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Amygdala response to pre-attentive masked fear is associated with callous-unemotional traits in children with conduct problems

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Abstract

Objective: In children with conduct problems, high levels of callous-unemotional traits are associated with amygdala hypoactivity to consciously-perceived fear, while low levels of callous-unemotional traits may be associated with amygdala *hyper*activity. Behavioral data suggest fear processing deficits in children with high callous-unemotional traits may extend to stimuli presented below conscious awareness (pre-attentively). We investigated the neural basis of this effect. Amygdala involvement was predicted on the basis of its role in pre-attentive affective processing in healthy adults, and its dysfunction in previous studies of conduct problems.

Method: fMRI was used to measure neural responses to fearful and calm faces **presented pre-attentively (for 17ms followed by backward masking)** in children with conduct problems and high callous-unemotional traits (n=15), conduct problems and low callous-unemotional traits (n=15), and typically developing controls (n=16). Amygdala response for Fear-Calm was predicted to differentiate groups, with greatest response in children with conduct problems and low callous-unemotional traits, and lowest in children with conduct problems and high callous-unemotional traits.

Results: In right amygdala, a greater response was seen in children with conduct problems and low callous-unemotional traits than in those with high callous-unemotional traits. Findings were not explained by levels of conduct disorder, attention-deficit hyperactivity disorder, anxiety, or depression. **Conclusions:** These data demonstrate differential amygdala activity to pre-attentively presented fear in children with conduct problems grouped by callous-unemotional traits, with high levels associated with lower amygdala reactivity. Our findings complement increasing evidence suggesting that callous-unemotional traits are an important specifier in the classification of children with conduct problems.

Introduction

Children with conduct problems are at risk of developing persistent antisocial behavior and other mental and physical health problems (1, 2). Conduct problems are a common reason for a referral to mental health and educational services, and represent a considerable public health cost (3). Callous-unemotional traits (lack of empathy and guilt, shallow affect) characterize a particularly problematic group of children with more severe conduct problems (2). Twin studies suggest that conduct problems with callous-unemotional traits are highly heritable, while conduct problems without callous-unemotional traits are driven primarily by environmental influences (4, 5). Children with conduct problems and high callous-unemotional traits are characterized by deficits in processing others' fearful and sad facial expressions and vocal tones (2, 6). In contrast, those with low callous-unemotional traits appear over-sensitive to perceived social threat, including anger and even ambiguous, neutral expressions (7, 2). Inclusion of callous-unemotional traits as a conduct disorder specifier is being considered for the revised Diagnostic and Statistical Manual of Mental Disorders (DSM-5) (8).

Most functional magnetic resonance imaging (fMRI) studies of children/adolescents with conduct problems have reported atypical activation of the amygdala (9, 10), a subcortical structure implicated in the processing of salient stimuli, including emotional facial expressions (11). fMRI data focusing on children with conduct problems without accounting for individual differences in callous-unemotional traits have been mixed, with evidence of both amygdala hypo- and hyper-activity to affective stimuli (12, 13, 14). These mixed findings may partly reflect differences in paradigms used across studies. They may also partly reflect variations in callous-unemotional traits across samples, given significant differences in emotional reactivity and behavioral responses to emotional stimuli in children with high vs. low callous-unemotional traits (2, 7).

Lower amygdala activity to fearful facial expressions has been reported for children with conduct problems and high callous-unemotional traits compared with typically developing children or children with attention deficit-hyperactivity symptoms (15, 16). A recent study from our group measured fMRI responses in children with conduct problems while they watched scenarios requiring affective (versus cognitive) Theory of Mind **(i.e. the ability to understand emotions compared with intentions and beliefs)**. Unique variance associated with callous-unemotional traits was negatively related, whilst unique variance associated with conduct disorder symptoms was positively related to amygdala response in children with conduct problems (17). This finding mirrors behavioral studies documenting deficits processing fear and sadness in children with conduct problems and high callous-unemotional traits (6), but heightened sensitivity to social threat in those with low callous-unemotional traits (7, 2); and further suggests heterogeneity in amygdala reactivity to emotional stimuli in children with conduct problems. Amygdala hypoactivity in children with conduct problems and high callous-unemotional traits (15, 16, 17) could partly explain associated clinical phenomena such as pre-meditated aggression, lack of empathy, and difficulties in learning from punishment (18). In contrast, amygdala hyperactivity in children with conduct problems and low callous-unemotional traits (12, 17) may partly explain clinical phenomena such as reactive aggression and difficulties in regulating emotions (2).

The amygdala responds to salient stimuli both when presented pre-attentively (i.e. before reaching conscious awareness or attention (19)), and under prolonged viewing conditions (11, 20, 21). This is consistent with the amygdala's role as part of a functional network engaged in triggering an orienting response to salient stimuli, including emotional facial expressions, so that appropriate processing of and behavioral responses to such stimuli can be prioritized. All previous fMRI studies of children with conduct problems have focused on affective stimuli presented under prolonged viewing conditions. However, atypical amygdala activity to pre-attentively presented affective stimuli may also characterize some children with conduct problems.

