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Archival Report

Deficient Amygdala Habituation to Threatening Stimuli in Borderline Personality Disorder Relates to Adverse Childhood Experiences

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ABSTRACT

BACKGROUND: Heightened amygdala response to threatening cues has been repeatedly observed in borderline personality disorder (BPD). A previous report linked hyperactivation to deficient amygdala habituation to repeated stimuli, but the biological underpinnings are incompletely understood.

METHODS: We examined a sample of 120 patients with BPD and 115 healthy control subjects with a well-established functional magnetic resonance imaging emotional face processing task to replicate the previously reported amygdala habituation deficit in BPD and probed this neural phenotype for associations with symptom severity and early social risk exposure.

RESULTS: Our results confirm a significant reduction in amygdala habituation to repeated negative stimuli in BPD (p_{FWE} = .015, peak-level familywise error [FWE] corrected for region of interest). Post hoc comparison and regression analysis did not suggest a role for BPD clinical state ($p_{FWE} > .56$) or symptom severity ($p_{FWE} > .45$) for this phenotype. Furthermore, deficient amygdala habituation was significantly related to increased exposure to adverse childhood experiences (p_{FWE} = .013, region of interest corrected).

CONCLUSIONS: Our data replicate a prior report on deficient amygdala habituation in BPD and link this neural phenotype to early adversity, a well-established social environmental risk factor for emotion dysregulation and psychiatric illness.

Keywords: Borderline personality disorder, Early adversity, Emotion processing, Functional neuroimaging, Habituation, Psychiatry

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Borderline personality disorder (BPD) is a common, debilitating, and costly psychiatric illness characterized by affective instability, impulsivity, and social impairments (1,2). Accumulating evidence supports the hypothesis of a central role of affective instability in the pathophysiology of BPD, which gives rise to a variety of downstream symptoms including intense negative emotions, inner tension, impulsivity, volatility of mood, and social deficits such as rejection sensitivity and lack of cooperation (1,3,4). Current etiological models of BPD emphasize an interaction between genetic risk factors and adverse social exposures during childhood and adolescence (5,6). In search of the underlying neural pathomechanisms for affective dysregulation, several small-sample studies used functional magnetic resonance imaging (fMRI) to study neural reactivity to threatening visual stimuli in BPD, especially to consecutively presented aversive pictures or negative facial expressions. Here, a significant increase in the mean response amplitude of the amygdala has been frequently observed, consistent with the idea of abnormally enhanced limbic processing of negative emotional stimuli in BPD (7–11).

Other work emphasizes the considerable variability in amygdala reactivity to threatening stimuli over the course of an fMRI experiment. This may be especially relevant for the high instability of emotional responses in BPD. Specifically, the rapid decline of amygdala responsivity to repeated emotional presentations is a well-established phenomenon related to neural habituation (12–14), an evolutionarily conserved plasticity mechanism tailoring innate responses toward salient stimuli with behavioral significance (13,15). Furthermore, researchers have cautioned that the between-group comparison of amygdala mean evoked amplitudes may lead to invalid neural mechanistic inferences (14,16), because an apparent overall increase in amygdala response to threatening cues may in fact relate to the absence of the natural signal decrement to repeated emotional stimulus presentations (i.e., a habituation deficit). Given the dynamic of amygdala reactivity, it is not surprising that the retest reliability of amygdala habituation

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indices is significantly higher than that of the mean response amplitude (17,18).

Despite the clinical and biological relevance of the phenomenon, only few fMRI studies have examined the question of whether deficient amygdala habituation to threatening stimuli plays a role in BPD. Hazlett *et al.* (19) modeled emotional picture repetitions over time and confirmed the abnormally sustained amygdala response amplitudes to successive stimulation in BPD. The researchers interpreted this result to indicate a plausible neural pathomechanism for the unusually intense and prolonged affective responses of BPD patients in clinical contexts (19). However, this finding still awaits independent confirmation in a larger-scale study, and the biological underpinnings remain to be clarified. Specifically, it is unclear whether the identified amygdala habituation phenotype reflects a neural mechanism for affective instability in BPD related to symptom severity or psychiatric risk. Given that, first, changes in mean amygdala reactivity *across* fMRI scans have been associated with differences in the BPD clinical state (20) and, second, deficient amygdala habituation to repeated emotional stimuli *within* an fMRI scan has been linked to genetic risk variants (21,22), the amygdala habituation phenotype may reflect a psychiatric risk–related neural mechanism.

In the present study we aimed to extend prior neuroimaging efforts in BPD to the examination of a large cohort of patients and healthy control subjects (HCs) using fMRI and a well-established facial emotion matching paradigm (23) known for yielding reliable amygdala habituation estimates (16). Specifically, we aimed to 1) confirm the previously identified within-scan amygdala habituation deficit in BPD, 2) test the potential relationship of this phenotype to symptom severity in BPD patients with acute and remitted symptoms, and 3) investigate the potential relationship of the amygdala habituation phenotype to adverse childhood experiences, a well-established social risk factor for BPD.

METHODS AND MATERIALS

Participants

We examined 235 individuals, 120 patients with BPD (29.94 ± 6.6 years [mean age ± SD], 111 women) and 115 HCs without a history of mental illness (28.43 ± 7.3 years, 96 women). All participants provided written informed consent for a protocol approved by the Ethics Committee of the University of Heidelberg. Participants were recruited through the central project of the Clinical Research Unit 256 through flyers, advertisement on social media/websites, and in newspapers. Before the study subjects underwent thorough screening and interviewing, conducted by M.Sc.-level or above psychologists or medical doctors extensively trained for diagnostics.

