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The effects of callous-unemotional traits and aggression subtypes on amygdala activity in response to negative faces

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Abstract

Background. Brain imaging studies have shown altered amygdala activity during emotion processing in children and adolescents with oppositional defiant disorder (ODD) and conduct disorder (CD) compared to typically developing children and adolescents (TD). Here we aimed to assess whether aggression-related subtypes (reactive and proactive aggression) and callous-unemotional (CU) traits predicted variation in amygdala activity and skin conductance (SC) response during emotion processing.

Methods. We included 177 participants ($n = 108$ cases with disruptive behaviour and/or ODD/CD and $n = 69$ TD), aged 8–18 years, across nine sites in Europe, as part of the EU Aggressotype and MATRICS projects. All participants performed an emotional face-matching functional magnetic resonance imaging task.

Results. Differences between cases and TD in affective processing, as well as specificity of activation patterns for aggression subtypes and CU traits, were assessed. Simultaneous SC recordings were acquired in a subsample ($n = 63$). Cases compared to TDs showed higher amygdala activity in response to negative faces (fearful and angry) *v.* shapes. Subtyping cases according to aggression-related subtypes did not significantly influence on amygdala activity; while stratification based on CU traits was more sensitive and revealed decreased amygdala activity in the high CU group. SC responses were significantly lower in cases and negatively correlated with CU traits, reactive and proactive aggression.

Conclusions. Our results showed differences in amygdala activity and SC responses to emotional faces between cases with ODD/CD and TD, while CU traits moderate both central (amygdala) and peripheral (SC) responses. Our insights regarding subtypes and trait-specific aggression could be used for improved diagnostics and personalized treatment.

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Introduction

Oppositional defiant disorder (ODD) and conduct disorder (CD) with a prevalence rate ranging from 2 to 4% (Polanczyk, Salum, Sugaya, Caye, & Rohde, 2015) are among the most commonly diagnosed mental health disorders in youth (Loeber, Burke, Lahey, Winters, & Zera, 2000). ODD is characterized by a frequent and persistent pattern of irritable and angry mood, vindictiveness and inappropriate, negativistic, defiant, and disobedient behavior toward authorities, while CD is defined as a repetitive and persistent pattern of behavior, which violates the rights of others and major age-appropriate societal rules (American Psychiatric Association, 2013). Additionally, the clinical representation of ODD/CD is heterogeneous, with distinct subtypes of aggression and high comorbidity rates with attention-deficit hyperactivity disorder (ADHD). Moreover, current research suggests that callous-unemotional (CU) traits, which include reduced guilt, callousness, uncaring behavior, and reduced empathy, contribute to this heterogeneity (Blair, Leibenluft, & Pine, 2014; Frick & Viding, 2009). On this basis, CU traits have been added to the fifth edition of the DSM (DSM-5) as a specifier for the diagnosis of CD called 'limited prosocial emotions'. Additionally, two distinctions in reactive (RA) and proactive (PA) aggression are often made to subtype aggressive behavior (Raine et al., 2006). RA is associated with impulsive, high arousal or affective aggression, whereas PA refers to goal-directed, planned behavior associated with reduced arousal and higher levels of CU traits (HCU) (Blair et al., 2014).

Recent brain imaging findings have provided insights into the underlying neural mechanisms of these aggression-related disorders. Altered neural activity has previously been found in brain areas, such as the amygdala, ventromedial prefrontal cortex, orbitofrontal cortex, anterior insula, and the caudate in children with ODD/CD when compared to typically developing children (TD) and children with ADHD (Noordermeer, Luman, & Oosterlaan, 2016; Viding, Seara-Cardoso, & McCrory, 2014). Moreover, different neural activity patterns of the amygdala in children with ODD/CD compared to TD children in response to negative (i.e. angry or fearful) face stimuli (Jones, Laurens, Herba, Barker, & Viding, 2009; Viding, Sebastian, et al., 2012) suggest impaired recognition of other's facial expressions (Blair, 2013; Veroude et al., 2016). However, previous studies have yielded inconsistent findings showing evidence of both hypo- and hyperactivity of the amygdala to affective stimuli (Coccaro, McCloskey, Fitzgerald, & Phan, 2007; Herpertz et al., 2008; Passamonti et al., 2010). This is consistent with the heterogeneity within aggression-related disorders. Two main theories might explain these divergent findings. First, the threat sensitivity theory, which describes an over-activation of limbic areas (i.e. amygdala), presumably associated with higher RA, and the deficient empathy theory, which is associated with reduced amygdala activity and more PA and higher CU traits (Blair et al., 2014). However, these studies with inconsistent findings did not take subtypes of aggression and the level of CU traits into account. Studies that considered the influence of CU traits have revealed amygdala hypoactivity in youth with HCU and amygdala hyperactivity in children with low CU traits (LCU) (Baker, Clanton, Rogers, & Brito, 2015; Blair, Veroude, & Buitelaar, 2016; Viding, Fontaine, & McCrory, 2012). Moreover, these altered amygdala responses, particularly to fearful expressions, were shown to be independent of comorbidities, such as ADHD (Hyde et al., 2016; Marsh et al., 2008; Posner et al., 2011). Nevertheless, several recent studies did not find a significant influence of CU traits on amygdala activity to negative stimuli (Dotterer, Hyde, Swartz, Hariri, & Williamson, 2017; Ewbank et al., 2018; Hyde et al., 2016), although the samples were not clinical.

Heterogeneous findings on the psychophysiological level [i.e. skin conductance response (SCR)] might also be explained by differential associations with aggression-related subtypes. Physiological hypo-arousal has been observed in children with HCU (Fanti, 2016), whereas hyper-arousal was most commonly associated with RA and internalizing symptoms (Gao, Tuvblad, Schell, Baker, & Raine, 2015; Scarpa, Haden, & Tanaka, 2010). Further, general reduced skin conductance (SC) (i.e. during resting state) has been found in ODD/CD (Lorber, 2004; Van Goozen, Matths, Cohen-Kettenis, Buitelaar, & Van England, 2000).

Our study aimed to evaluate if accounting for aggression-related subtypes and CU traits in children and adolescents with high aggression can disentangle the heterogeneity of amygdala responses and SCR to negative face stimuli into more consistent patterns, and to characterize divergent neural reactivity in response to negative face stimuli in these groups by comparing them with a large sample of TD children.