A recent behavioral study by Sylvers et al. (22) assessed time taken for emotional faces increasing in contrast salience, to break into consciousness during a continuous flash suppression task. Elevated callous-unemotional trait scores were associated with greater lag times for fearful, and, to a lesser extent, disgusted faces, to break through to conscious awareness relative to neutral faces. This effect was particularly pronounced in children with high levels of impulsive behavior. These pre-attentive data complement studies showing a fear processing deficit to overtly presented stimuli in children with conduct problems and high callous-unemotional traits.

The current study is the first to use fMRI to investigate pre-attentive fear processing in children with conduct problems. We focused on fear processing as fearful faces signal potential threat in the surroundings and index distress. Children with conduct problems and high callous-unemotional traits are fearless and insensitive to others' distress (2). In contrast, children with conduct problems and low callous-unemotional traits are emotionally reactive to threat (2). Extrapolating from previous data, we predicted that children with conduct problems and high callous-unemotional traits would show the lowest amygdala response to pre-attentively presented fearful vs. calm faces, children with conduct problems and low callous-unemotional traits would show the greatest response, and typically developing controls would show an intermediate response.

Methods

Participants: Males aged 10-16 were recruited from the community via newspaper advertisements/local schools. Screening questionnaires were administered to parents and teachers of 176 boys expressing an interest in taking part. These yielded: a research diagnosis of conduct problems; dimensional assessment of callous-unemotional traits; an overall psychopathology screen; demographic data; and information regarding neurological or psychiatric diagnoses. Current conduct problems were assessed using the Child and Adolescent Symptom Inventory-4R (23) Conduct Disorder scale, and callous-unemotional traits were assessed using the Inventory of Callous-Unemotional Traits (24). Both were scored by taking the highest ratings from either the parent or teacher questionnaire for any given item (25). The Strengths and Difficulties Questionnaire (26) was used as a brief screening measure for psychopathology (Table S1).

On the basis of the screening information participants were invited for an fMRI scan; this group largely overlapped with a previous sample (17). Child and Adolescent Symptom Inventory-4R Conduct Disorder cut-off scores for inclusion in the conduct problems group were 3+ (ages 10-14) and 6+ (ages 15-16). Scores of this magnitude and above are associated with a clinical diagnosis of conduct disorder (27). Children with conduct problems were divided into low and high callous-unemotional trait groups based on a median split of callous-unemotional trait scores (median=44.5).

Groups were matched on IQ, age, handedness, ethnicity and SES. All controls scored below the conduct problems group median on callous-unemotional traits, and scored in the normal range on each subscale (including conduct disorder) of the Strengths and Difficulties Questionnaire. For all groups, exclusion criteria included a previous diagnosis of neurological or psychotic disorder, or a current prescription for psychiatric medication. **(We later found that two participants had been medicated for ADHD symptoms at the time of scanning. However, analyses conducted with and without these participants were very similar, and so their data were included in reported analyses)**. To ensure a representative group of children with conduct problems, common co-morbidities (attention deficit hyperactivity disorder, generalized anxiety disorder, depression and substance/alcohol abuse) were not used as exclusion criteria, but current parent-reported symptom counts were obtained during fMRI sessions using the Child and Adolescent Symptom Inventory-4R, so any contribution to the imaging data could be systematically assessed.

After complete description of the study to participants, written informed consent from parents and written assent from participants was obtained. Fifty-five children were scanned (38 conduct problems, 17 controls), yielding a final sample of usable data from 30 children with conduct problems (15 in each callous-unemotional group) and 16 controls. Exclusions were due to: excessive motion (5 with conduct problems, 1 control), scanner refusal (2 with conduct problems), and technical problems (1 with conduct problems). Group assignment based on callous-unemotional traits took place after exclusions, based on the median for the final sample. See Table 1 for participant data.

*****************************Table 1******************************

Experimental Task: The task was based on backward masking methods used previously to elicit amygdala response to pre-attentively presented stimuli in healthy adults (21, 28). Stimuli comprised fearful and calm faces of 6 individuals taken from the NimStim (29); (3 male, 3 female). Calm (not neutral) faces were used, as previous studies suggest that children with conduct problems may interpret neutral faces as hostile (7). Image size was standardized, and all faces were presented in greyscale with hair cropped. Stimuli were presented on a mid-grey background in 20 blocks; 10 fear, 10 calm, each lasting 15 seconds. Block order was randomized, with the constraint that the same block type was never presented more than twice in a row. A fixation cross was displayed for 15 seconds after every second block.

Each block consisted of 30 trials comprising a target face presented for 17ms, followed by a backward mask face presented for 183ms. **The subjective experience is of seeing the backward masked face only, with the target face presented below the level of conscious awareness (pre-attentively).** A crosshair ISI was presented for 300ms at the centre of the screen, with the centre of the cross approximating the centre of the nose of the target and mask faces. Each trial lasted 500ms. The only difference between fear and calm blocks was that target (masked) faces were either fearful or calm. All mask stimuli were calm faces. Presentation of the target face for 1 frame (17ms) was verified with a high-speed video camera (Casio EX-FH25) set to capture 1000 frames per second.