In patients, DSM-IV Axis I and II disorders were assessed by the Structured Clinical Interview for DSM-IV (24) and the International Personality Disorder Examination (25). Among the patients, 98 had a diagnosis of current BPD (BPD-C, defined as currently fulfilling 5 or more DSM-IV criteria), whereas 22 had a lifetime diagnosis of BPD but were currently in remission (BPD-R, defined as fulfilling a maximum of 3 DSM-IV criteria in the preceding 2 years).

HCs were recruited from the general population in and around Mannheim (Germany). All HCs underwent Structured Clinical Interview for DSM-IV screening after recruitment. From these individuals, 51 further underwent the full Structured Clinical Interview for DSM-IV Axis I Disorders and International Personality Disorder Examination assessment to confirm the absence of a lifetime history of psychiatric disorders or substance dependency.

General exclusion criteria were MRI contraindications, a history of head trauma or neurological illness, and current alcohol abuse or other drug use. BPD patients were excluded for substance dependency for 1 year before participation or a lifetime diagnosis of schizophrenia or bipolar disorder. Current use of selective serotonin reuptake inhibitors, but no other drug use, was tolerated in patients (BPD-C, n = 15).

Psychological and Clinical Assessments

We used the Raven Advanced Progressive Matrices test (26) to assess general intelligence and the Childhood Trauma Questionnaire (CTQ) (27) to assess retrospective self-ratings of the severity of abuse and neglect during childhood. In individuals with confirmed BPD diagnosis, severity of clinical impairment was assessed using German versions of the Borderline Symptom List (BSL) (28), Dissociative Experience Scale (29), Non-Suicidal Self-Injury Scale, Global Assessment of Functioning Scale (Axis V in DSM-IV) (30), Beck Depression Inventory (31), and the Barratt Impulsiveness Scale (32). Shortly before the fMRI scan, all subjects completed the self-assessment manikin for quantification of current subjective arousal and inner tension, dominance, and valence (33).

Emotional Face Matching Task

Participants completed a well-established emotional face matching paradigm during fMRI designed to engage implicit emotion processing (23). The task is known for robustly activating the amygdala (17) and providing reliable measures of amygdala habituation (16). Experimental stimuli consisted of the visual presentation of face stimuli from the Facial Action Coding System (34) showing angry and fearful expressions. Control stimuli were simple geometric shapes (circles, horizontal ellipses, vertical ellipses). The task comprised 8 blocks of 30 seconds each, with blocks alternating between experimental and control conditions. Within each block, each trial was presented for 5 seconds. In each trial 3 stimuli were simultaneously presented, and the task was to correctly match, via button-press, 1 of 2 images displayed either on the left or right to a target image located above. Total duration of the task was 4 minutes 34 seconds.

fMRI Data Acquisition and Processing

Blood oxygen level–dependent fMRI was performed on 2 identical 3T MRI scanners (Siemens Trio, Erlangen, Germany) using an echo planar imaging sequence with the following parameters: repetition time 2000 ms, echo time 30 ms, 130 volumes, 28 oblique slices per volume, 4-mm slice thickness, 1-mm slice distance, 80° flip angle, 192-mm field of view, and 64 × 64 matrix. The fMRI data were processed and analyzed with SPM12 (www.fil.ion.ucl.ac.uk/spm) implemented in

MATLAB (The MathWorks, Inc., Natick, MA; https://www.mathworks.com/products/matlab.html) using standard procedures. Briefly, during preprocessing the images were realigned to the mean image of the scan run using a 6-parameter rigid body spatial transformation, spatially normalized to the standard stereotactic space of the Montreal Neurological Institute template, resampled to 3-mm isotropic voxels, and smoothed with an 8-mm full width at half maximum Gaussian kernel.

fMRI Data Analysis

Amygdala habituation analysis consisted of a 2-level procedure. At the first level we defined a general linear model for each subject including boxcar reference vectors for each task block (convolved with the standard SPM hemodynamic response function) and the head motion parameters from the realignment step. We defined a high-pass filter with a cut-off frequency of 0.001 Hz and used first-order autoregressive modeling to correct for temporal autocorrelations. Voxelwise habituation indices for each subject were quantified as previously described (35) by calculating the mean response amplitude difference between the first and the second half of the face matching condition blocks ([block 1 + block 2] > [block 3 + block 4]). Second-level statistical inference included univariate analyses of variance (ANOVA) models with group (BPD-C, BPD-R, HC) as a factor. Group contrasts were used to test for the effects of diagnosis (BPD-C + BPD-R vs. HC) and BPD clinical state (BPD-C vs. BPD-R). Associations with BPD symptom severity were examined with a univariate ANOVA model with group (BPD-C, BPD-R) as a factor and BSL total score as covariate of interest. In addition, we tested for the potential effects of prescan arousal and inner tension by including self-assessment manikin ratings (arousal subscale) as a covariate of interest. The relationship to childhood adversity was examined with a univariate ANOVA model with group (BPD-C, BPD-R, HC) as a factor and CTQ total scores as covariates of interest. To test for potential group-specific associations between childhood adversity and amygdala habituation, we defined an analogous univariate ANOVA model with an additional interaction term (group × CTQ). We further included sex as a covariate of no interest in all our fMRI analyses because we observed a significant difference in the sex distribution of the HC and BPD-R groups (Table 1).

fMRI Statistical Inference

Statistical significance was assessed at $p < .05$, peak voxel-level familywise error (FWE) corrected (p_{FWE}) for multiple comparisons in an a priori defined mask of the right amygdala derived from the Automated Anatomical Labeling atlas (36). The choice of the right amygdala as region of interest was motivated by prior work demonstrating 1) different functional roles and habituation rates for the left and right amygdala during emotion processing, with the right amygdala specialized to rapid and dynamical detection of emotional stimuli (14,37,38), and 2) considerably higher retest reliabilities of habituation estimates in the right amygdala (16).

