHD scores [18,19].

METHOD

Participants

Participants in the current study were part of the joint EU-MATRICS and EU-Aggressotype project. Children and adolescents aged 8-18 years with and without aggressive behavior were recruited from resident hospitals, ambulatories, and eligible (boarding) schools. A total of 207 (n = 150 males) cases (n = 118) and healthy controls (n = 89) were included from nine different sites in Europe (mean age = 13.30 ± 2.60 years). As the main goal was to conduct aggression subtype-specific analyses, recruitment focused on including cases presenting with a diagnosis of CD and/or ODD and/or aggression scores in a clinical range (T > 70) according to the Child Behavior Checklist (CBCL), Youth Self Report (YSR), or Teacher Report Form (TRF) [35]. Controls were not allowed to have clinical aggression scores (T > 70) or a DSM-diagnosis, cases had to be without other main diagnoses. Further exclusion criteria for all participants were contra-indications for MRI scanning (i.e., braces, metal parts) and insufficient intellectual and cognitive functioning as further described in the Supplement. All participants gave written informed consent. Each site obtained ethical approval separately.

Clinical Assessments

The semi-structured interview Kiddie-Schedule for Affective Disorders and Schizophrenia, present and lifetime version (K-SADS-PL) [36] was used to assess diagnostic criteria for all participants by trained psychologists or interns based on the reports of participants and their parents were interviewed separately. In order to gain further diagnostic and aggression-related information, participants, their parents and teachers completed the YSR, CBCL, and TRF [35], respectively. The self-reported Reactive Proactive Aggression Questionnaire [37] measured reactive and proactive forms of aggression. To assess CU traits, parents filled out the Inventory

of Callous-Unemotional traits (ICU) [7,38] consisting of three subscales assessing callousness, uncaring, and unemotional behaviors. ADHD symptoms were evaluated using the inattention, hyperactivity, and impulsivity counts of the K-SADS-PL [36]. As part of a larger study protocol, further behavioral, neuropsychological, and neurophysiological tests and MRI sequences were conducted with details provided in the Supplemental Information.

Image Acquisition and Preprocessing

For data acquisition, six sites used Siemens 3 Tesla (T) scanners, two sites Philips 3T scanners, and one site a GE 3T scanner (see Supplemental Table S1 and Table S2). T1-weighted anatomical scans with largely similar parameters across sites (see Supplemental Table S1) were used to include white matter (WM) and cerebrospinal fluid (CSF) parameters as counfound regressors during temporal preprocessing. T2*-weighted echo-planar imaging resting state functional imaging was performed with predominantly similar parameters across sites (TR 2.45s or less, at least 32 slices; see Supplemental Table S2). During an average acquisition time of 8 min 25 sec, participants were instructed to lie still, look at a white cross-hair presented against a black background, and let their mind wander by not thinking about anything in particular. Standard preprocessing steps were applied using SPM12 (Welcome Trust Centre for Neuroimaging, UCL, United Kingdom; http://www.fil.ion.ucl.ac.uk/spm) and the SPM-based CONN toolbox v17.b (http://www.nitrc.org/projects/conn). Ten cases were excluded due to excessive motion (> 0.94 mm), twelve participants due to missing or insufficient quality of the structural scans, and 14 individuals based on image artifacts. More information on the preprocessing steps is described in the Supplement.

Regions of Interest

Using MarsBar toolbox (v0.44) (http://marsbar.sourceforge.net), PCC and amPFC as main cores of the DMN were centered on coordinates recently used [18] provided by Andrews-Hanna et al. [21]. Additionally, bilateral amygdala and bilateral anterior insula ROIs as part of the SN were derived from the Broadman-atlas implemented in the WFU Pickatlas (http://fmri.wfubmc.edu/software/pickatlas).

