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Methods

Participants

Patients were recruited from mental health clinics at local sites, Virenze-Riagg Maastricht (The Netherlands), PsyQ Heerlen (The Netherlands), the BPD treatment unit of the Department of Psychiatry and Psychotherapy at the Medical Center Freiburg (Germany), the Department of Psychiatry and Psychotherapy at the University Hospital Lübeck (Germany) and the Institute for Behavior Therapy Training Hamburg (Germany). Borderline personality disorder (BPD) patients were recruited within the context of an international multicenter RCT on group schema therapy versus treatment-as-usual.[1] Only females were chosen since gender might influence emotional processing[2], and because in mental health care BPD is more often diagnosed in females. We excluded homosexual females, because we used heterosexual erotic stimuli. General exclusion criteria were lifetime psychotic or bipolar disorder type-I, attention-deficit/hyperactivity disorder, dissociative identity disorder, serious and/or unstable medical illness, substance dependence needing clinical detoxification and fMRI exclusion criteria (i.e. claustrophobia, metal objects, cardiac arrhythmia, epilepsy, tattoos at neck/head and pregnancy).

Four BPD patients, one non-patient (NPC) and one cluster-C personality disorder patient (CCP) were excluded because of invalid or incomplete data, one BPD and one CCP because of too much head motion (if head motion in 2 or more runs was > 4 mm) during scanning, three NPC because of scores above 0.70 on the Brief Symptom Inventory and three CCP because of scores above 100 on the BPD checklist. Two BPD and two CCP were excluded because they had

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an estimated IQ outside the range of 70-120. Additionally, as there was a disproportionate number of NPC with IQ higher than the highest IQ in the BPD patients, we excluded two NPC with IQ scores above the 95-percentile to guarantee that NPC were better matched to the BPD patients.

Measures

Brief Symptom Inventory (BSI) - The BSI is a brief psychological self-report inventory of general symptoms of psychopathology during the past week.[3] It is a short alternative for the Symptom Checklist-90-R from which it was developed. It contains 53 items divided over nine dimensions: somatization, obsession-compulsion, interpersonal sensitivity, depressive mood, anxiety, hostility, phobic anxiety, paranoid ideation and psychoticism. Answers are scored on a 5-point Likert scale, ranging from 0 (not at all) to 4 (extremely). Scores of the dimensions are calculated by summing the values for the items divided by the number of items within the subscales. The total score measures the level of symptomatology, which is the sum of the nine dimensions plus the four additional items divided by total number of items. The internal consistency showed a Cronbach's α of 0.96 for the total instrument and ranged between 0.71 and 0.85 for its subscales[3,4] To distinguish patients from non-patients a cutoff score of 0.70 is suggested.[5]

BPD Checklist - The BPD Checklist is a self-report questionnaire used to assess the burden of BPD symptoms as experienced during the last month.[6] It consists of 47 items based on the nine

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dimensions of BPD in DSM-IV. Items must be rated on 5-point Likert scale, ranging from 1 (not at all) to 5 (extremely). Next to the total sumscore, also the scores for the nine subscales can be calculated. Scores above 100 signify BPD pathology and therefore is indicative as cutoff for inclusion criteria. When control patients showed an elevated score, an extra check with the SCID-II BPD section was done.

Interview for Traumatic Events in Childhood (ITEC) - The ITEC is a retrospective, semi-structured interview to measure childhood maltreatment prior the age of 18 including sexual (12 items), physical (13 items) and emotional abuse (9 items), and emotional (6 items) and physical neglect (15 items).[7] For each item the participant experienced maltreatment, follow-up questions are used to gather more detailed information about the perpetrator(s), age of onset, frequency, duration of the trauma and the impact on the victim in the past and in the present. This information was used to calculate a severity score between 0 and 1, such that the score increased with the severity of the event itself, the closeness of the perpetrator, the younger age of onset, the longer duration of the event, and the higher impact on the victim. For each subscales the severity scores for the events are summed, the higher this score, the more severe the maltreatment. Internal consistencies of these scales were moderate to excellent, with Cronbach's α varying between 0.58 and 0.89 with a mean of 0.79.[7] In addition to the victimization scales, similar scales were created for witnessing the various forms of maltreatment. In current study only the

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victim scales were reported. On average the administration time is about 30 minutes but can take up to one hour in case of multiple maltreatments.

Dissociation and Anxiety - Present state dissociative experiences were assessed using four items of the Dissociation-Tension-Scale[8], containing derealization and changes in perception of one's body, hearing and pain. Additionally two items concerning the level of anxiety and the level of nervousness were added. Responses were indicated on a visual analogue scale, ranging from 0 not at all to 100 extreme. The dissociation score was the averaged across the four dissociation items. Internal consistencies of the dissociation score proved to be good in current sample, with a Cronbach's α of 0.83 concerning dissociation before scanning and a Cronbach's α of 0.86 concerning dissociation after scanning.

Self-Assessment Manikin Scale - The pictures shown during the scanning session were qualitatively assessed for valence and arousal using the Self-Assessment Manikin Scale. The Self-Assessment Manikin Scale consists of a series of human-like figures to measures the affective reaction of a person to stimuli.[9] Intensity of valence and arousal were both rated on a 9-point scale, with for valence 1 being extremely unpleasant and 9 being extremely pleasant, and for arousal 1 being most calm and 9 being most aroused.