For each block, the 30 trials comprised 5 presentations of each of the 6 target faces in a pseudorandom order, with each target face (fear or calm) masked by each of the other 5 individuals' calm faces. The task lasted 7.5 mins and comprised 600 trials (300 fear, 300 calm). During the task participants were asked to keep their eyes fixed on the central cross and attend to the faces (passive viewing). Participants were monitored by video link to ensure alertness. Aftewards, participants were asked what they had seen. Three participants mentioned seeing emotion, although none explicitly mentioned fear. Removing these participants from the analysis did not alter the results, and their data were retained in the final sample.

Psychometric and questionnaire measures: Participants completed the two-subtest version of the Wechsler Abbreviated Scale of Intelligence (30) and the Alcohol/Drug Use Disorder Identification Tests (31,32). A parent/guardian also completed the Child and Adolescent Symptom Inventory-4R scales for attention deficit-hyperactivity disorder, generalized anxiety disorder and major depressive episode to ascertain symptom counts for disorders most commonly co-morbid with conduct problems (Table 1). Group differences were found for all symptoms, and were controlled for in subsidiary analyses. They were not included as covariates in the main analysis since a strong case has been made that when participants are not randomly assigned to groups, it is inappropriate to covary for variables intrinsically related to grouping assignment (33).

fMRI data acquisition: A Siemens Avanto 1.5T MRI scanner was used to acquire a 5.5 min 3D T1-weighted structural scan, and multislice T2*-weighted echo planar volumes with BOLD contrast. The EPI sequence was designed to optimize signal detection and reduce dropout in orbitofrontal cortex and amygdala, based on (34), and used the following acquisition parameters: 35 2mm slices acquired in an ascending trajectory with a 1mm gap, TE=50ms; TR=2975ms; slice tilt=-30° (T>C); flip angle=90°; field of view=192mm; matrix size=64x64. Functional data were acquired in a single run of 7.5mins, with 158 volumes per run. Fieldmaps (phase and magnitude images) were also acquired for use in the unwarping stage of data pre-processing.

fMRI data analysis: Imaging data were analysed using SPM8 (www.fil.ion.ucl.ac.uk/spm). Data pre-processing followed a standard sequence: the first five volumes were discarded; data were realigned; unwarped using a fieldmap; normalized with a voxel size of 2x2x2mm; and smoothed with an 8mm Gaussian filter. A block analysis compared neural responses associated with masked fearful and calm faces. Three regressors, each comprising 10 15-second blocks of Fear, Calm and Fixation, were modelled as boxcar functions convolved with a canonical haemodynamic response function. The six realignment parameters were modelled as effects of no interest. For 10 participants (two controls, four in each conduct problem group) an extra regressor was included to model a small number of corrupted images resulting from excessive motion. These images (≤10% of each participant's data) were removed and the adjacent images interpolated in order to prevent distortion of the between-subjects mask. Data were high-pass filtered at 128 s to remove low-frequency drifts.

First-level contrast images for Fear-Calm for each participant were entered into second-level analyses. Based on our prediction that amygdala responses to Fear-Calm would vary by group, a regression analysis was conducted with groups coded as 1, 0 and -1 (1=conduct problems with low callous-unemotional traits; 0=controls; -1=conduct problems with high callous-unemotional traits). A *t*-contrast of 1 was used to look for regions showing a linear relationship across groups in the predicted direction, and -1 for the reverse direction. To explore dimensional associations between callous-unemotional traits and amygdala response to masked fear within the conduct problems group, an additional regression analysis was conducted in which individuals' callous-unemotional trait scores were regressed against neural responses to Fear-Calm.

We report data from the amygdala region of interest in the main text. For completeness, we also report results from whole brain analyses at $p<.001$, uncorrected, $k\geq5$ in the Supplemental Data. The amygdala region of interest was defined both structurally (the bilateral Talairach Daemon amygdala mask supplied by WFU PickAtlas (35) and functionally (an 8mm sphere centred on the peak co-ordinate [18 -6 -18] for masked fear>masked happy reported by (21), converted from Talairach to MNI co-ordinates).

Results

A cluster showing the predicted pattern (conduct problems with low callous-unemotional traits>controls>conduct problems with high callous-unemotional traits) was found in the right amygdala at $p<.001$, uncorrected (peak voxel=[20 -2 -22], t=3.85, z=3.55, k=9, Figure 1). The whole cluster survived small-volume correction using both the structurally and functionally defined amygdala regions of interest ($p<.05$ familywise-error corrected at both voxel and cluster levels). This finding also remained significant with familywise-error correction when controlling for variables on which the groups differed (conduct disorder, attention deficit-hyperactivity disorder, anxiety, and depression symptoms; Table S2). For completeness, a list of all clusters showing the predicted pattern for Fear-Calm, and the reverse, is displayed in Table S3. As no regions survived whole-brain correction, these data are not discussed further.