Functional Connectivity Analysis

Seed-based rsFC analyses were performed using CONN toolbox. First-level analysis computed Pearson´s correlation coefficients between the time course of previously denoised BOLD-signals from a seed and whole-brain voxel clusters. After Fisher's transformation to normally distributed z-scores, general linear model (GLM) analyses were computed. Second-level analysis included random-effects analysis of covariance (ANCOVA) for group comparisons, with further analyses including ADHD symptoms as additional covariates of no interest [18,22]. Linear regressions separately tested the association between reactive and proactive aggression and CU total score and dimensions on rsFC within cases. Site was added as dummy-coded covariate of no interest in second-level analyses. Results of the seed-based analyses are reported at a statistical threshold of $p < .001$, p-FWE $< .008$ cluster-level corrected (=0.05/6, using additional Bonferroni corrections for number of seeds). For post hoc analyses, we additionally controlled for age, sex, IQ, medication, and handedness given previous reports on possible influences for instance on rsFC of RSNs, such as the DMN[39]. Results of rsFC analysis were correlated with the behavioral measures using SPSS Edition 24 (IBM, SPSS, Inc.).

RESULTS

Sample Characteristics

Out of 118 cases, 48 had a diagnosis of ODD, 25 of CD plus ODD, and seven of CD. Seventy-seven cases (additionally) presented with a clinically relevant score (T > 70) on aggression or rule-breaking behavior subscales of the CBCL, and 41 cases on both subscales. Twenty-nine cases exhibited comorbid ADHD and 70 cases received medication (Table 1). Cases consisted of more males than females and exhibited a lower IQ than healthy controls. They also showed a wide distribution of CU traits, reactive and proactive aggression levels, and ADHD symptoms. For the distribution of diagnoses, aggression scores, medication, and demographic variables across sites, see Supplemental Table S3.

Table 1. Sample Characteristics (based on the group for the current resting state analysis)

Characteristic	Cases (n = 118)	HC (n = 89)	Test statistic	p-value
Age (Years)	13.23 ± 2.68	13.40 ± 2.49	$t(205) = -0.45$	0.65
Sex, m/f	99/19	51/38	$X^2 = 17.98$	< 0.001
IQ[a]	100.78 ± 11.00	106.64 ± 10.42	$t(195) = -3.81$	< 0.001
Handedness, left/right	16/95	10/77	$X^2 = 0.37$	0.55
CD plus ODD Diagnosis[b]	25			
ODD Diagnosis[b]	48			
CD Diagnosis[b]	7			
ADHD Diagnosis[b]	29			
CBCL – Aggression T-score	74.46 ± 10.10			
CBCL – Rule-breaking T-score	69.00 ± 12.14			
K-SADS - Inattention	3.33 ± 2.91			
K-SADS - Hyperactivity	1.66 ± 1.91			
K-SADS - Impulsivity	1.08 ± 1.20			
ICU – Total Score	33.68 ± 10.16			
ICU – Callousness	12.00 ± 6.11			
ICU – Uncaring	17.00 ± 3.93			
ICU – Unemotional	7.17 ± 3.31			
RPQ – Reactive Aggression	12.55 ± 5.09			
RPQ – Proactive Aggression	3.00 ± 5.01			
Medication use[c]	70			
- Stimulants	52			
- Neuroleptics	18			
- Antidepressants	2			
Mean RMS-FD	0.12 ± 0.17	0.09 ± 0.18	$U = 4134.50$	<0.01

Values are means or in case of non-normal distribution medians ± SD, or counts.
HC, healthy controls; ADHD, attention-deficit hyperactivity disorder; CD, conduct disorder; ODD, oppositional defiant disorder; K-SADS, Kiddie Schedule for Affective Disorders and Schizophrenia; ICU, Inventory of Callous-Unemotional traits, parent report; RPQ, Reactive and Proactive aggression Questionnaire; CBCL; Child Behavior Checklist; RMS-FD, root mean square framewise displacement.
[a]IQ estimated according to four sub-tests derived from the Wechsler Intelligence Scale for Children IV.
[b]Diagnoses derived from the Kiddie-Schedule for Affective Disorders and Schizophrenia, present and lifetime version.
[c]Medication use according to parental or clinicial report.