RESULTS

Demographic and Psychological Assessments

Univariate ANOVA revealed no group differences in age, education, or intelligence. We detected a significant difference in the sex distribution of the HC and BPD-R groups ($\chi^2 = 4.22$, $p = .040$). CTQ total scores differed significantly in the group comparisons between HC and BPD-C (Games-Howell test, $p < .001$) and HC and BPD-R ($p < .001$), respectively. There was no significant CTQ difference between the BPD-C and BPD-R groups ($p = .24$). Compared with patients with BPD-R, the patients with BPD-C showed a significantly higher severity of clinical symptoms including borderline-typical symptoms, dissociative experiences, and impulsivity (all $p < .001$). Further statistical details on the assessed demographic and psychological variables are provided in Table 1. For details on the clinical symptoms and comorbidities of patients with BPD-C and BPD-R, see Table 2.

fMRI Data Quality Parameters and Task Performance

There were no significant group differences in fMRI data quality parameters including signal-to-noise ratio, spiking, framewise displacement, and mean head motion translation or rotation (all $p > .23$). Likewise, we detected no significant group differences in fMRI task performance parameters including percent correct responses and reaction times (all $p > .29$). Further statistical details are provided in Table 1. Whole-brain results are reported in Supplemental Table S1.

Amygdala Habituation Group Comparisons

Group comparison of patients with BPD and HCs confirmed a significant reduction of right amygdala habituation to repeated negative stimuli in BPD patients ($t = 3.27$, $p_{FWE} = .015$, all p values are peak-level corrected for region of interest) (Figure 1A, left), a finding also surviving correction for bilateral amygdala volume ($t = 3.27$, $p_{FWE} = .028$). Data inspection suggested a rapid decline of right amygdala responsivity to emotional block repetitions in HCs, whereas a comparable decline in amygdala responsivity over time was absent in patients with BPD (Figure 1A, right). This observation was confirmed through subgroup contrasts revealing a significant decline in amygdala responsivity across time in HCs ($t = 4.52$, $p_{FWE} = .0002$) but not in patients with BPD ($t = 0.41$, $p_{FWE} = .749$). No difference in amygdala habituation was evident in the comparison between patients with BPD-C and BPD-R (all $t < 1.20$, $p_{FWE} > .56$) (Figure 1B).

Amygdala Habituation Regression Analyses

A regression analysis with BSL total score as a predictor did not provide evidence for a significant association between symptom severity and amygdala habituation in BPD ($t = 1.92$, $p_{FWE} > .279$) (Figure 1C). Also, we detected no significant correlation between any of these BSL scale items and our habituation estimates (all $r < .18$, all $p > .11$), including items 9, 14, and 20 specifically relating to affective instability (capturing inner tension, mood swings, and fear of losing control, respectively). Similarly, arousal at the beginning of the experiment was not associated with amygdala habituation estimates ($t = 1.9$, $p_{FWE} >$

Table 1. Sample Description for Healthy Control Subjects and Patients With Current and Remitted BPD

Characteristics	BPD-C			BPD-R			HC			ANOVA/ χ^2, p	HC vs. BPD-C, p	HC vs. BPD-R, p	BPD-C vs. BPD-R, p
	M ± SD or *n*	Min/Max	*n*	M ± SD or *n*	Min/Max	*n*	M ± SD or *n*	Min/Max	*n*				
Demographic Data													
Age, years	29.03 ± 7.12	18/49	98	28.55 ± 3.94	21/38	22	28.43 ± 7.36	18/51	115	.824[a]	.821	.994	.900
Sex, male/female	9/89	–	98	0/22	–	22	19/96	–	115	.050[b]	.114	.04	.139
Education, years	12.06 ± 1.47	9/15	98	12.52 ± 1.53	10/16	22	12.09 ± 1.37	8/14	115	.374[c]	.858	.189	.185
Psychological Data													
Intelligence (RPM)	54.12 ± 6.26	33/60	94	54.37 ± 3.89	44/59	19	54.84 ± 4.71	35/60	51	.757[c]	.479	.696	.867
Early adversity (CTQ)	61.45 ± 18.76	26/103	85	54.38 ± 17.41	32/93	21	32.82 ± 10.21	25/68	115	<.001[a]	<.001	<.001	.244
Emotional abuse	17.22 ± 5.60	5/25	85	14.27 ± 6.35	7/25	22	6.83 ± 2.91	5/18	115	<.001[a]	<.001	<.001	.132
Physical abuse	8.74 ± 4.22	5/22	85	8.38 ± 6.07	5/25	21	5.60 ± 1.55	5/13	115	<.001[a]	<.001	.117	.964
Sexual abuse	8.95 ± 5.64	5/25	86	6.90 ± 4.68	5/25	21	5.31 ± 1.21	5/14	115	<.001[a]	<.001	.290	.210
Emotional neglect	17.15 ± 5.33	6/25	86	15.68 ± 5.79	6/25	22	8.26 ± 4.24	5/25	115	<.001[a]	<.001	<.001	.534
Physical neglect	9.91 ± 4.08	5/25	86	8.77 ± 3.29	5/16	22	6.43 ± 2.41	5/17	115	<.001[a]	<.001	.011	.367
Perceived arousal (SAM; low ≙ arousal)	3.14 ± 0.92	1/5	95	3.57 ± 0.98	2/5	22	3.79 ± 0.78	2/5	107	<.001[c]	<.001	.260	.055
Perceived pleasure (SAM; low ≙ pleasure)	2.75 ± 0.73	1/5	95	3.25 ± 0.70	2/5	22	3.77 ± 0.67	2/5	107	<.001[c]	<.001	.001	.005
Perceived dominance (SAM; high ≙ dominance)	2.86 ± 0.67	1/5	95	2.52 ± 0.76	1/4	22	1.86 ± 0.71	1/4	107	<.001[c]	<.001	<.001	.038
fMRI Task Performance													
Correct ratio face, %	99.33 ± 2.90	75/100	93	99.24 ± 2.09	92/100	22	99.64 ± 1.18	96/100	115	.506[c]	.284	.215	.641
Correct ratio form, %	96.81 ± 4.89	75/100	93	97.34 ± 3.29	88/100	22	97.43 ± 3.19	83/100	115	.291[a]	.254	.994	.254
fMRI Data Quality													
Signal-to-noise ratio	94.18 ± 12.95	55.40/119.46	98	93.14 ± 10.39	68.60/107.01	22	94.28 ± 16.10	41.29/128.60	115	.943[c]	.961	.751	.727
Spikes	3.08 ± 11.86	0/90	98	1.86 ± 6.57	0/30	22	3.87 ± 17.74	0/168	115	.819[c]	.709	.602	.643
Sum motion translation, mm	1.15 ± 0.62	0.35/3.29	98	1.15 ± 0.39	0.30/1.79	22	1.04 ± 0.56	0.36/3.99	115	.336[c]	.170	.389	.980
Sum motion rotation, degree	1.02 ± 0.73	0.28/4.90	98	1.07 ± 0.53	0.33/2.38	22	0.91 ± 0.45	0.29/3.27	115	.235[a]	.378	.396	.941
Mean FWD, mm	0.19 ± 0.07	0.08/0.39	98	0.18 ± 0.08	0.06/0.41	22	0.17 ± 0.07	0.06/0.46	115	.278[c]	.108	.523	.776