Methods and materials

Participants

Participants in the current study were part of both the EU-Aggressotype and EU-MATRICS projects. In total, 208 participants aged 8–18 years were assessed using functional magnetic resonance imaging (fMRI) across nine sites in Europe. The measures used here were part of a larger test battery including questionnaires, neuropsychological testing, MR scanning, and genotyping. Participants who were included as 'cases' were diagnosed with ODD and/or CD based on the structured diagnostic interviews with child and parents using the Kiddie Schedule for Affective Disorders and Schizophrenia (K-SADS) (Kaufman et al., 1997) according to DSM-5, or scored above the clinical cut-off (T value $\geqslant$70) for the subscales of aggressive and/or rule-breaking behavior as measured with the Child Behavior Checklist completed by parents, teachers, or youths (CBCL/TRF/YSR; Achenbach, Howell, Quay, Conners, & Bates, 1991). Exclusion criteria for all participants were any contraindications for MRI, an IQ<80 measured from four subtests (vocabulary, similarities, block design, and picture completion/matrix reasoning) of the Wechsler Intelligence Scale for Children-IV (Wechsler, 2003), and a primary DSM-5 diagnosis of psychosis, bipolar disorder, major depression, and/or an anxiety disorder. In the typically developing comparison group, no DSM axis I disorder, assessed via the K-SADS, and no clinical score in the CBCL, TRF, or YSR was allowed. For cases, medication use had to be stable for at least 2 weeks prior to inclusion. The parent-rated Inventory of Callous-Unemotional Traits (ICU) (Essau, Sasagawa, & Frick, 2006) and the self-reported Reactive Proactive Aggression Questionnaire (RPQ) (Raine et al., 2006) were used to subtype aggressive behavior. ADHD symptoms were measured with the parent-rated SNAP-IV questionnaire (Bussing et al., 2008). Ethical approval for the study was obtained for all sites separately from local ethics committees. Written (or oral) informed consent was given by the participants and their parents or legal representatives.

fMRI task

Participants performed a modified version of the emotional face-matching task (Hariri, Bookheimer, & Mazziotta, 2000). In this

task, participants completed four blocks of a perceptual face-matching task in which they had to match the presented emotions. Stimuli comprised a trio of faces in which the participants had to select one of two emotions (displayed on the bottom) identical to the target stimulus (displayed on the top). Each block consisted of six images derived from a standard set of facial affect with either negative (anger and fear) or positive faces (happy and neutral). Interleaved between these blocks, participants completed two blocks of a sensorimotor control task with geometric shapes (horizontal ellipses or vertical ellipses).

Skin conductance recording and pre-processing

SCR was recorded simultaneously with fMRI data in a subsample collected at three sites (Mannheim, Nijmegen, and Zürich), using a pair of Ag/AgCI electrodes and electrode paste with 0.5% saline (TD-246 Skin Resistance–Skin Conductance Electrode Paste, Discount Disposables, Vermont, USA) placed on the distal phalanges of digits I and II on the non-dominant hand. MR-compatible amplifiers and sensors were used (Brain Products GmbH Gilching, Germany). Data were downsampled to 10 Hz and analyzed in Ledalab (Version 3.4.9; www.ledalab.de) applying the continuous decomposition analysis and we extracted the time integral of the SCR (Benedek & Kaernbach, 2010) for further analysis.

Image acquisition and pre-processing

MRI scans were performed in nine different sites across Europe (online Supplementary Table S1). Whole-brain data were acquired with echo-planar T2*-weighted imaging (EPI), sensitive to the Blood Oxygenation Level Dependent (BOLD) signal contrast [36 axial slices (except for one site with 39 slices), 3 mm thickness; repetition time 2100 ms; echo time 35 ms; voxel size: 3 × 3 × 3 mm; Flipangle 74°; FOV = 192 mm]. Data were analyzed using SPM12 (www.fil.ion.ucl.ac.uk/spm/). The first five volumes were discarded to allow longitudinal magnetization to reach equilibrium. A high-resolution structural magnetization-prepared rapid gradient echo (MP-RAGE) scan was also acquired at a resolution of 1 × 1 × 1.2 mm. EPIs were interpolated in time to correct for slice time differences and realigned to the middle scan to correct for head movements. EPIs were co-registered and normalized to the standard EPI template in MNI space (Montreal Neurological Institute) using linear and non-linear transformations, and smoothed with a full-width-half-maximum Gaussian kernel of 8 mm. Realignment parameters were examined to ensure head movement did not exceed 3 mm.

Statistical analysis

Analysis of demographic and behavioral data

Group differences in demographic variables were analyzed using analysis of variance (ANOVA) or χ^2 tests, when appropriate. Further, behavioral performance data of the face-matching task were assessed by repeated-measures ANOVA with an experimental condition (negative faces *v.* shapes) as the within-subject factor and a between-subject factor of group. Behavioral data were corrected using age, sex, IQ, and medication as covariates of non-interest.

fMRI analysis

For each participant, a General Linear Model (GLM) assessed regionally specific effects of task parameters on BOLD indices of activation (Friston et al., 1994). The model included experimental conditions (negative and positive faces and shapes), instructions, and task end, plus six realignment parameters as covariates of no interest, to account for residual motion-related variance. Low-frequency signal drift was removed using a high-pass filter (cut-off 128 s) and an autoregressive [AR(1)] correction for serial correlations was applied.

Contrast images for the comparisons of negative faces *v.* shapes and positive faces *v.* shapes were generated. Since we expected the largest effects in the negative faces *v.* shapes condition, we concentrated on this contrast. Exploratory analyses of the positive faces *v.* shapes and negative *v.* positive faces are reported in the Supplementary material. Group differences were assessed by means of a two-sample *t* test. For group comparisons, several brain regions, including the bilateral amygdala, insula, orbitofrontal cortex, and anterior cingulate cortex, were defined as region of interest (ROI) thresholded at a corrected FWE <0.05 level and corrected for each ROI analysis (0.05/4 = 0.0125). Further, the influence of the CU traits was analyzed by regression analysis coding groups as −1 for HCU, 0 for TDs, and 1 for LCU. Participants for the HCU group were selected based on the ICU means previously published (Lozier, Cardinale, VanMeter, & Marsh, 2014; Sebastian et al., 2014; Viding, Fontaine, et al., 2012). To obtain a reliable subgroup with HCU in our sample, participants had to score ⩾38, which represents 27.7% ($n = 30$) of the cases sample. Additionally, the influence and the differential effects of subtypes of aggression were analyzed applying a regression analysis including continuous measurements of RA and PA, separately.

Brain regions were defined with the Talairach Daemon atlas implemented in the Wake Forest University (WFU) PickAtlas (Lancaster et al., 2000) using the atlas for automated anatomical labeling (Tzourio-Mazoyer et al., 2002). Exploratory whole-brain analyses are reported at an uncorrected $p < 0.001$ level for clusters including at least 10 voxels. All analyses were controlled for age, sex, IQ, medication, and site.