Group differences in functional connectivity

Cases demonstrated a reduced DMN (PCC seed) connectivity with a cluster including left frontal pole ($t(197) = 5.46$, cluster-size p-FWE < .008, peak p-uncorrected < .00001, $\beta = .10$) (Figure 1, Supplemental Table S4). This pattern only survived post hoc control for site, age, sex, IQ at a less conservative significance threshold (cluster-size p-FWE < .05). After taking ADHD symptoms into account, cases showed a diminished left hemispheric rsFC of SN (anterior insula seed) connectivity with a cluster extending from orbitofrontal cortex (OFC) to frontal pole compared to healthy controls ($t(194) = 5.07$, cluster-size p-FWE < .008, peak p-uncorrected < .00001, $\beta = .10$) (Figure 1, Supplemental Table S4). This group difference survived post hoc correction for site, age, sex, IQ, and handedness, but disappeared when medication was added as a covariate. Subsequent analysis within cases revealed a positive correlation of ADHD inattention and hyperactivity counts and SN (left anterior insula seed) rsFC, however at a lower significance threshold ($t(85) > 5.27$, all cluster-size p-FDR < .05, peak p-uncorrected < .00001, $\beta = .04$-$.09$) (Figure 1).

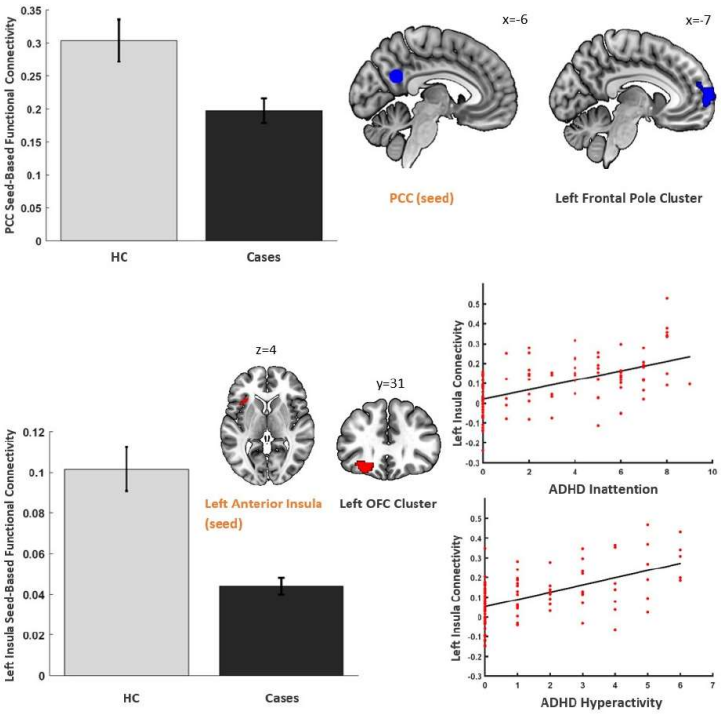

Figure 1. The bar charts on the left show reduced seed-based functional connectivity patterns for cases compared to healthy control (HC) subjects (see statistics in Table S4). DMN seed posterior cingulate cortex (PCC) connectivity with a cluster including left frontal pole and left superior frontal gyrus is corrected for site. The connectivity pattern for SN seed left anterior insula with a cluster including left orbitofrontal cortex (OFC) is corrected for site and ADHD scores. The statistical threshold for the reported results is $p < 0.001$, FWE cluster-level corrected ($p < 0.008 = 0.05/6$ using additional Bonferroni correction for number of seeds) for multiple comparisons. The scatterplots depict the main effect of ADHD inattention and hyperactivity counts within cases on SN seed left anterior insula seed-based connectivity with a cluster in left OFC and left frontal pole, post hoc corrected for site, age, sex, IQ, medication, and handedness ($p < 0.001$, FDR cluster-level corrected $p < 0.05$).