ANOVA, univariate analysis of variance; BPD, borderline personality disorder; BPD-C, current BPD; BPD-R, remitted BPD; CTQ, Childhood Trauma Questionnaire; fMRI, functional magnetic resonance imaging; FWD, framewise displacement; HC, healthy control subjects; M, mean; *n*, number of available data points; RPM, Raven's Progressive Matrices; SAM, self-assessment manikin (acquired immediately before fMRI); spikes, number of time points in which the signal intensity is larger than 10 × SD of the mean signal.

[a]Welch-ANOVA for all participant groups and Games-Howell test for 2-group comparisons (unequal group variances identified by the Levene test).

[b]χ^2 for all participant groups and 2-group comparisons.

[c]ANOVA for all participant groups and independent *t* test for 2-group comparisons.

Table 2. Clinical Symptoms and Comorbidities in Currently Manifest and Remitted Patients With BPD

	BPD-C			BPD-R			t Test/χ^2
	M ± SD or n	Min/Max	n	M ± SD or n	Min/Max	n	p Value
Clinical Symptoms							
BPD symptoms (BSL)	1.82 ± 0.80	0.09/3.48	89	0.51 ± 0.43	0.09/1.65	22	<.001[a]
Self-injury (if occurred; NSSI)	10.58 ± 13.82	0/59	78	1.55 ± 1.03	0/3	11	<.001[a]
Dissociation (FDS)	21.01 ± 11.28	1.14/53.18	88	7.64 ± 5.89	0.45/23.18	22	<.001[a]
Impulsivity (BIS)	82.10 ± 11.92	50/111	88	70.73 ± 9.73	55/87	22	<.001[b]
Depression (BDI-II)	24.99 ± 11.19	0/54	81	8.35 ± 6.84	0/29	20	<.001[a]
Global functioning (GAF)	51.72 ± 9.07	35/76	93	69.59 ± 7.47	57/82	22	<.001[b]
Suicide attempts (number if occurred)	2.47 ± 1.94	1/10	49	1.92 ± 0.99	1/4	12	.344[b]
Age first suicide attempt (years if occurred)	19.23 ± 6.56	9/42	48	16.73 ± 4.22	11/25	11	.285[b]
Axis I Disorders (DSM-IV Criteria, SCID-I)							
Any current comorbid diagnosis, yes/no	82/16	–	98	11/11	–	22	.001[c]
Any lifetime comorbid diagnosis, yes/no	93/5	–	98	22/0	–	22	.279[c]
Axis II Disorders (DSM-IV Criteria, IPDE)							
Current antisocial PD, yes/no	1/87	–	88	0/21	–	21	.624[c]
Current avoidant PD, yes/no	26/68	–	94	0/22	–	22	.005[c]
Lifetime antisocial PD, yes/no	4/87	–	91	0/22	–	22	.317[c]
Lifetime avoidant PD, yes/no	27/66	–	93	3/19	–	22	.139[c]

BDI, Beck Depression Inventory; BIS, Barratt Impulsivity Scale; BPD, borderline personality disorder; BPD-C, current BPD; BPD-R, remitted BPD; BSL, Borderline Symptom List; FDS, Dissociative Experience Scale; GAF, Global Assessment of Functioning; IPDE, International Personality Disorder Examination; M, mean; n, number of available data points; NSSI, nonsuicidal self-injurious behavior within the past 12 months; PD, personality disorder; SCID-I, Structured Clinical Interview for DSM-IV Axis I Disorders.

[a]Independent t test with unequal variances.

[b]Independent t test with equal variances.