Finally, to account for possible influences of ADHD, we repeated all analysis adding parent-rated ADHD (continuous variable measured with the SNAP-IV questionnaire). In addition to covariate analyses, it was tested how sensitive amygdala activity was for the variables of no interest (site, medication, and sex) in general. Additionally, we matched both groups for IQ and age and repeated the main analyses. Participants were randomly selected using MedCalc Software 18.9 (MedCalc Software, Mariakerke, Belgium)

Analysis of skin conductance response

In analogy to the behavioral data, SCR data were analyzed by repeated-measures ANOVA with within-subject factors experimental condition (negative faces and shapes) and a between-subject factor of group. Additionally, the relation between SCR, RPQ, and ICU total score was investigated with Pearson's correlations. SCRs were defined as responses between 0.9 and 4 s after stimulus presentation that needed to exceed 0.01 µs (Boucsein et al., 2012). The SCR amplitude was log-transformed by means of 1 + logSCR to obtain normally distributed data.

Results

Sample characteristics

Table 1 shows the sample characteristics. From the 208 participants available for fMRI analysis, 31 participants were excluded due to excessive motion. Finally, 177 participants were included for analysis, 69 TDs and 108 cases (43 [39.8%] with ODD, 10

Table 1. Characteristics of the participants included in the functional magnetic resonance imaging analysis

	Cases ($n = 108$)		TD ($n = 69$)		ANOVA
	Mean	S.D.	Mean	S.D.	p values
Age	13.19	2.69	13.91	2.59	0.078
Sex(m)	82.4%(m)		58.0%(m)		χ^2<0.001
IQ[a]	99.28	10.62	107.44	10.69	<0.001
CBCL T score Aggression	74.45	9.99	52.14	3.58	<0.001
CBCL T score Rule breaking	67.05	9.05	52.03	3.66	<0.001
ICU total	32.99	10.02	20.45	7.73	<0.001
RPQ reactive[b]	12.40	4.73	5.85	3.54	<0.001
RPQ proactive[b]	4.71	4.69	0.88	1.45	<0.001
RPQ total[b]	17.11	8.33	6.73	4.42	<0.001
SNAP-IV[c]	31.14	12.15	5.93	6.62	<0.001
Medication (%)	60.20%	–	–	–	–
Stimulants	60.00%	–	–	–	–
Antipsychotics	30.76%	–	–	–	–
Antidepressants	4.61%	–	–	–	–
Other	4.61%	–	–	–	–

CBCL, Child Behavior Checklist; ICU, Inventory of Callous-Unemotional Traits; RPQ, Reactive Proactive Questionnaire; SNAP-IV, ADHD total score; m, male; S.D., standard deviation; TD, typically developing peers.
[a]IQ estimated from a subset of the Wechsler Intelligence Scale for Children III (Wechsler, 2003).
[b]For cases $n = 98$.
[c]For cases $n = 81$.

[9.2%] with CD alone, 19 [17.6%] with both diagnoses and 36 [33.3%] with a CBCL T value >70 in aggression or rule-breaking behavior). Compared to TDs, the cases group consisted of more males ($p < 0.001$), lower IQ ($p < 0.001$), and did differ marginally with regard to age ($p = .078$).

Behavioral data

Repeated-measures ANOVA for accuracy of correct emotional matching showed only a trend for significance between groups [$F_{(1,171)} = 2.826$, $p = 0.095$]. Cases showed overall less accuracy compared to TDs. As expected, older participants showed a higher accuracy regardless of condition ($p = 0.015$). All other covariates were not significant. In a further exploratory RM-ANOVA with a within-condition factor for further separating emotions into angry, fearful, happy, and neutral faces and shapes, the interaction term condition × group was significant [$F_{(4,684)} = 2.805$, $p = 0.026$]. Post-hoc tests revealed that the cases made more mistakes than TD in matching fear ($p = 0.018$) or neutral faces ($p < 0.001$). Regarding reaction times, no significant group differences were found [$F_{(1,171)} = 1.118$, $p = 0.292$] but a trend for a condition × group interaction effect [$F_{(1,171)} = 2.775$, $p = 0.098$] was found (online Supplementary Table S2).

fMRI

Group comparisons (cases v. TDs) for negative faces v. shapes

Figure 1 shows the group comparisons for the amygdala ROI using a t test, which revealed that cases had higher left amygdala activity compared to TDs [$t_{(163)} = 3.66$, $p_{\text{fwe-corrected}} = 0.007$, $k = 9$; $x = -27$, $y = -4$, $z = -13$]. No other effects were found in the ROIs. Group effects on a whole-brain level are depicted in online Supplementary Table S3. For positive faces $v.$ shapes, see online Supplementary Table S4.

Effects of CU traits

In total, 166 participants were available with complete CU traits data, resulting in 30 cases HCU group, 64 TDs, and 72 cases LCU group. Interestingly, HCU participants were significantly older than the LCU subgroup, and showed significantly higher scores for proactive aggression ($p < 0.001$), but not for reactive aggression. For more details, see Table 2.

Regression analysis was only performed for the left amygdala, since only this ROI was significant at the group differences, and showed a significant association with CU traits [$t_{(153)} = 3.27$, $p_{\text{fwe-corrected}} = 0.012$, $k = 3$, $x = -12$, $y = -1$, $z = -16$]. The HCU group showed the lowest amygdala activity for negative faces $v.$ shapes, TD children showing intermediate activity, and LCU children the highest activity for this contrast (Fig. 2). The whole-brain analysis is shown in online Supplementary Table S5. For the positive faces $v.$ shapes contrast, no significant group difference or an association with CU traits was found.