Functional connectivity correlates of reactive versus proactive aggression

Reactive and proactive aggression yielded distinct and increasing rsFC patterns for DMN and SN seeds within cases (Figure 2, Supplemental Table S6). After correction for site, age, sex, IQ, medication, and handedness, proactive aggression scores related to SN (left amygdala seed) connectivity with a cluster including the precuneus and PCC. In contrast, cases with higher reactive aggression demonstrated increased DMN (PCC seed) connectivity with a cluster extending from left parahippocampal gyrus to left inferior temporal gyrus. Furthermore, reactive aggression scores correlated with SN (left amygdala seed) rsFC with a cluster extending from left lateral occipital cortex to the precuneus. Reactive aggression scores also related to SN (right anterior insula seed) rsFC with a cluster in the right caudate nucleus (all cluster-size p-FWE < .008, β = .04-.05).

Functional connectivity correlates of specific CU dimensions

The different CU dimensions surviving post hoc correction for site, age, sex, IQ, medication, and handedness were positively associated with distinct seed-based rsFC patterns within cases (Figure 3, Supplemental Table S7). Callousness-related DMN seed-based connectivity patterns included clusters in the right hemispheric precentral gyrus. Clusters correlating with amPFC seed extended to the parietal and occipital areas and the cluster for PCC seed further included cingulate regions. Uncaring and unemotional behavior scores related to SN (left anterior insula seed) rsFC with precuneal and cingulate clusters. However, uncaring-related clusters extended to right hemispheric central gyrus, while unemotional-related clusters expanded to left hemispheric occipital and parietal areas, such as the angular gyrus. Uncaring behavior scores also related to SN (right anterior insula seed) rsFC with a cluster in the left central gyrus and to

DMN (amPFC seed) rsFC with right hemispheric cerebellar regions (all cluster-size p-FWE < .008, β = -.06-.08) (Figure 3, Supplemental Table S7).