[c]χ^2 test.

.288). However, in a model including all study participants, group as a factor, and CTQ total scores as a predictor, decreased right amygdala habituation was significantly related to higher exposure to adverse childhood experiences (t = 3.26, p_{FWE} = .013 [Figure 2]; p_{FWE} = .024 after correction for bilateral amygdala volume). In the same model the main effect of group (i.e., the comparison of patients with BPD and HCs in the presence of a CTQ covariate) provided a clear null finding (t = 0.88, p_{FWE} = .622). Also, we detected no significant CTQ × group interaction effects on amygdala habituation ($F_{2,213}$ = 4.61, p = .161), suggesting that the initially observed group difference in right amygdala habituation between HCs and patients with BPD in fact related to the group-dependent differences in exposure to adverse childhood experiences.

Based on the observed relationship of amygdala habituation to childhood adversity, we explored post hoc the potential contribution of different trauma types to this association (as defined by the CTQ subscales quantifying the severity of emotional abuse, physical abuse, sexual abuse, emotional neglect, and physical neglect). Consistent with prior reports (39–41), the CTQ subscales were significantly intercorrelated (all $r > .38$, all $p < .001$). Exploratory correlation analyses with peak voxel habituation estimates showed that apart from sexual abuse ($r > -.11$, all $p = .119$), all other CTQ subscales were significantly related to amygdala habituation (all $r > .20$, all $p < .002$).

DISCUSSION

In this study we used an established fMRI paradigm (23) providing reliable amygdala habituation estimates (16) to confirm the finding of deficient amygdala habituation to repeated emotional stimuli in BPD (19). We further aimed to gain insight into the neurobiological underpinnings of this phenotype by probing its relationship to BPD symptom severity and early social risk exposure.

In line with our expectations, we detected a significantly lower amygdala habituation in our BPD patient group. This observation is consistent with a prior study (19) and extends the current knowledge on neural emotional processing. First, similar to Hazlett *et al.* (19) , the group difference resulted from a rapid decline of right amygdala responsivity to repeated emotional blocks in HCs, whereas patients with BPD failed to show a decline in amygdala responsivity to these repetitions. Second, we detected a similar result even though there were several noticeable methodical differences between this study and that of Hazlett *et al.* (19), including specifics of the stimulus material (Ekman faces vs. International Affective Picture System), task design (blocked vs. event-related design), and analysis strategy (voxelwise in imaging space vs. analysis of extracted region of interest response estimates). Third, our findings come from an independent cohort with a considerably larger sample size. We conclude from these points that deficient amygdala habituation to successive emotional stimuli is a reproducible finding in BPD that appears to be relatively robust to variations in experimental methods.

Deficits in amygdala habituation to affective stimulation in BPD have previously been interpreted as a plausible neural mechanism for the unstable and intense affective reactions seen in these patients (19,20). This points to a clinically relevant signal, that is, the scale of amygdala habituation across

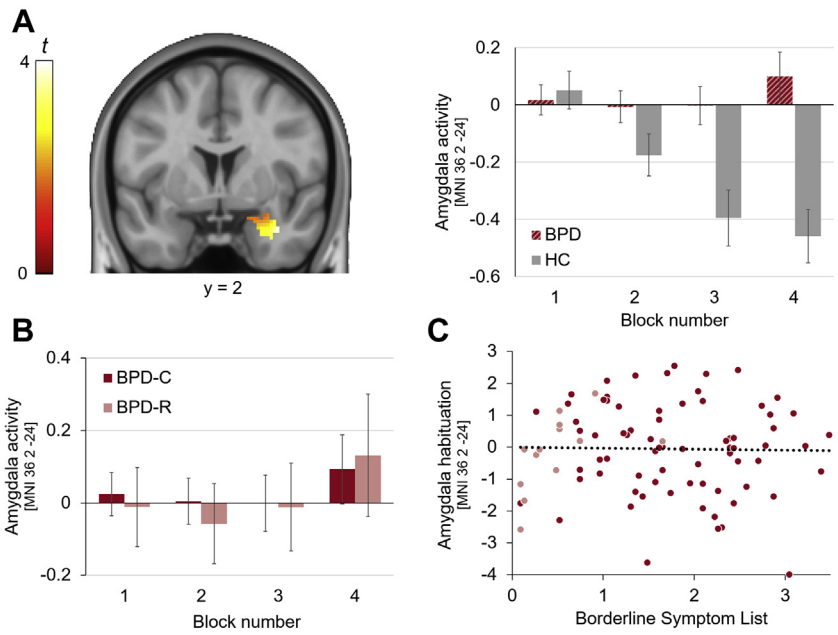

Figure 1. Changes in amygdala reactivity to threatening stimuli over the course of the functional magnetic resonance imaging experiment. **(A)** Significant difference in amygdala habituation manifesting as a rapid signal decrement to successive emotional stimulation blocks in the healthy control (HC) but not the borderline personality disorder (BPD) group (t = 3.27, p_{FWE} = .015). **(B)** No significant difference in amygdala habituation between currently manifest (BPD-C) and remitted (BPD-R) patients (all values $t <$ 1.20, $p_{FWE} >$ .56). **(C)** Amygdala habituation estimates do not relate to current BPD symptom severity scores (t = 1.92, $p_{FWE} >$ .279). The functional map in panel **(A)** is thresholded at p = .05, uncorrected for illustration purposes, and is displayed on the coronal section of a structural-template magnetic resonance image. In panels **(A)** and **(B)**, blockwise habituation estimates are plotted for presentation purposes. MNI, Montreal Neurological Institute standard space.

the fMRI scan is expected to reflect the degree of the current severity of the condition. We specifically examined this question by probing the sensitivity of the habituation phenotype to variations in current BPD diagnostic status (BPD-C, BPD-R) and symptom load (BSL). Interestingly, neither the comparison between acutely ill and remitted patients nor our association analysis with an established instrument for the severity assessment of borderline-typical symptoms was related to this amygdala phenotype. Furthermore, habituation did not relate to differences in the perceived arousal state immediately before the fMRI scan.