Effects of reactive and proactive aggression subtypes

When adding reactive and proactive aggression as covariates in the same model of CU traits, the effect of CU traits on the left amygdala remained significant [$t_{(144)} = 3.08$, $p_{\text{fwe-corrected}} = 0.018$, $k = 1$, $x = -12$, $y = -1$, $z = -16$]. Additionally, we assessed the RPQ without CU traits which did not show any significant association. Exploratory analysis for the cases group only showed a negative relationship with the proactive subscale for the left amygdala at a trend level [$t_{(85)} = 2.37$, $p_{\text{fwe-corrected}} = 0.091$, $k = 1$, $x = -12$,

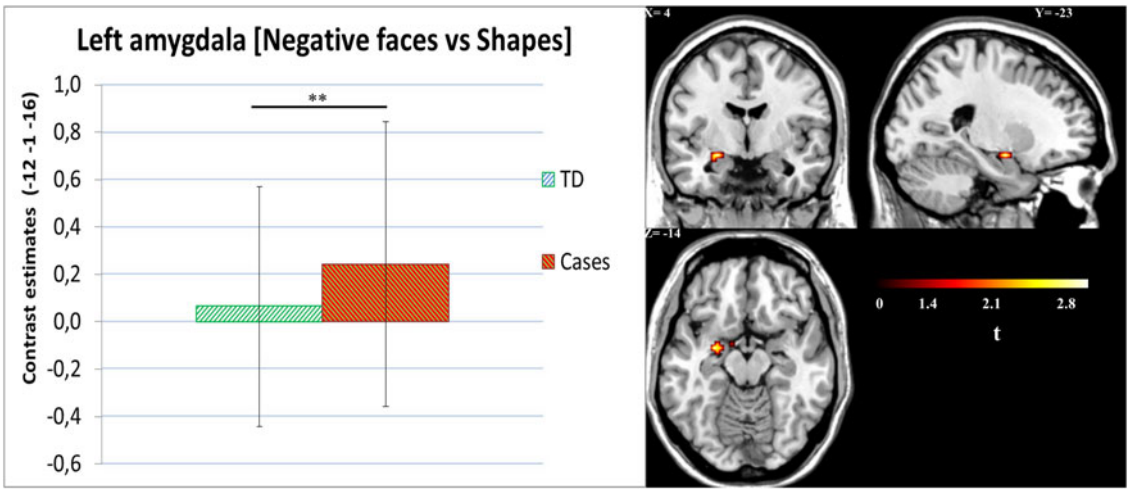

Fig. 1. Negative faces *v.* shapes. Left amygdala activity for cases *v.* TD group. Cases showed higher amygdala activity [$t_{(163)} = 3.66$, $p_{\text{fwe-corrected}} = 0.007$, $k = 9$; $x = -27$, $y = -4$, $z = -13$].Cases = ODD/CD, oppositional defiant disorder; CD, conduct disorder; TD, typically developing peers.

Table 2. Characteristics of the participants included in the regression analysis

	LCU			TD			HCU			ANOVA	
	n	Mean	S.D.	*n*	Mean	S.D.	*n*	Mean	S.D.	*p* values	Post-hoc
Age	72	12.75	2.64	64	13.93	2.54	30	13.91	2.77	0.019	LCU < TD = HCU
Sex(m)	72	84.9%		64	56.30%		30	83.30%		$\chi^2 < 0.001$	TD < LCU = HCU
Medication (%)	72	53.4%		64	0.00%		30	63.30%		$\chi^2 < 0.001$	TD < LCU = HCU
IQ[a]	72	99.71	9.99	64	107.81	10.87	30	99.61	12.10	<0.001	LCU = HCU < TD
CBCL *T* score Rule breaking	72	64.92	8.79	64	52.09	3.74	30	72.84	7.52	<0.001	TD < LCU < HCU
CBCL *T* score Aggression	72	73.72	10.65	64	52.22	3.65	30	76.83	7.97	<0.001	TD < LCU = HCU
ICU total	72	27.89	5.96	64	20.45	7.73	30	45.23	6.55	<0.001	TD < LCU < HCU
RPQ reactive	69	12.24	4.50	63	5.83	3.55	29	13.10	5.05	<0.001	TD < LCU =HCU
RPQ proactive	69	3.59	3.90	63	0.90	1.48	29	7.45	5.56	<0.001	TD < LCU < HCU
RPQ total	69	15.82	7.24	63	6.73	4.47	29	20.55	9.68	<0.001	TD < LCU < HCU
SNAP-IV	58	28.80	10.87	61	5.92	6.75	23	38.74	12.41	<0.001	TD < LCU <HCU
Medication (%)		52.0%						63.3%		ns	LCU =HCU
Stimulants		73.6%						47.3%		$\chi^2 = 0.040$	LCU > HCU
Antipsychotics		31.5%						36.8%		ns	LCU = HCU
Antidepressants		5.2%						5.2%		ns	LCU = HCU
Other		2.6%						10.5%		ns	LCU = HCU

CBCL, Child Behavior Checklist; ICU, Inventory of Callous-Unemotional Traits; RPQ, Reactive Proactive Questionnaire; SNAP-IV, ADHD total score; S.D., standard deviation; LCU, low ICU; HCU, high ICU; TD, typically developing peers.
[a]IQ estimated from a subset of the Wechsler Intelligence Scale for Children III (Wechsler, 2003).

$y = -1, z = -16$]. At whole-brain level for both groups (at an uncorrected level), a positive relationship with the right fusiform area [$t_{(150)} = 4.10$, $p_{\text{uncor}}<0.001$, $k = 15$, $x = -42$, $y = -34$, $z = -16$] was observed.

Sensitivity analyses

ADHD as an additional covariate

To control for potential influences of ADHD symptoms, we added the SNAP-IV as a covariate. In total, 158 participants were available with complete ADHD symptom data. The inclusion of this covariate further strengthened the results of the main group comparison [left amygdala: $t_{(143)} = 3.63$, $p_{\text{fwe-corrected}} = 0.008$, $k = 16$; $x = -24$, $y = -4$, $z = -13$; right amygdala $t_{(143)} = 3.35$, $p_{\text{fwe-corrected}} = 0.018$, $k = 16$; $x = 27$, $y = -4$, $z = -13$] and for the CU effect [left amygdala: $t_{(133)} = 3.60$, $p_{\text{fwe-corrected}} = 0.010$, $k = 6$; $x = -12$, $y = -1$, $z = -16$].

Effects of control variables: medication, site, age, and sex

Medication was related to amygdala activity [$F_{(1,164)} = 7.814$, $p = 0.006$], with higher amygdala activity during the negative *v.*