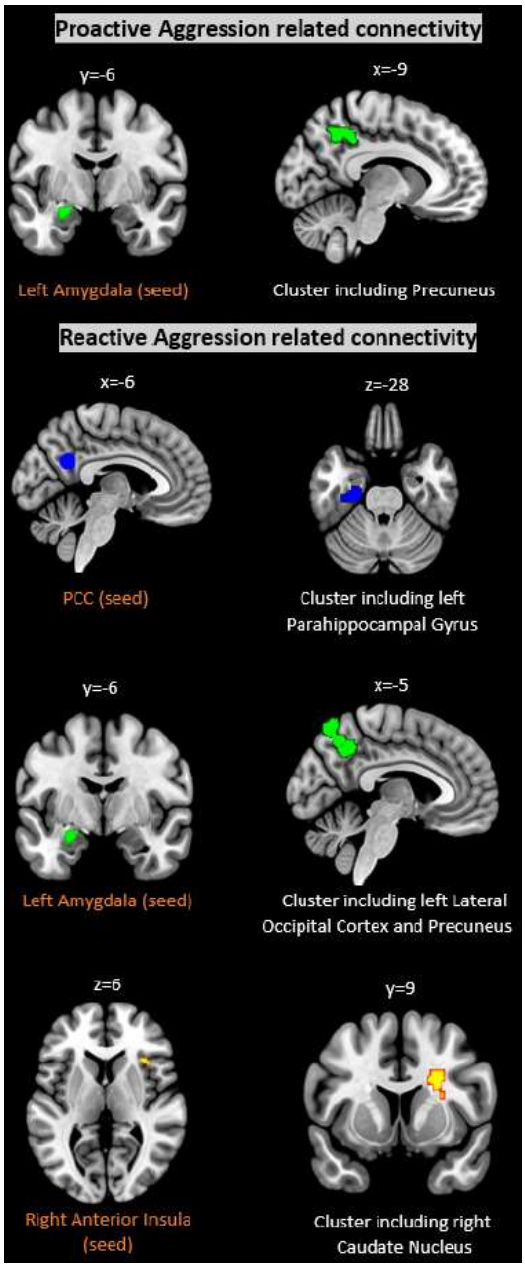

Figure 2. Higher proactive aggression scores related to increased salience network (SN) (left amygdala seed) connectivity with a cluster including precuneus and posterior cingulate gyrus (PCC). Higher reactive aggression scores related to increased default mode network (DMN) (PCC seed) connectivity with a left hemispheric cluster extending from parahippocampal gyrus to inferior temporal gyrus. Moreover, with higher reactive scores, SN (left amygdala seed) connectivity with a voxel cluster extending from left lateral occipital cortex to precuneus increased along with SN (right anterior insula seed) connectivity with a voxel cluster in right caudate nucleus.

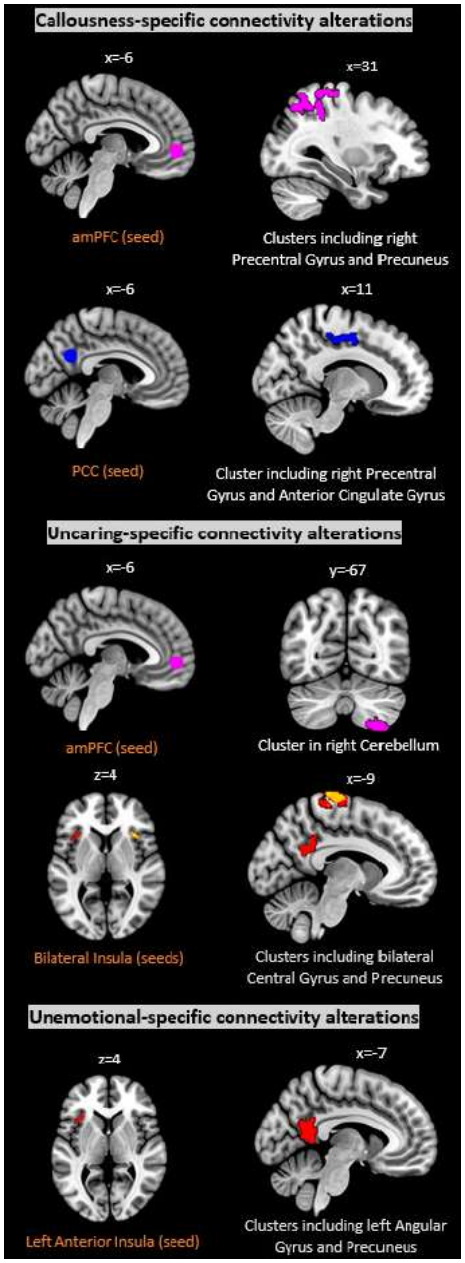

Figure 3. With higher callousness scores, default mode network (DMN) (anterior medial prefrontal cortex (amPFC) and posterior cingulate cortex (PCC) seed) connectivity with clusters including precentral gyrus increased and extended to parietal, occipital, and cingulate areas, respectively. Uncaring scores correlated positively with DMN (amPFC seed) connectivity with cerebellar regions and with salience network (SN) (bilateral anterior insula seeds) connectivity with frontal and parietal regions including bilateral central gyrus. With higher unemotional scores, SN (left anterior insula seed) connectivity with occipital, cingulate, and parietal areas including the precuneus increased.