********************************Figure 1********************************

Planned *t*-tests were conducted using mean responses across the right amygdala cluster. One-sample *t*-tests comparing responses to Fear-Calm in each group revealed a significant positive difference in children with conduct problems and low callous-unemotional traits (*t*(14)=2.82, p=.**014**), no significant difference between conditions in controls (*t*(15)=-.85, p=.**41**), and a significant negative difference in children with conduct problems and high callous-unemotional traits (*t*(14)=-2.30, *p*=.0**37**). Across groups, responses to Fear-Calm in children with conduct problems and low callous-unemotional traits were significantly greater than both controls (*t*(29)=2.49, *p*=.0**19**) and children with conduct problems and high callous-unemotional traits (*t*(28)=3.46, *p*=.00**2**). **The difference between controls and children with conduct problems and high callous-unemotional traits was not significant (*t*(29)=1.38, *p*=.18).**

A separate regression analysis within the conduct problems group investigated the association between dimensional callous-unemotional trait scores and neural responses to pre-attentively presented fear (relative to calm). At a whole-brain uncorrected threshold of *p*=.001, one voxel in the right amygdala showed a significant negative relationship with callous-unemotional trait scores ([24 -2 -18], *t*=3.38, z=3.07; Figure 2). This voxel also survived small volume correction (*p*<.05 with familywise-error correction) using the regions of interest defined both anatomically and functionally as described above. Other regions showing negative or positive relationships with callous-unemotional trait scores are listed in Table S4. None survived whole-brain correction, and these data are not discussed further.

*******************************Figure 2*******************************

Discussion

We employed a backward masking paradigm (21) to investigate differential amygdala activity to pre-attentively presented fear in children with conduct problems, and found significantly lower amygdala activity to backwardly masked fearful versus calm faces in children with high compared with low callous-unemotional traits. Amygdala activity in the control group was intermediate between the conduct problems groups. These data are the first to demonstrate differential amygdala activity to pre-attentively presented fear across the spectrum of callous-unemotional traits in children with conduct problems. Our findings indicate that reduced amygdala activation to salient stimuli in children with conduct problems and high callous-unemotional traits extends to early stages of information processing, suggestive of an affective processing deficit in this group. Reduced amygdala activation is characteristic of this sub-group, rather than conduct problems more generally. Our finding adds to increasing evidence regarding the utility of callous-unemotional specifier in the classification of children with conduct problems.

In a previous study we reported a negative association between callous-unemotional traits and amygdala activity using an explicit and complex affective processing task (17). The current study demonstrated a negative association between callous-unemotional traits and amygdala response to pre-attentively presented fear, further highlighting the dimensional relationship between callous-unemotional traits and amygdala activity. This finding is consistent with a recent behavioral study demonstrating reduced pre-attentive processing of negative emotions in individuals with high callous-unemotional traits (22).

Lesion studies indicate an important role for amygdala in at least some aspects of pre-attentive processing of salient stimuli (see (11) for a review). For example, reduced reflexive gaze orientation to fearfully widened eyes is seen in amygdala patients (36). It has been proposed that amygdala dysfunction may interfere with the initial processing of salient facial features (e.g. widened fearful eyes), which typically trigger attentional shifts (36, 37). Studies of healthy adults have found that displays of fearful eye whites are sufficient to elicit amygdala activation (38) and that extent of amygdala activation to emotional faces positively correlates with degree of fixation to the eye region (37). Our data suggest that children with conduct problems and high callous-unemotional traits show reduced amygdala activity to pre-attentively presented salient facial information that could compromise their orienting to critical affective cues relevant for social interaction. Recent behavioral and eye-tracking data are in line with this possibility (7, 39). Under free-viewing conditions, children with high callous-unemotional traits have difficulty recognising fearful expressions and focus less on the eye-region of the face than children with low callous-unemotional traits. However, when asked to effortfully focus on the eye-region of the face, fear recognition performance improves.

It is important to acknowledge that the amygdala is part of a network that triggers an orienting response (37). Future paradigms, sensitive to exploring functional connectivity between amygdala and other brain regions, will be informative. We also note that the control group did not show increased amygdala response to pre-attentively presented fear. Some studies of healthy adults using masked stimuli have also failed to find robust amygdala response (40), suggesting that there may be individual differences in response to pre-attentively presented stimuli. Critically, in the present study the task was sensitive enough to elicit differences between conduct problem subgroups based on callous-unemotional traits. Additionally our group difference finding was right-lateralized, in line with most previous effects reported for masked fear stimuli (40).

Several limitations of the present study must be acknowledged. Our sample was selected using a research diagnosis: replication in a sample of youth with a clinical diagnosis would be of interest. In addition, we studied only males; it is unknown whether girls with conduct problems show a similar pattern. The use of a passive viewing task means that it was not possible to delineate specific computations contributing to activation differences between groups. Future imaging studies, including more temporally sensitive methods, could explore the time-course of amygdala activity and connectivity with other brain regions. Finally, the current study was cross-sectional. Longitudinal studies will be necessary to chart possible changes in patterns of neural responses associated with conduct problems.