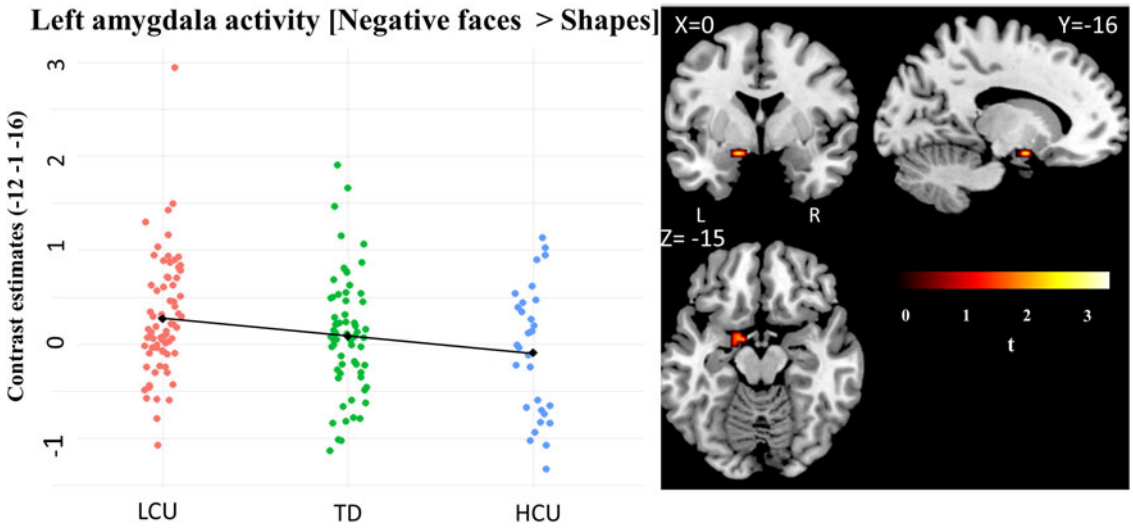

Fig. 2. Left amygdala activity for negative *v.* shapes. Group-specific amygdala activity for negative *v.* shapes contrast depending on the CU subtypes [$t_{(153)} = 3.27$, $p_{\text{fwe-corrected}} = 0.012$, $k = 3$; $x = -12$, $y = -1$, $z = -16$]. HCU, high callous-unemotional traits; TD, typically developing peers; LCU, low callous-unemotional traits.

shapes contrast in non-medicated participants [$t_{(164)} = 3.32$, $p_{\text{fwe-corrected}} = 0.010$, $k = 8$; $x = -15$, $y = 2$, $z = -16$]. A significant effect of site on amygdala activity [$F_{(8,164)} = 2.259$, $p = 0.026$] was observed. Nevertheless, when excluding unbalanced sites (four sites with fewer than five participants per group), no significant impact of site was found [$F_{(8,131)} = 1.159$, $p = 0.181$], and the main result did not change [$t_{(131)} = 3.53$, $p_{\text{fwe-corrected}} = 0.011$, $k = 19$; $x = -24$, $y = -4$, $z = -13$]. In addition, there was no significant effect of age and sex. For details, see online Supplementary Tables S6, S7, and S8.

Matching groups for age and IQ

Matching both groups left 112 participants (56 in each group) for the main analysis including sex and medication as covariates. Amygdala activity in the cases group remained significantly increased [$t_{(110)} = 3.12$, $p_{\text{fwe-corrected}} = 0.018$, $k = 3$; $x = -27$, $y = -4$, $z = -13$]. However, the CU effect reached only a trend ($p = 0.081$), presumably related to the lower sample size.

Skin conductance

Simultaneous fMRI and SC data were available for 37 cases and 26 TDs. A significant interaction between experimental condition and group was found [$F_{(1,61)} = 5.523$, $p = 0.022$]. In the cases group, a lower SCR to negative facial stimuli ($p = 0.002$), but not to shapes ($p = 0.280$) was seen compared to TDs. The total score on the ICU scale was negatively associated with SCR for negative faces ($r = -0.395$, $p = 0.001$) and shapes ($r = -0.300$, $p = 0.018$) (Fig. 3). Additionally, significant correlations between RA ($r = -0.292$, $p = 0.021$), PA aggression ($r = -0.286$, $p = 0.024$), and RPQ total scale ($r = -0.324$, $p = 0.010$) were found for SCR of negative faces only.

Discussion

Our study addressed the neural characterization of aggression-related subtypes and CU traits in children and adolescents with aggression-related problems from a large multicenter cohort during a well-established and robust fMRI task. Cases showed different levels of amygdala activity during the presentation of negative faces *v.* shapes than TDs. However, even more importantly, when considering effects of CU traits, our results showed that increased CU traits were associated with amygdala hypoactivation, and that only patients with low CU traits showed increased amygdala activity to negative faces. This finding is in line with previous studies showing higher amygdala activity in youth with low CU traits, and lower activity in those with high CU traits (Viding et al., 2014). Regarding subtypes of aggression (reactive and proactive aggression), we did not find any significant association that survived family-wise correction, but there was a trend for a negative relationship between PA and amygdala activity to negative faces.

The general higher activity in the amygdala adds evidence to the heightened threat sensitivity theory in aggression-related disorders (Blair et al., 2014; Dotterer et al., 2017; Viding, Fontaine, et al., 2012). Importantly, this effect remained stable after controlling for age, sex, medication, site, IQ, ADHD, and after excluding one outlier (>3SD). Additionally, our results revealed that this higher amygdala activity showed a phenotype-specific pattern for participants with significantly lower PA.

Concerning the differential effect of CU traits, our results showed that these traits are able to disentangle specific neural alterations, which is in line with previous findings (Jones et al., 2009; Lozier et al., 2014; Marsh et al., 2008; Viding et al., 2014; White et al., 2012). It is worth noting that in our study, only the most severe CU patients (ICU >38) showed amygdala underactivation. This probably underlies our finding of on average increased amygdala activity across the entire sample of cases and highlights the importance of subtyping approaches when investigating the neural basis of aggression. Earlier studies using the same instrument found LCU-specific effects on amygdala activity, in an even higher CU traits population (ICU mean of 52) (Lozier et al., 2014; Sebastian et al., 2014; Viding, Fontaine, et al., 2012). In line with a recent study (Docherty, Boxer, Huesmann, O'Brien, & Bushman, 2017) which evaluated a cut-off for the ICU questionnaire (ICU>40), our finding might be an important result which might reflect a biological correlate (ICU⩾38; lower amygdala) which could be used in the