The habituation abnormality seen in BPD may indicate that the phenotype reflects a neural risk marker for BPD illness vulnerability. We probed this matter in the context of early exposure to social risk and detected a significant association between reduced amygdala habituation and higher severity of adverse childhood experiences. Although our post hoc exploratory analyses suggested that nearly all assessed trauma subtypes may be of relevance for this association, the significant intercorrelations among CTQ subscales do not support any firm conclusion on the contribution of specific trauma subtypes to amygdala habituation. As required from a neural risk marker, this association was detected across the full sample and was independent of the clinical status of the participants. In addition, this analysis provided evidence that the group difference in amygdala habituation between patients with BPD and HCs is largely accounted for by the variant exposure to adverse childhood experiences. We conclude from these findings that deficient amygdala habituation to successive emotional stimuli relates to neurobiological risk for BPD—in other words, the susceptibility for developing long-lasting and intense affective reactions to affectively stimulating encounters related to environmental adversity in childhood.

Deficient amygdala habituation to threat-related stimuli has been previously related to serotonin receptor density and genetic variants modulating the neurotransmission of biogenic amines, neuroplasticity, and psychiatric risk

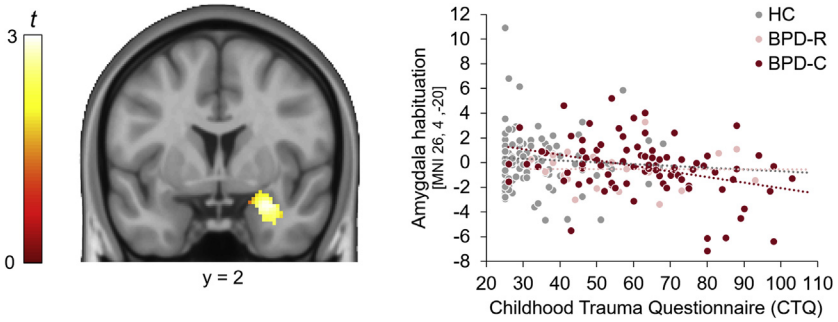

Figure 2. Decreased right amygdala habituation to repeated threatening stimuli during functional magnetic resonance imaging significantly relates to higher exposure to adverse childhood experiences (t = 3.26, p_{FWE} = .013). BPD, borderline personality disorder; BPD-C, current BPD; BPD-R, remitted BPD; FWE, familywise error; HC, healthy control subjects; MNI, Montreal Neurological Institute.

(21,22,35,42). Our work extends this knowledge by suggesting that in addition to genetic risk, an established social environmental risk factor for psychiatric disorders is relevant for the same phenotype, thereby possibly shaping the evolutionarily conserved sensitivity for the detection of threatening social stimuli. Notably, the question of the clinical specificity of the neural habituation phenotype was beyond the scope of this study. However, amygdala habituation deficits have been linked to other clinical syndromes and risk constellations, in particular to autism spectrum disorder (35,43,44) and temperamental risk for social anxiety (45). Moreover, recent evidence suggests that the genetic risk architecture of BPD overlaps substantially with that of other psychiatric disorders (46). In light of this literature, it thus appears implausible that deficient amygdala habituation would reflect a neural phenotype that is specific to BPD. Instead, we posit that multiple sources of illness risk (and possibly also resilience) may converge on this neural system and phenotype, thereby shaping the vulnerability for developing BPD and possibly other psychiatric disorders with a shared genetic and environmental risk architecture and overlapping affective and social deficits. Further research is needed to corroborate this notion.

Although we adopted previously validated methods and examined a large group of individuals, several limitations of our study merit comment. First, compared with the size of the current BPD sample, the included group of 22 patients with BPD-R is still relatively small. Second, we cannot fully rule out the potential influence of antidepressant treatment on our findings. However, less than 13% of our patients were on medication (all from the BPD-C group, and only selective serotonin reuptake inhibitors and serotonin and norepinephrine reuptake inhibitors), and the lack of an amygdala habituation difference between patients with BPD-C and BPD-R makes a relevant drug effect appear unlikely. Third, our opinions on the lack of clinical specificity of the amygdala habituation risk phenotype are based on the current literature and require more explicit confirmation in future studies including multiple clinical conditions as well as healthy populations at genetic and environmental risk. Fourth, the reduced variance of CTQ scores in the control subjects may have limited the examination of associations to habituation. Finally, we acknowledge the presence of conflicting reports in the literature. For example, Denny *et al.* (47) did not detect a difference in within-session habituation between samples of 26 patients with BPD and 24 HCs but detected a significant increase in salience network sensitization to the same stimuli in a later study session. This discrepancy to our habituation finding may plausibly be related to differences in task design and the stimuli used. Although Denny *et al.* applied an event-related design with 5 presentations of the same negative social images, we compared early and late block-related activations during cognitive matching of different emotional stimuli. Alternatively, or in addition, we believe that higher statistical power (i.e., the larger sample sizes in our study) may have facilitated the detection of a significant within-session habituation deficit in BPD in our work.