Despite these limitations, the current study extends our understanding of the neural correlates of conduct problems. We show that right amygdala responses to pre-attentively presented fear differentiate children with conduct problems and varying levels of callous-unemotional traits, with significantly greater responses in those with low relative to high levels of these traits. This mirrors findings from studies using explicitly presented affective stimuli, and additionally suggests that altered amygdala responses characterize the very earliest stages of affective processing. In children with conduct problems and high callous-unemotional traits, an attenuated amygdala response to pre-attentive fearful faces may reduce orienting to salient features of these stimuli, reducing opportunities for learning from these important social cues. Conversely, heightened pre-attentive amygdala responses in children with conduct problems and low callous-unemotional traits may predispose these children to affective hypervigilance. Our regression analysis also contributes to existing data indicating a dimensional relationship between callous-unemotional traits and amygdala response.

From a clinical perspective, divergent patterns of amygdala response to pre-attentively presented emotion point to differential underlying neural vulnerabilities in conduct problem subgroups. This may have important implications for how we formulate and intervene in conduct disorder. Specifically, it may be important to evaluate the therapeutic efficacy of helping children with conduct problems develop a more balanced appraisal of other people’s emotions. This may include a process of explicit verbalisation. This kind of approach has already been shown to be effective in Attention Bias Modification Treatment for anxiety disorder (41). In addition, clinically focused intervention studies that investigate how treatment response relates to a child's level of callous-unemotional traits, will help us better understand the variation in treatment response seen in this group.

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References

1 Odgers CL, Caspi A, Broadbent JM, Dickson N, Hancox RJ, Harrington H, Poulton R, Sears MR, Thomson WM, Moffitt TE: Prediction of differential adult health burden by conduct problem subtypes in males. Arch Gen Psychiatry 2007; 64:476-484.

2 Frick PJ, Viding E: Antisocial behavior from a developmental psychopathology perspective. Dev Psychopathol 2009; 21:1111-1131.

3 Romeo R, Knapp M, Scott S: Economic cost of severe antisocial behaviour in children--and who pays it. Br J Psychiatry 2006; 188:547-553.

4 Viding E, Blair RJ, Moffitt TE, Plomin R: Evidence for substantial genetic risk for psychopathy in 7-year-olds. J Child Psychol Psychiatry 2005; 46:592-597.

5 Viding E, Jones AP, Frick PJ, Moffitt TE, Plomin R: Heritability of antisocial behaviour at 9: do callous-unemotional traits matter? Dev Sci 2008; 11:17-22.

6 Blair RJ: Neuroimaging of psychopathy and antisocial behavior: a targeted review. Curr Psychiatry Rep 2010; 12:76-82.

7 Dadds MR, Perry Y, Hawes DJ, Merz S, Riddell AC, Haines DJ, Solak E, Abeygunawardane AI: Attention to the eyes and fear-recognition deficits in child psychopathy. Br J Psychiatry. 2006; 189:280-281.

8 Kahn RE, Frick PJ, Youngstrom E, Findling RL, Youngstrom JK: The effects of including a callous-unemotional specifier for the diagnosis of conduct disorder. J Child Psychol Psychiatry 2011; 10-7610.

9 Viding E, McCrory EJ, Blakemore SJ, Frederickson N: Behavioural problems and bullying at school: can cognitive neuroscience shed new light on an old problem? Trends Cogn Sci 2011; 15:289-291.

10 Sterzer P, Stadler C: Neuroimaging of aggressive and violent behaviour in children and adolescents. Front Behav Neurosci 2009; e-pub ahead of print 12 October 2009; doi:10.3389/neuro.08.035.2009.

11 Adolphs R: Fear, faces, and the human amygdala. Curr Opin Neurobiol 2008; 18:166-172.

12 Herpertz SC, Huebner T, Marx I, Vloet TD, Fink GR, Stoecker T, Shah NJ, Konrad K, Herpertz-Dahlmann B: Emotional processing in male adolescents with childhood-onset conduct disorder. J Child Psychol Psychiatry 2008; 49:781-791.

13 Passamonti L, Fairchild G, Goodyer IM, Hurford G, Hagan CC, Rowe JB, Calder AJ: Neural abnormalities in early-onset and adolescence-onset conduct disorder. Arch Gen Psychiatry. 2010; 67:729-738.

14 Sterzer P, Stadler C, Krebs A, Kleinschmidt A, Poustka F: Abnormal neural responses to emotional visual stimuli in adolescents with conduct disorder. Biol Psychiatry 2005; 57:7-15.

15 Jones AP, Laurens KR, Herba CM, Barker GJ, Viding E: Amygdala hypoactivity to fearful faces in boys with conduct problems and callous-unemotional traits. Am J Psychiatry 2009; 166:95-102.

16 Marsh AA, Finger EC, Mitchell DG, Reid ME, Sims C, Kosson DS, Towbin KE, Leibenluft E, Pine DS, Blair RJ: Reduced amygdala response to fearful expressions in children and adolescents with callous-unemotional traits and disruptive behavior disorders. Am J Psychiatry 2008; 165:712-720.

17 Sebastian CL, McCrory EJP, Cecil CAM, Lockwood PL, De Brito SA, Fontaine NMG, Viding E: Neural responses to affective and cognitive Theory of Mind in children with conduct problems and varying levels of callous-unemotional traits. Archives of General Psychiatry (in press).

18 Blair RJ, Viding E: Psychopathy, in Rutter's Child and Adolescent Psychiatry. Edited by Rutter M, Bishop D, Pine D, Scott S, Stevenson J, Taylor E, Thapar A. Oxford, UK, Blackwell Publishing Ltd, 2008.