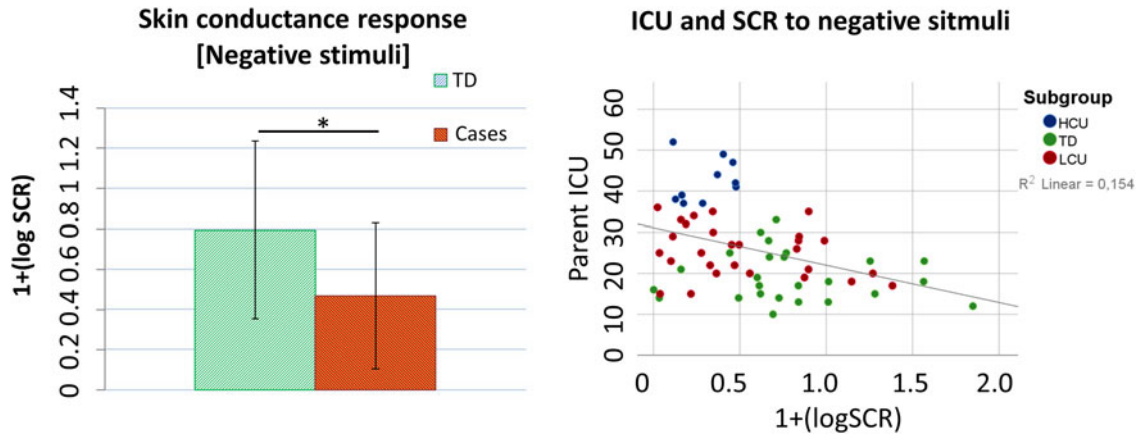

Fig. 3 Skin conductance response and ICU. Skin conductance response (SCR). Cases showed lower SCR activity to negative faces. ICU was negatively associated with SCR and the CU. TD, typically developing peers; HCU, high callous-unemotional traits; LCU, low callous-unemotional traits; ICU, inventory of callous-unemotional traits.

classification of aggression-related disorders (specifier) and probably subtypes. Interestingly, the HCU subgroup showed higher scores for PA compared to the LCU subgroup. However, no differences between high and low CU subgroups were found for RA. Some studies showed that both aggression-related subtypes are associated with high CU traits (Kimonis et al., 2008; Pechorro, Ray, Gonçalves, & Jesus, 2017; Waller et al., 2015), while one study reported that only PA is correlated with CU traits (Urben et al., 2018), whereas in our sample, both PA and RA correlated high with CU traits (PA $r = 0.51$, RA $r = 0.41$). Further, while CU traits were obtained using parent-ratings, PA and RA were self-rated. Thus, we cannot rule out informant-based biases in self-ratings and further investigations using the same informants for all measures are warranted. Altogether, our result might suggest that CU traits measured via the parent-reported ICU questionnaire reflect stable traits, which are more prone to identify subtype-specific aggression.

Finally, the SC data showed general physiological hypoactivation in response to negative emotions in cases compared to TDs, and a negative association between SCR, CU traits, PA, and RA were found. These findings are in line with numerous studies (Blair, 1999; Fanti, 2016; Herpertz et al., 2005, 2008) showing reduced SC in aggression-related disorders, particularly those with high CU traits. However, the SCR and fMRI data showed divergent patterns with higher amygdala activity in the LCU subgroup when compared with TDs. This, together with the overall reduced SCR, might suggest an interrupted physiological circuit with neural processes involved in response to affective stimuli in cases within the LCU subgroup. However, this should be interpreted with caution, since our fMRI-SCR data were only based on a subsample of cases.

Strengths and limitations

The strengths of this study include a large sample of aggression-related disorders and TDs children and adolescents, the assessments of reactive and proactive aggression and CU traits, enabling to disentangle subtype- and trait-specific differences, and a well-established fMRI task to elicit amygdala activity. There are also limitations worth noting. First, the multicenter nature of this study, in which nine different institutes participated and contributed to a sample size which would have been difficult to reach at an individual site, might have also introduced heterogeneity. However, sensitivity analysis with fewer sites did not change the main results, indicating that this did not negatively influence the results. Second, our relatively small proportion of subjects high in CU traits (27.7%) suggests that our sample is predominantly reactively aggressive, since there were no significant differences between low and high CU subgroup on reactive aggression. Moreover, within the emotional face-matching task, there are two major shortcomings; (1) our task was an explicit emotional-matching task, whereas the majority of other studies used an implicit task (identifying gender) or passive viewing of emotions and (2) the negative faces included two emotions in a block design (fear and angry). These shortcomings could have diluted our effects as studies which showed CU effects on amygdala activity found mainly effects for fearful faces (Jones et al., 2009; Lozier et al., 2014; Marsh et al., 2008; Viding, Fontaine, et al., 2012). Interestingly, this is confirmed by our performance data with fewer correct responses specifically during the matching of fearful faces.

Conclusion

In summary, this large study compared children and adolescents with aggression-related problems *v.* TD during an fMRI emotional face-matching task, investigating the role of subtypes of aggression and CU traits. Overall, children and adolescents with high aggression showed amygdala hyperactivity in emotion and face processing areas, particularly in the subgroup with low CU traits. In contrast, in those with high CU traits, amygdala hypoactivity was observed. Our findings underline the importance to specify subtypes and CU traits in aggression-related disorders, brain-based evidence and therefore providing a possible biomarker, which could be used for improved diagnostics and personalized treatment.

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References

Achenbach, T. M., Howell, C. T., Quay, H. C., Conners, C. K., & Bates, J. E. (1991). National survey of problems and competencies among four- to sixteen-year-olds: Parents' reports for normative and clinical samples. *Monographs of the Society for Research in Child Development*, *56*(3), i–130. doi:10.2307/1166156.

American Psychiatric Association (2013). (n.d.). American Psychiatric Association, 2013. Retrieved July 15, 2015, from http://www.dsm5.org/Pages/Default.aspx.

Baker, R. H., Clanton, R. L., Rogers, J. C., & Brito, S. A. D. (2015). Neuroimaging findings in disruptive behavior disorders. *CNS Spectrums*, *20*(4), 369–381. doi:10.1017/S1092852914000789.

Benedek, M., & Kaernbach, C. (2010). A continuous measure of phasic electrodermal activity. *Journal of Neuroscience Methods*, *190*(1–5), 80–91. doi:10.1016/j.jneumeth.2010.04.028.

Blair, R. J. R. (1999). Responsiveness to distress cues in the child with psychopathic tendencies. *Personality and Individual Differences*, *27*(1), 135–145. doi:10.1016/S0191-8869(98)00231-1.

Blair, R. J. R. (2013). Psychopathy: Cognitive and neural dysfunction. *Dialogues in Clinical Neuroscience*, *15*(2), 181–190.

Blair, R. J. R., Leibenluft, E., & Pine, D. S. (2014). Conduct disorder and callous–unemotional traits in youth. *New England Journal of Medicine*, *371*(23), 2207–2216. doi:10.1056/NEJMra1315612.

Blair, R. J. R., Veroude, K., & Buitelaar, J. K. (2016). Neuro-cognitive system dysfunction and symptom sets: A review of fMRI studies in youth with conduct problems. *Neuroscience & Biobehavioral Reviews*, *91*, 69–90. doi:10.1016/j.neubiorev.2016.10.022.

Boucsein, W., Fowles, D. C. F., Grimnes, S., Ben-Shakhar, G., Roth, W., Dawson, M. E., & Filion, D. L. (2012). Publication recommendations for electrodermal measurements. *Psychophysiology*, *49*(8), 1017–1034. doi:10.1111/j.1469-8986.2012.01384.x.