DISCUSSION

The present multi-center study investigated aggression subtype-specific rsFC in a sizeable sample of children and adolescents ($n > 200$, mean age < 14 years, including both genders) with disruptive behavior including diagnoses of CD and ODD. Cases exhibited diminished DMN (PCC seed) rsFC with a frontal cluster including the left frontal pole compared to healthy controls. The additional control for ADHD symptoms further identified decreased SN (left anterior insula seed) rsFC with a left hemispheric cluster extending from the OFC to the frontal pole. We found distinct and increasing aggression subtype-specific rsFC patterns within cases in brain regions linked to processes like emotion, empathy, and decision-making. Proactive aggression scores related to SN connectivity with a cluster including the precuneus, and reactive aggression scores related to distinct DMN and SN seed-based rsFC patterns with (para-)limbic and precuneal clusters. Callousness and uncaring behavior scores related to different DMN and SN rsFC with voxel clusters including frontal, parietal, cingulate, precuneal, and cerebellar areas. Both uncaring and unemotional behavior scores correlated with SN rsFC with precuneal and cingulate clusters, which extended to distinct brain areas including the central and angular gyrus, respectively.

Abnormal rsFC patterns for the PCC have been demonstrated in male adolescents with CD [17–19] and may suggest impaired self-referential processes [21]. As observed group differences only survived post hoc analysis when other covariates were not controlled for or a lower threshold was applied, cautious interpretation is advised. Our results support the crucial role of comorbid ADHD symptoms previously shown in male adolescents with CD within the DMN [18] and extend analysis to DMN and SN seed-based rsFC with whole-brain voxel clusters. Diminished SN (left anterior insula seed) rsFC with left hemispheric clusters extending from the OFC to the frontal pole in cases compared to controls were only demonstrated when considering ADHD scores. In line with previous rs-fMRI studies [18,19,23], cases with higher

ADHD scores exhibited an increase in rsFC. This result further corroborates the recent finding of overlapping deficit functioning of prefrontal regions like the OFC and limbic areas including insular cortices in ADHD and disruptive behavior disorders [4].

To our knowledge, the present study is the first to address reactive and proactive aggression in rsFC of children and adolescents with disruptive behavior. In line with our hypothesis, reactive and proactive aggression related to distinct SN and DMN seed-based rsFC patterns in brain areas implicated in emotion, empathy, and cognitive control. Reactive scores correlated with increased rsFC within paralimbic and limbic regions, shown to function abnormally in psychopathy[28]. This is partly in line with the previously reported behavioral overlap of reactive and proactive aggression with CU-related traits [40]. The cluster involved in SN (right anterior insula seed) rsFC included the right caudate nucleus, a region implicated in integration of performance and cognitive control [41] and morality [42]. Neural circuits including the anterior insula have been linked to responses to frustration and perceived social provocations [33]. Moreover, both aggression subtypes related to increased SN (left amygdala seed) rsFC with a cluster including the precuneus. The amygdala, when impaired, seems involved in both a neural threat circuitry, related to a higher risk for reactive aggression and in moral behavior increasing the risk for proactive aggression [43]. Pathological aggression has been related to hyperactivity in neural circuits such as the amygdala, implicated in emotion generation, and to hypoactivity of cognitive control-related neural circuits including the precuneus [34]. Increased amygdala activity has been implicated in threat responses [34,44] which in turn could trigger reactive aggression [33]. Further, rsFC of the precuneus related to impulsivity in a recent study [19], which is relevant given the association of impulsivity and reactive aggression [11]. However, the cluster implicated in SN rsFC extended to distinct paralimbic and occipital areas for reactive and proactive aggression, thereby underpinning their differentiation and corroborating findings at the behavioral level [11].