19 Ohman A: Evolutionary preparedness for preattentive processing of threat., in Attention and orienting: Sensory and motivational processes. Edited by Lang PJ, Simons RF, Balaban M. Mahwah, NJ, Erlbaum, 2008.

20 Pessoa L, Adolphs R: Emotion processing and the amygdala: from a 'low road' to 'many roads' of evaluating biological significance. Nat Rev Neurosci 2010; 11:773-783.

21 Whalen PJ, Rauch SL, Etcoff NL, McInerney SC, Lee MB, Jenike MA: Masked presentations of emotional facial expressions modulate amygdala activity without explicit knowledge. J Neurosci 1998; 18:411-418.

22 Sylvers PD, Brennan PA, Lilienfeld SO: Psychopathic traits and preattentive threat processing in children: a novel test of the fearlessness hypothesis. Psychol Sci 2011; 22:1280-1287.

23 Gadow KD, Sprafkin J: The Symptom Inventories: An Annotated Bibliography. Stony Brook, NY, Checkmate Plus, 2009.

24 Essau CA, Sasagawa S, Frick PJ: Callous-unemotional traits in a community sample of adolescents. Assessment 2006; 13:454-469.

25 Piacentini JC, Cohen P, Cohen J: Combining discrepant diagnostic information from multiple sources: are complex algorithms better than simple ones? J Abnorm Child Psychol 1992; 20:51-63.

26 Goodman R: The Strengths and Difficulties Questionnaire: a research note. J Child Psychol Psychiatry 1997; 38:581-586.

27 Sprafkin J, Gadow KD: Adolescent Symptom Inventory-4 Norms Manual. Stony Brook, NY, Checkmate Plus, 1998.

28 Kim MJ, Loucks RA, Neta M, Davis FC, Oler JA, Mazzulla EC, Whalen PJ: Behind the mask: the influence of mask-type on amygdala response to fearful faces. Soc Cogn Affect Neurosci 2010; 5:363-368.

29 Tottenham N, Tanaka JW, Leon AC, McCarry T, Nurse M, Hare TA, Marcus DJ, Westerlund A, Casey BJ, Nelson C: The NimStim set of facial expressions: judgments from untrained research participants. Psychiatry Res 2009; 168:242-249.

30 Weschler D: Wechsler Abbreviated Scale of Intelligence (WASI). San Antonio, TX, Harcourt Assessment, 1999.

31 Babor TF, Higgins-Biddle JC, Saunders JB, Monteiro MG: The Alcohol Use Disorders Identification Test guidelines for use in primary care. World Health Organisation, Department of Mental Health and Substance Dependence, 2001.

32 Berman AH, Bergman H, Palmstierna T, Schlyter F: Evaluation of the Drug Use Disorders Identification Test (DUDIT) in criminal justice and detoxification settings and in a Swedish population sample. Eur Addict Res 2005; 11:22-31.

33 Miller GA, Chapman JP: Misunderstanding analysis of covariance. J Abnorm Psychol 2001; 110:40-48.

34 Weiskopf N, Hutton C, Josephs O, Deichmann R: Optimal EPI parameters for reduction of susceptibility-induced BOLD sensitivity losses: a whole-brain analysis at 3 T and 1.5 T. Neuroimage 2006; 33:493-504.

35 Maldjian JA, Laurienti PJ, Kraft RA, Burdette JH: An automated method for neuroanatomic and cytoarchitectonic atlas-based interrogation of fMRI data sets. Neuroimage 2003; 19:1233-1239.

36 Adolphs R, Tranel D, Buchanan TW: Amygdala damage impairs emotional memory for gist but not details of complex stimuli. Nat Neurosci 2005; 8:512-518.

37 Gamer M, Buchel C: Amygdala activation predicts gaze toward fearful eyes. J Neurosci 2009; 29:9123-9126.

38 Whalen PJ, Kagan J, Cook RG, Davis FC, Kim H, Polis S, McLaren DG, Somerville LH, McLean AA, Maxwell JS, Johnstone T: Human amygdala responsivity to masked fearful eye whites. Science 2004; 306:2061.

39 Dadds MR, El Masry Y, Wimalaweera S, Guastella AJ: Reduced eye gaze explains "fear blindness" in childhood psychopathic traits. J Am Acad Child Adolesc Psychiatry 2008; 47:455-463.

40 Costafreda SG, Brammer MJ, David AS, Fu CH: Predictors of amygdala activation during the processing of emotional stimuli: a meta-analysis of 385 PET and fMRI studies. Brain Res Rev 2008; 58:57-70.

41 Hakamata Y, Lissek S, Bar-Haim Y, Britton JC, Fox NA, Leibenluft E, Ernst M, Pine DS. Attention bias modification treatment: a meta-analysis toward the establishment of novel treatment for anxiety. Biol Psychiatry 2010; 68:982-90.

Table and Figure Legends

Table 1: Participant characteristics (means, SDs) presented by group.