Bussing, R., Fernandez, M., Harwood, M., Hou, W., Garvan, C. W., Swanson, J. M., & Eyberg, S. M. (2008). Parent and teacher SNAP-IV ratings of attention deficit/hyperactivity disorder symptoms: Psychometric properties and normative ratings from a school district sample. *Assessment*, *15*(3), 317–328. doi:10.1177/1073191107313888.

Coccaro, E. F., McCloskey, M. S., Fitzgerald, D. A., & Phan, K. L. (2007). Amygdala and orbitofrontal reactivity to social threat in individuals with impulsive aggression. *Biological Psychiatry*, *62*(2), 168–178. doi:10.1016/j.biopsych.2006.08.024.

Docherty, M., Boxer, P., Huesmann, L. R., O'Brien, M., & Bushman, B. (2017). Assessing callous-unemotional traits in adolescents: Determining cutoff scores for the inventory of callous and unemotional traits. *Journal of Clinical Psychology*, *73*(3), 257–278. doi:10.1002/jclp.22313.

Dotterer, H. L., Hyde, L. W., Swartz, J. R., Hariri, A. R., & Williamson, D. E. (2017). Amygdala reactivity predicts adolescent antisocial behavior but not callous-unemotional traits. *Developmental Cognitive Neuroscience*, *24* (Supplement C), 84–92. doi:10.1016/j.dcn.2017.02.008.

Essau, C. A., Sasagawa, S., & Frick, P. J. (2006). Callous-unemotional traits in a community sample of adolescents. *Assessment*, *13*(4), 454–469. doi:10.1177/1073191106287354.

Ewbank, M. P., Passamonti, L., Hagan, C. C., Goodyer, I. M., Calder, A. J., & Fairchild, G. (2018). Psychopathic traits influence amygdala–anterior cingulate cortex connectivity during facial emotion processing. *Social Cognitive and Affective Neuroscience*, *13*(5), 525–534. doi:10.1093/scan/nsy019.

Fanti, K. A. (2016). Understanding heterogeneity in conduct disorder: A review of psychophysiological studies. *Neuroscience & Biobehavioral Reviews*, *91*, 4–20. doi:10.1016/j.neubiorev.2016.09.022.

Frick, P. J., & Viding, E. (2009). Antisocial behavior from a developmental psychopathology perspective. *Development and Psychopathology*, *21*(4), 1111–1131. doi:10.1017/S0954579409990071.

Friston, K. J., Holmes, A. P., Worsley, K. J., Poline, J.-P., Frith, C. D., & Frackowiak, R. S. J. (1994). Statistical parametric maps in functional imaging: A general linear approach. *Human Brain Mapping*, *2*(4), 189–210. doi:10.1002/hbm.460020402.

Gao, Y., Tuvblad, C., Schell, A., Baker, L., & Raine, A. (2015). Skin conductance fear conditioning impairments and aggression: A longitudinal study. *Psychophysiology*, *52*(2), 288–295. doi:10.1111/psyp.12322.

Hariri, A. R., Bookheimer, S. Y., & Mazziotta, J. C. (2000). Modulating emotional responses: Effects of a neocortical network on the limbic system. *Neuroreport*, *11*(1), 43–48.

Herpertz, S. C., Huebner, T., Marx, I., Vloet, T. D., Fink, G. R., Stoecker, T., … Herpertz-Dahlmann, B. (2008). Emotional processing in male adolescents with childhood-onset conduct disorder. *Journal of Child Psychology and Psychiatry*, *49*(7), 781–791. doi:10.1111/j.1469-7610.2008.01905.x.

Herpertz, S. C., Mueller, B., Qunaibi, M., Lichterfeld, C., Konrad, K., & Herpertz-Dahlmann, B. (2005). Response to emotional stimuli in boys with conduct disorder. *The American Journal of Psychiatry*, *162*(6), 1100–1107. doi:10.1176/appi.ajp.162.6.1100.

Hyde, L. W., Shaw, D. S., Murray, L., Gard, A., Hariri, A. R., & Forbes, E. E. (2016). Dissecting the role of amygdala reactivity in antisocial behavior in a sample of young, low-income, urban men. *Clinical Psychological Science*, *4*(3), 527–544. doi:10.1177/2167702615614511.

Jones, A. P., Laurens, K. R., Herba, C. M., Barker, G. J., & Viding, E. (2009). Amygdala hypoactivity to fearful faces in boys with conduct problems and callous-unemotional traits. *American Journal of Psychiatry*, *166*(1), 95–102. doi:10.1176/appi.ajp.2008.07071050.

Kaufman, J., Birmaher, B., Brent, D., Rao, U., Flynn, C., Moreci, P., … Ryan, N. (1997). Schedule for affective disorders and schizophrenia for school-age children-present and lifetime version (K-SADS-PL): Initial reliability and validity data. *Journal of the American Academy of Child & Adolescent Psychiatry*, *36*(7), 980–988. doi:10.1097/00004583-199707000-00021.

Kimonis, E. R., Frick, P. J., Skeem, J. L., Marsee, M. A., Cruise, K., Munoz, L. C., … Morris, A. S. (2008). Assessing callous–unemotional traits in adolescent offenders: Validation of the Inventory of Callous–Unemotional Traits. *International Journal of Law and Psychiatry*, *31*(3), 241–252. doi:10.1016/j.ijlp.2008.04.002.

Lancaster, J. L., Woldorff, M. G., Parsons, L. M., Liotti, M., Freitas, C. S., Rainey, L., … Fox, P. T. (2000). Automated Talairach atlas labels for functional brain mapping. *Human Brain Mapping*, *10*(3), 120–131.

Loeber, R., Burke, J. D., Lahey, B. B., Winters, A., & Zera, M. (2000). Oppositional defiant and conduct disorder: A review of the past 10 years,

part I. *Journal of the American Academy of Child and Adolescent Psychiatry*, *39*(12), 1468–1484. doi:10.1097/00004583-200012000-00007.

Lorber, M. F. (2004). Psychophysiology of aggression, psychopathy, and conduct problems: A meta-analysis. *Psychological Bulletin*, *130*(4), 531–552. doi:10.1037/0033-2909.130.4.531.

Lozier, L. M., Cardinale, E. M., VanMeter, J. W., & Marsh, A. A. (2014). Mediation of the relationship between callous-unemotional traits and proactive aggression by amygdala response to fear among children with conduct problems. *JAMA Psychiatry*, *71*(6), 627–636. doi:10.1001/jamapsychiatry.2013.4540.