The reported different rsFC patterns for the distinct CU dimensions support their differentiation in line with previous neural [12,13,24,27,28] and behavioral [8] findings. Affected brain areas extended from (para-)limbic regions implicated in adult psychopathy[28] to frontal, parietal, and cingulate areas. These regions have been linked to emotion, empathy, moral, decision-making and self-referential processes, and are impaired in youth with disruptive behaviors [44]. In line with rsFC studies in adolescents [12,13,24] and adults [27,30], cases exhibited increases in rsFC with higher CU total and dimension scores. Callousness and uncaring behavior-related rsFC patterns included clusters in the precentral gyrus. This region has been previously linked to altered rsFC in adolescents with CU-related traits [23,24] and psychopathic adults [27,28,30,31]. Precentral gyrus functioning was related to reward-related processes in adolescents and young adults with a wide distribution of CU scores [45]. A recent meta-analytic review found that the right anterior insula was implicated in the evaluation of feelings and the left anterior insula in the expression of anger [46]. Moreover, altered anterior insula functioning has been linked to abnormal empathic processes in children with higher callousness scores [47], and impaired decision making problems that may in turn increase frustration and subsequent antisocial behavior [44]. The DMN seed-based rsFC patterns reported here might suggest uncaring and callousness dimension-specific alterations in (affective) self-referential processes [21]. Altered rsFC in the DMN was reported in children and adolescents with CU-related traits [23,24] and in adult psychopathy [29,30,48]. For uncaring and unemotional behavior scores, the overlapping SN rsFC with a cluster in precuneal and cingulate areas may reflect their behavioral correlation [38]. The precuneus as part of the DMN contributed to classifying adults with antisocial personality disorder [48]. Unemotional behavior scores further related to SN rsFC with a cluster including left angular gyrus. This area seems to be implicated in moral processing [42], which has also been related to altered insular and cingulate functioning in adult psychopathy [49]. Interestingly, we did not find overlapping rsFC between CU dimensions and reactive or proactive aggression,

despite well-known commonalities on a behavioral level [40]. Despite the involvement of the amygdala in altered rsFC patterns [12,13] of adolescents with CU-related traits, no rsFC pattern included the amygdala. Our CU dimension-specific results may provide more support for recent findings that suggest neural alterations in broader brain areas beyond (para-)limbic regions in adult psychopathy [49].

There are some limitations in the present study. Firstly, including 38 cases with clinically relevant scores (T > 70) without a DSM-diagnosis of CD and/or ODD lowered the severity of aggression subtype-specific scores except for CU total and subdimension scores, which were comparable for cases with and without a diagnosis. However, subsequent sensitivity analyses led to comparable results of case-control group comparisons in rsFC (see Supplement). Secondly, we used parent reported CU traits and did not consider self-reports. Yet, a recent study reported a higher criterion validity for parent reported ICU compared to self- and teacher reports [50]. Third, data for the current study was collected at different sites with varying scanner manufacturers and partly deviating scan parameters, which affected data homogeneity and limited our study power. However, the larger sample size enabled by the multi-center design compensated for this and is likely to have increased the reliability and generalizability of our results.

Taken together, in the present study children and adolescents with disruptive behavior exhibited decreased rsFC of the DMN (PCC seed) and also of the SN (left anterior insula seed) with left hemispheric frontal voxel clusters, when ADHD symptoms were controlled for. More importantly, cases demonstrated an aggression subtype-specific increase in DMN and SN seed-based whole-brain rsFC with clusters beyond (para-)limbic areas in frontal, parietal, and cingulate regions related to processes like emotion, empathy, and decision-making. By evaluating the effect of reactive and proactive aggression along with CU dimensions, we have extended previous research in mainly male adolescents with CD and limited exploration of

distinct manifestations and subtypes of aggression. Our aggression subtype-specific findings provide support for a more precise diagnostic specification of aggression-related disorders. The present results also point to specific neural correlates of aggression subtypes, which may lend themselves to a tailored application of medication and treatment of criminal behavior. Particularly treatment of children and adolescents with disruptive behavior may be improved through careful exploration of distinct aggression subtypes and a better understanding of neural correlates. Further, our results may indicate developmental trajectories with some of the observed brain areas relating to adult psychopathy. In order to deepen the knowledge on neural correlates of aggression subtypes, future studies could extend the present a priori defined seed-based rsFC findings by investigating hypothesis-free whole-brain aggression dimension-specific rsFC alterations and relate these connectivity alterations to those during task performance.

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