Figure 1: The region of the right amygdala showing the pattern: conduct problems with low callous-unemotional traits>controls>conduct problems with high callous-unemotional traits for the contrast Fear-Calm. The overlay shows the cluster at *p*<.001, uncorrected; and the bars represent mean responses across this cluster (k=9). The cluster also survived small volume correction (*p*<.05, familywise error corrected).

Figure 2: Scatterplot showing the continuous relationship between right amygdala response to Fear-Calm and callous-unemotional traits within the conduct problems group. One voxel ([24 -2 -18], *t*=3.38) was significant at p=.001 (uncorrected), and survived small volume correction (*p*<.05, familywise error corrected).

Table 1

	Group			
Measure	Control group (N=16)		Conduct problems/ Low callous-unemotional traits (N=15)	
	Mean	SD	Mean	SD
Age[b]	13.73	1.37	14.70	1.53
Socio-Economic Status[b]	2.78	0.84	2.67	1.16
Full IQ score from 2-subtest WASI[c]	108.44	10.30	103.73	11.36
Ethnicity[b,e]	15:1	-	10:3:2	-
Handedness[b,f]	11:4:1	-	10:5	-
Inventory of Callous-Unemotional Traits[d]	24.56	5.50	34.73	8.16
Child and Adolescent Symptom Inventory				
Conduct Disorder[d]	0.44	0.73	7.85	3.74
Attention Deficit Hyperactivity Disorder[g]	10.13	5.98	21.53	11.80
Generalised Anxiety Disorder[g]	3.75	3.19	7.22	4.59
Major Depressive Episode[g,h]	2.75	1.98	5.40	2.92
Alcohol Use and Disorders[c]	1.25	1.73	4.40	5.88
Drug Use and Disorders[c]	0.00	0.00	2.60	5.59

* $p < .05$, Bonferroni corrected

[a] All p-values obtained using t-tests except for Ethnicity and Handedness (Fisher's exact tests use

[b] Measures taken at screening phase, parent report

[c] Child at scanning session

[d] Measures taken at screening phase, parent and teacher report

[e] White:Black:Mixed

[f] Right:Left:Ambidextrous

[g] Measures taken at scanning session - parent report

[h] Missing data from 1 participant with conduct problems

Conduct problems/ High callous-unemotional traits (N=15)		Analysis	
Mean	SD	p^a	Post Hoc*
14.22	1.93	.261	
3.27	1.06	.234	
98.80	12.08	.069	
12:1:2	-	.339	
13:2	-	.503	
53.47	5.50	.001	1<2<3
13.88	7.04	.001	1<2<3
31.40	9.39	.001	1<2<3
8.48	5.16	.012	1<3
5.71	3.31	.009	1<2<3
5.07	7.40	.128	
1.13	2.70	.136	

d)

Figure 1

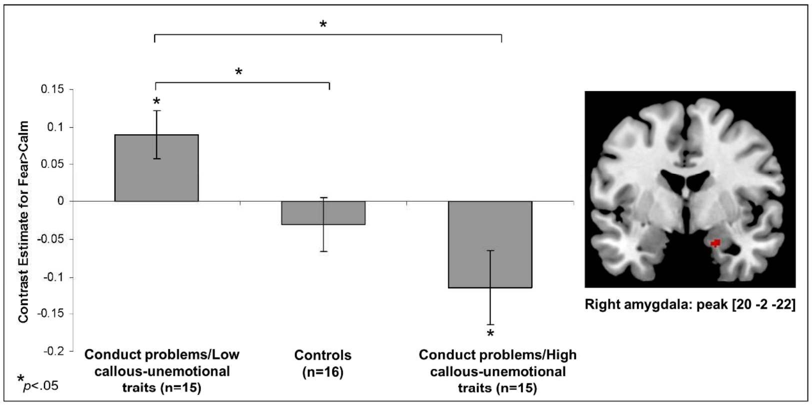

Figure 1: The region of the right amygdala showing the pattern: conduct problems with low callous-unemotional traits>controls>conduct problems with high callous-unemotional traits for the contrast Fear-Calm. The overlay shows the cluster at p<.001, uncorrected; and the bars represent mean responses across this cluster (k=9). The cluster also survived small volume correction (p<.05, familywise error corrected).
254x190mm (300 x 300 DPI)

Figure 2

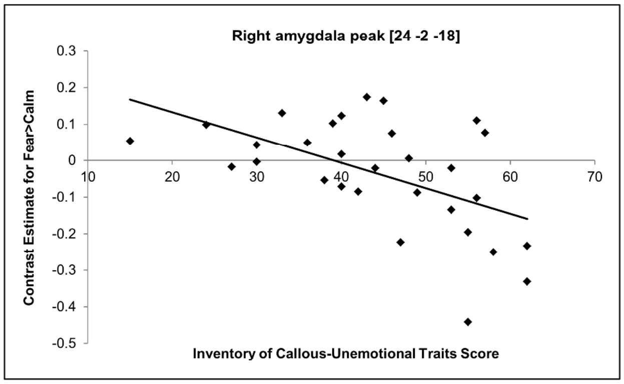

Figure 2: Scatterplot showing the continuous relationship between right amygdala response to Fear-Calm and callous-unemotional traits within the conduct problems group. One voxel ([24 -2 -18], t=3.38) was significant at p=.001 (uncorrected), and survived small volume correction (p<.05, familywise error corrected).
254x190mm (300 x 300 DPI)

Supplemental Data for 'Amygdala response to pre-attentive masked fear is associated with callous-unemotional traits in children with conduct problems.'