Marsh, A. A., Finger, E. C., Mitchell, D. G. V., Reid, M. E., Sims, C., Kosson, D. S., … Blair, R. J. R. (2008). Reduced amygdala response to fearful expressions in children and adolescents with callous-unemotional traits and disruptive behavior disorders. *The American Journal of Psychiatry*, *165*(6), 712–720. doi:10.1176/appi.ajp.2007.07071145.

Noordermeer, S. D. S., Luman, M., & Oosterlaan, J. (2016). A systematic review and meta-analysis of neuroimaging in oppositional defiant disorder (ODD) and conduct disorder (CD) taking attention-deficit hyperactivity disorder (ADHD) into account. *Neuropsychology Review*, *26*(1), 44–72. doi:10.1007/s11065-015-9315-8.

Passamonti, L., Fairchild, G., Goodyer, I. M., Hurford, G., Hagan, C. C., Rowe, J. B., & Calder, A. J. (2010). Neural abnormalities in early-onset and adolescence-onset conduct disorder. *Archives of General Psychiatry*, *67*(7), 729–738. doi:10.1001/archgenpsychiatry.2010.75.

Pechorro, P., Ray, J. V., Gonçalves, R. A., & Jesus, S. N. (2017). The Inventory of Callous-Unemotional Traits: Psychometric properties among referred and non-referred Portuguese female juveniles. *International Journal of Law and Psychiatry*, *54*, 67–75. doi:10.1016/j.ijlp.2017.05.002.

Polanczyk, G. V., Salum, G. A., Sugaya, L. S., Caye, A., & Rohde, L. A. (2015). Annual research review: A meta-analysis of the worldwide prevalence of mental disorders in children and adolescents. *Journal of Child Psychology and Psychiatry*, *56*(3), 345–365. doi:10.1111/jcpp.12381.

Posner, J., Nagel, B. J., Maia, T. V., Mechling, A., Oh, M., Wang, Z., & Peterson, B. S. (2011). Abnormal amygdalar activation and connectivity in adolescents with attention-deficit/hyperactivity disorder. *Journal of the American Academy of Child and Adolescent Psychiatry*, *50*(8), 828–837.e3. doi:10.1016/j.jaac.2011.05.010.

Raine, A., Dodge, K., Loeber, R., Gatzke-Kopp, L., Lynam, D., Reynolds, C., … Liu, J. (2006). The reactive-proactive aggression questionnaire: Differential correlates of reactive and proactive aggression in adolescent boys. *Aggressive Behavior*, *32*(2), 159–171. doi:10.1002/ab.20115.

Scarpa, A., Haden, S. C., & Tanaka, A. (2010). Being hot-tempered: Autonomic, emotional, and behavioral distinctions between childhood reactive and proactive aggression. *Biological Psychology*, *84*(3), 488–496. doi:10.1016/j.biopsycho.2009.11.006.

Sebastian, C. L., McCrory, E. J., Dadds, M. R., Cecil, C. A. M., Lockwood, P. L., Hyde, Z. H., … Viding, E. (2014). Neural responses to fearful eyes in children with conduct problems and varying levels of callous-unemotional traits. *Psychological Medicine*, *44*(1), 99–109. doi:10.1017/S0033291713000482.

Tzourio-Mazoyer, N., Landeau, B., Papathanassiou, D., Crivello, F., Etard, O., Delcroix, N., … Joliot, M. (2002). Automated anatomical labeling of activations in SPM using a macroscopic anatomical parcellation of the MNI MRI single-subject brain. *NeuroImage*, *15*(1), 273–289. doi:10.1006/nimg.2001.0978.

Urben, S., Habersaat, S., Pihet, S., Suter, M., de Ridder, J., & Stéphan, P. (2018). Specific contributions of age of onset, callous-unemotional traits and impulsivity to reactive and proactive aggression in youths with conduct disorders. *The Psychiatric Quarterly*, *89*(1), 1–10. doi:10.1007/s11126-017-9506-y.

Van Goozen, S. H. M., Matths, W., Cohen-Kettenis, P. T., Buitelaar, J. K., & Van England, H. (2000). Hypothalamic-pituitary-adrenal axis and autonomic nervous system activity in disruptive children and matched controls. *Journal of the American Academy of Child & Adolescent Psychiatry*, *39*(11), 1438–1445. doi:10.1097/00004583-200011000-00019.

Veroude, K., von Rhein, D., Chauvin, R. J. M., van Dongen, E. V., Mennes, M. J. J., Franke, B., … Buitelaar, J. K. (2016). The link between callous-unemotional traits and neural mechanisms of reward processing: An fMRI study. *Psychiatry Research: Neuroimaging*, *255*, 75–80. doi:10.1016/j.pscychresns.2016.08.005.

Viding, E., Fontaine, N. M., & McCrory, E. J. (2012). Antisocial behaviour in children with and without callous-unemotional traits. *Journal of the Royal Society of Medicine*, *105*(5), 195–200. doi:10.1258/jrsm.2011.110223.

Viding, E., Seara-Cardoso, A., & McCrory, E. J. (2014). Antisocial and callous behaviour in children. *Current Topics in Behavioral Neurosciences*, *17*, 395–419. doi:10.1007/7854_2013_266.

Viding, E., Sebastian, C. L., Dadds, M. R., Lockwood, P. L., Cecil, C. A. M., De Brito, S. A., & McCrory, E. J. (2012). Amygdala response to preattentive masked fear in children with conduct problems: The role of callous-unemotional traits. *American Journal of Psychiatry*, *169*(10), 1109–1116. doi:10.1176/appi.ajp.2012.12020191.

Waller, R., Wright, A. G. C., Shaw, D. S., Gardner, F., Dishion, T. J., Wilson, M. N., & Hyde, L. W. (2015). Factor structure and construct validity of the parent-reported inventory of callous-unemotional traits among high-risk 9-year-olds. *Assessment*, *22*(5), 561–580. doi:10.1177/1073191114556101.

Wechsler, D. (2003). Wechsler Intelligence Scale for Children-WISC-IV. Retrieved from http://homepages.abdn.ac.uk/j.crawford/pages/dept/sf_wisc4.htm.

White, S. F., Marsh, A. A., Fowler, K. A., Schechter, J. C., Adalio, C., Pope, K., … Blair, R. J. R. (2012). Reduced amygdala response in youths with disruptive behavior disorders and psychopathic traits: Decreased emotional response versus increased top-down attention to nonemotional features. *American Journal of Psychiatry*, *169*(7), 750–758. doi:10.1176/appi.ajp.2012.11081270.