To conclude, this study confirms the finding of deficient amygdala habituation to repeated emotional stimuli in BPD in a large sample of participants and suggests that this phenotype is, at least to some extent, robust to variations in neuroimaging methods. Our data do not support a link of amygdala habituation to the clinical severity of BPD. Instead, we identify a significant association between this phenotype and the severity of adverse experiences during childhood, which largely accounts for group difference between patients with BPD and HCs. In light of the existing clinical literature on amygdala habituation, we propose that multiple sources of illness risk (and possibly also resilience) may converge on this neural system and shape the vulnerability to the development of BPD and also likely other psychiatric disorders with pronounced affective and social deficits and a shared genetic and environmental risk architecture.

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REFERENCES

1. Schmahl C, Herpertz SC, Bertsch K, Ende G, Flor H, Kirsch P, *et al.* (2014): Mechanisms of disturbed emotion processing and social interaction in borderline personality disorder: State of knowledge and research agenda of the German Clinical Research Unit. Borderline Personal Disord Emot Dysregul 1:12.
2. Tomko RL, Trull TJ, Wood PK, Sher KJ (2014): Characteristics of borderline personality disorder in a community sample: Comorbidity, treatment utilization, and general functioning. J Person Disord 28:734–750.
3. Tragesser SL, Solhan M, Schwartz-Mette R, Trull TJ (2007): The role of affective instability and impulsivity in predicting future BPD features. J Person Disord 21:603–614.
4. Linehan MM, Kehrer CA (1993): Borderline personality disorder. In: Barlow DH, editor. Clinical Handbook of Psychological Disorders: A Step-By-Step Treatment Manual, 2 ed. New York: Guilford Press, 396–441.
5. Leichsenring F, Leibing E, Kruse J, New AS, Leweke F (2011): Borderline personality disorder. Lancet 377:74–84.
6. Torgersen S (2000): Genetics of patients with borderline personality disorder. Psychiatr Clin North Am 23:1–9.
7. Herpertz SC, Dietrich TM, Wenning B, Krings T, Erberich SG, Willmes K, *et al.* (2001): Evidence of abnormal amygdala functioning in borderline personality disorder: A functional MRI study. Biol Psychiatry 50:292–298.
8. Donegan NH, Sanislow CA, Blumberg HP, Fulbright RK, Lacadie C, Skudlarski P, *et al.* (2003): Amygdala hyperreactivity in borderline personality disorder: Implications for emotional dysregulation. Biol Psychiatry 54:1284–1293.
9. Minzenberg MJ, Fan J, New AS, Tang CY, Siever LJ (2007): Fronto-limbic dysfunction in response to facial emotion in borderline personality disorder: An event-related fMRI study. Psychiatry Res 155:231–243.
10. Schulze L, Schmahl C, Niedtfeld I (2016): Neural correlates of disturbed emotion processing in borderline personality disorder: A multimodal meta-analysis. Biol Psychiatry 79:97–106.
11. Koenigsberg HW, Siever LJ, Lee H, Pizzarello S, New AS, Goodman M, *et al.* (2009): Neural correlates of emotion processing in borderline personality disorder. Psychiatry Res 172:192–199.
12. Bordi F, LeDoux J (1992): Sensory tuning beyond the sensory system: An initial analysis of auditory response properties of neurons in the lateral amygdaloid nucleus and overlying areas of the striatum. J Neurosci 12:2493–2503.
13. Breiter HC, Etcoff NL, Whalen PJ, Kennedy WA, Rauch SL, Buckner RL, *et al.* (1996): Response and habituation of the human amygdala during visual processing of facial expression. Neuron 17:875–887.
14. Phillips ML, Medford N, Young AW, Williams L, Williams SC, Bullmore ET, *et al.* (2001): Time courses of left and right amygdalar responses to fearful facial expressions. Hum Brain Mapp 12:193–202.
15. Rankin CH, Abrams T, Barry RJ, Bhatnagar S, Clayton DF, Colombo J, *et al.* (2009): Habituation revisited: An updated and revised description of the behavioral characteristics of habituation. Neurobiol Learn Mem 92:135–138.
16. Plichta MM, Grimm O, Morgen K, Mier D, Sauer C, Haddad L, *et al.* (2014): Amygdala habituation: Areliable fMRI phenotype. NeuroImage 103:383–390.
17. Plichta MM, Schwarz AJ, Grimm O, Morgen K, Mier D, Haddad L, *et al.* (2012): Test-retest reliability of evoked BOLD signals from a cognitive-emotive fMRI test battery. NeuroImage 60:1746–1758.
18. Gee DG, McEwen SC, Forsyth JK, Haut KM, Bearden CE, Addington J, *et al.* (2015): Reliability of an fMRI paradigm for emotional processing in a multisite longitudinal study. Hum Brain Mapp 36:2558–2579.
19. Hazlett EA, Zhang J, New AS, Zelmanova Y, Goldstein KE, Haznedar MM, *et al.* (2012): Potentiated amygdala response to repeated emotional pictures in borderline personality disorder. Biol Psychiatry 72:448–456.
20. Goodman M, Carpenter D, Tang CY, Goldstein KE, Avedon J, Fernandez N, *et al.* (2014): Dialectical behavior therapy alters emotion regulation and amygdala activity in patients with borderline personality disorder. J Psychiatr Res 57:108–116.
21. Lonsdorf TB, Golkar A, Lindstom KM, Fransson P, Schalling M, Ohman A, *et al.* (2011): 5-HTTLPR and COMTval158met genotype gate amygdala reactivity and habituation. Biol Psychol 87:106–112.
22. Perez-Rodriguez MM, New AS, Goldstein KE, Rosell D, Yuan Q, Zhou Z, *et al.* (2017): Brain-derived neurotrophic factor Val66Met genotype modulates amygdala habituation. Psychiatry Res Neuroimag 263:85–92.
23. Hariri AR, Mattay VS, Tessitore A, Kolachana B, Fera F, Goldman D, *et al.* (2002): Serotonin transporter genetic variation and the response of the human amygdala. Science 297:400–403.
24. Wittchen H-U, Wunderlich U, Gruschwitz S, Zaudig M (1997): SKID I. Strukturiertes Klinisches Interview für DSM-IV. Achse I: Psychische Störungen. Interviewheft und Beurteilungsheft. Eine deutschsprachige, erweiterte Bearb. d. amerikanischen Originalversion des SKID I. Hogrefe-Verlag: Goettingen.
25. Loranger AW (1999): IPDE: International Personality Disorder Examination: DSM-IV and ICD-10 Interviews. Lutz, FL: PARS Psychological Assessment Resources.
26. Raven J, Raven JC, Court JH (1998): Raven Manual: Section 4, Advanced Progressive Matrices. Oxford, UK: Oxford Psychologists Press.
27. Bernstein DP, Fink L (1998): Childhood Trauma Questionnaire: A Retrospective Self-Report: Manual. San Antonio, TX: Harcourt Brace & Co.
28. Bohus M, Limberger MF, Frank U, Chapman AL, Kuhler T, Stieglitz RD (2007): Psychometric properties of the Borderline Symptom List (BSL). Psychopathology 40:126–132.
29. Freyberger HJ, Spitzer C, Stieglitz RD (1986): Fragebogen zu Dissoziative Symptomen (FDS). German adaptation of the Dissociative Experience Scale (DES). Bern: Huber.
30. American Psychiatric Association (2000): Diagnostic and Statistical Manual Of Mental Disorders, 4th ed, text revision. Washington, DC: American Psychiatric Publishing.
31. Beck AT (1996): BDI-II, Beck Depression Inventory: Manual, 2nd ed. San Antonio, TX: Psychological Corp.
32. Patton JH, Stanford MS, Barratt ES (1995): Factor structure of the Barratt impulsiveness scale. J Clin Psychol 51:768–774.
33. Bradley MM, Lang PJ (1994): Measuring emotion: the self-assessment manikin and the semantic differential. J Behav Ther Exp Psychiatry 25:49–59.
34. Ekman P, Friesen WV (1976): Measuring facial movement. Environment Psychol Nonverbal Behav 1:56–75.
35. Wiggins JL, Swartz JR, Martin DM, Lord C, Monk CS (2014): Serotonin transporter genotype impacts amygdala habituation in youth with autism spectrum disorders. Soc Cogn Affect Neurosci 9:832–838.
36. Tzourio-Mazoyer N, Landeau B, Papathanassiou D, Crivello F, Etard O, Delcroix N, *et al.* (2002): Automated anatomical labeling of activations in SPM using a macroscopic anatomical parcellation of the MNI MRI single-subject brain. NeuroImage 15:273–289.
37. Wright CI, Fischer H, Whalen PJ, McInerney SC, Shin LM, Rauch SL (2001): Differential prefrontal cortex and amygdala habituation to repeatedly presented emotional stimuli. Neuroreport 12:379–383.
38. Baas D, Aleman A, Kahn RS (2004): Lateralization of amygdala activation: a systematic review of functional neuroimaging studies. Brain Res Brain Res Rev 45:96–103.
39. Liebschutz JM, Buchanan-Howland K, Chen CA, Frank DA, Richardson MA, Heeren TC, *et al.* (2018): Childhood Trauma Questionnaire (CTQ) correlations with prospective violence assessment in a longitudinal cohort. Psychol Assess 30:841–845.