Supplementary Results Tables

Table S1: Combined parent and teacher ratings for the Strengths and Difficulties Questionnaire, taken during participant screening.

	Group							
Strengths and Difficulties Questionnaire	1) Control group (n=16)		2) Conduct problems/Low callous-unemotional traits (n=15)		3) Conduct problems/High callous-unemotional traits (n=15)		Analysis	
	Mean	SD	Mean	SD	Mean	SD	p	Post-hoc*
Conduct Problems	1.06	1.24	5.93	1.39	8.20	1.47	.001	1<2<3
Hyperactivity/Inattention	3.81	2.20	7.53	2.17	8.80	1.42	.001	1<(2,3)
Peer Relationship Problems	2.31	1.25	3.27	2.96	4.67	2.97	.041	1<3
Emotional symptoms	2.25	1.57	4.00	3.21	4.73	2.69	.028	1<3
Pro-social behaviour	9.56	0.63	7.47	1.81	6.40	2.16	.001	1<(2,3)
Total difficulties	9.44	3.69	20.73	6.24	26.80	6.34	.001	1<2<3

**p* <.05, Bonferroni corrected

Table S2: The effect of controlling for participant symptom counts on the cluster reported in the main text in right amygdala showing the pattern: conduct problems with low callous-unemotional traits>controls>conduct problems with low callous-unemotional traits. Voxels displayed survive familywise error correction at *p*<.05 within a bilateral amygdala mask.

Covariate (symptom counts, N=46)	Peak amygdala voxel			t	z	k
No Covariate	20	-2	-22	3.85	3.55	9
Conduct Disorder	20	-2	-22	3.97	3.65	9
Attention Deficit Hyperactivity Disorder	20	-2	-22	4.24	3.85	16
Generalised Anxiety Disorder	20	-2	-22	4.05	3.70	14
Major Depressive Episode[a]	20	-2	-22	3.60	3.34	4

[a] Missing data from one participant with conduct problems

x y z=peak voxel MNI co-ordinates; k=cluster size (2x2x2mm voxels).

Table S3: Significant clusters at *p*<.001, uncorrected, *k*≥5, showing the pattern conduct problems with low callous-unemotional traits>controls>conduct problems with low callous-unemotional traits, and the reverse, for Fear-Calm.

Brain Region	BA	L/R	x	y	z	peak t	peak z	k
Conduct problems/low callous-unemotional traits>Controls>Conduct problems/high callous-unemotional traits								
Cerebellum, posterior lobe	-	L	-28	-70	-22	4.34	3.94	142
Amygdala	-	R	20	-2	-22	3.85	3.55	9
Occipital cortex	18	R	18	-84	-16	3.62	3.36	6
Dorsolateral prefrontal cortex	46	R	48	38	26	3.37	3.16	5
Middle frontal gyrus	8	L	-40	24	44	3.37	3.16	5
Conduct problems/low callous-unemotional traits<Controls<Conduct problems/high callous-unemotional traits								
Middle temporal gyrus	20	R	40	-18	-20	4.55	4.10	31
Precentral gyrus	6	L	-38	-12	40	4.21	3.84	73
Uncus	20	R	30	-10	-32	3.95	3.64	33
Cerebellum, anterior lobe	-	R	14	-40	-30	3.65	3.39	10
Superior frontal gyrus	9	L	-16	34	40	3.63	3.37	16
Mid-cingulate gyrus	24	R	12	8	34	3.61	3.36	8
Fusiform gyrus	20	L	-36	-8	-28	3.61	3.36	9

BA=Brodmann Area; L/R=Left/Right; x y z=peak voxel MNI co-ordinates; k=cluster size (2x2x2mm voxels).

Table S4: Significant clusters at $p<.001$, uncorrected, $k\geq 5$, showing a correlation between callous-unemotional trait scores and neural responses to Fear-Calm in children with conduct problems.

Brain Region	BA	L/R	x	y	z	peak t	peak z	k
Negative Relationship								
Caudate tail	-	R	22	-44	12	5.21	4.32	32
Middle frontal gyrus	46	R	46	38	28	4.14	3.62	34
Occipital cortex	19	L	-36	-82	-4	4.12	3.61	73
			-24	-88	-12	4.09	3.59	
Occipital cortex	17	L	-20	-82	4	4.02	3.54	7
Claustrum	-	L	-34	-24	-6	3.82	3.40	9
Cerebellum, posterior lobe	-	L	-30	-70	-22	3.78	3.37	44
			-38	-72	-18	3.56	3.20	
Positive Relationship								
Parahippocampal gyrus	28	R	28	-10	-32	4.10	3.60	14
Inferior temporal gyrus	21	R	42	-8	-34	4.00	3.53	11

BA=Brodmann Area; L/R=Left/Right; x y z=peak voxel MNI co-ordinates; k=cluster size (2x2x2mm voxels).