40. Glaesmer H, Schulz A, Hauser W, Freyberger HJ, Brahler E, Grabe HJ (2013): [The childhood trauma screener (CTS) - development and validation of cut-off-scores for classificatory diagnostics]. Psychiatr Praxis 40:220–226.
41. Pieritz K, Rief W, Euteneuer F (2015): Childhood adversities and laboratory pain perception. Neuropsychiatr Dis Treat 11:2109–2116.
42. Fisher PM, Meltzer CC, Price JC, Coleman RL, Ziolko SK, Becker C, *et al.* (2009): Medial prefrontal cortex 5-HT(2A) density is correlated with amygdala reactivity, response habituation, and functional coupling. Cereb Cortex 19:2499–2507.
43. Kleinhans NM, Johnson LC, Richards T, Mahurin R, Greenson J, Dawson G, *et al.* (2009): Reduced neural habituation in the amygdala and social impairments in autism spectrum disorders. Am J Psychiatry 166:467–475.
44. Kleinhans NM, Richards T, Greenson J, Dawson G, Aylward E (2016): Altered dynamics of the fMRI response to faces in individuals with autism. J Autism Dev Disord 46:232–241.
45. Blackford JU, Allen AH, Cowan RL, Avery SN (2012): Amygdala and hippocampus fail to habituate to faces in individuals with an inhibited temperament. Soc Cogn Affect Neurosci 8:143–150.
46. Witt SH, Streit F, Jungkunz M, Frank J, Awasthi S, Reinbold CS, *et al.* (2017): Genome-wide association study of borderline personality disorder reveals genetic overlap with bipolar disorder, major depression and schizophrenia. Transl Psychiatry 7:e1155.
47. Denny BT, Fan J, Fels S, Galitzer H, Schiller D, Koenigsberg HW (2018): Sensitization of the neural salience network to repeated emotional stimuli following initial habituation in patients with borderline personality disorder. Am J Psychiatry 175:657–664.