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Wechsler Adult Intelligence Scale - IQ was estimated by means of four subtests of the Wechsler Adult Intelligence Scale, including two verbal (i.e. Vocabulary and Similarities) and two nonverbal tests (i.e. Block design and Matrix reasoning). Together these subtests correlate strongly with general intellectual ability.[10] IQ was estimated based on the optimized regression equation: 39.05 + (1.54 * comprehension score) + (1.64 * matrix reasoning score) + (1.48 * similarities score) + (0.98 * picture arrangement score). If the WAIS score was not available (BPD patients n = 11, NPC n = 3 and CCP n = 1) an estimation of the IQ was made based on the education level, using the regression equation from our present sample per group; IQ BPD patients = 85.932 + (3.360 * ISCED code), IQ NPC = 85.996 + (3.659 * ISCED code) and IQ CCP = 81.529 + (4.226 * ISCED code). These regression equations explained respectively 22%, 31% and 36% of the variance in IQ. Level of education of both the Dutch and German educational systems were transformed into the International Standard Classification of Education (ISCED).

Borderline Personality Disorder Severity Index (BPDSI) - The BPDSI is a semi-structured clinical interview assessing frequency and severity of BPD manifestations.[11-13] The 70-items reflect the nine BPD criteria described in the DSM-IV. For each item the frequency of the last three months is rated on an 11-point scale, ranging from 0 (never) to 10 (daily). The scores on the subscales provide information on the severity of each of the DSM-IV dimensions, derived by averaging the items scores. The total score is the sum of the nine dimensions scores, ranging

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from 0 to 90, with an internal consistency of Cronbach's $\alpha = 0.93$ and subscales ranged between 0.41 and 0.83.[11] A total score of 20 distinguishes BPD from other personality disorders.[12]

Procedure

To ensure correct and confident use of the emotion regulation strategy and the nature of the stimuli, prior to scanning the participant was trained with a practice task outside the scanner. This task was similar to the experimental task inside the scanner. After completing the practice task the participant entered the scanner and the scanning session was performed, lasting 75 minutes. As part of the scanning session participants also underwent two resting state scans (results reported separately[14]). Before and after scanning the anxiety and dissociation state was assessed.[8] In addition, at the end of the session the participant completed an 'exit'-questionnaire, assessing information about a description of which strategy the participant used concerning both instructions (look and safe) and general experience with the fMRI-measurement. Finally, the participants had to rate their subjective reaction of each picture they had seen during the session.

fMRI acquisition

Functional MRI was performed on 3-Tesla scanners at the three sites, at Maastricht on a Siemens Magnetom Allegra head-only scanner equipped with a birdcage headcoil (Siemens Medical Systems, Erlangen, Germany), at Freiburg on a Siemens tim-Trio Magnetom whole body scanner (Siemens Medical Systems, Erlangen, Germany) equipped with an 8-channel

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headcoil, and at Lübeck on a Philips Achiva whole body scanner equipped with an 8-channel headcoil (Philips Healthcare, Best, The Netherlands). The BPD patients from Heerlen were scanned in Maastricht and the BPD patients from Hamburg were scanned in Lübeck. In Maastricht 13 BPD, 10 NPC and 11 CCP were scanned, Freiburg scanned 14 BPD, 18 NPC and 8 CCP, finally 28 BPD, 14 NPC and 5 CCP were scanned in Lubeck.

Participants were scanned in head first supine position. Head movements were minimalized using foam paddings. Additionally, participants were instructed not to look away from the stimuli or to close her eyes, and avoid moving as much as possible during scanning. T2*-weighted images were acquired via echo planar imaging (EPI), using the following imaging parameters: repetition time = 2000 ms, echo time = 27 ms, flip angle = 90°, field of view = 192 x 192 mm, voxel size = 3 x 3 x 3 mm, and matrix = 64 x 64. Images were recorder in four runs of 240 images in Maastricht and 252 images in Freiburg and Lübeck. One volume in Maastricht consisted of 32, and in Freiburg and Lübeck of 34, interleaved measured axial slices. The T2*-weighted slices were optimized with a negative tilt of 30°, to minimize susceptibility and distortion artifacts within the amygdala[15] in Maastricht and Freiburg. A whole brain anatomical scan in sagittal plane was acquired, using a high resolution T1-weigthed sequence (repetition time = 2250 ms, echo time = 2.6 ms, flip angle = 9°, field of view = 256 x 256 mm, voxel size 1 x 1 x 1 mm). In total, 192 images were obtained in Maastricht, 160 in Freiburg and 170 in Lübeck.

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fMRI preprocessing

All preprocessing and statistical analyses were performed with BrainVoyager 2.6 (Brain Innovation, Maastricht, The Netherlands). The first two images of each run were discarded because of saturation effects. Preprocessing comprised slice time correction with sinc interpolation, 3D motion correction for three translation and three rotation parameters with trilinear interpolation for detection, and sinc interpolation for motion correction and removal of low-frequency drifts was performed by high-pass temporal filtering of 2 sines/cosines per run.[16] In all anatomical scans, the brain was peeled from the skull and corrected for intensity inhomogeneities. Participants underwent a second session concerning another task data presented elsewhere[17], in which also an anatomical scan was conducted. To obtain a high resolution and high contrast anatomical scan, both anatomical scans were averaged when possible. After preprocessing the functional data were coregistered with the anatomical data per run, and for each run a volume-time-course was created. Each volume-time-course was spatially smoothed with a 6 mm full-width-at-half-maximum isotropic Gaussian kernel. Spatial normalization was performed using standard Talairach transformation procedures (Talairach and Tournoux, 1988).

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Supplementary Results

Behavioral data

To examine whether the experienced emotion during scanning (Table S1) after each trial depended on the stimulus category and/or differed between groups an instruction (look vs. safe) x stimulus (negative, positive, erotic vs. neutral) x group (BPD, NPC vs. CCP) repeated measures ANOVA was performed. No significant three-way interaction was shown. Besides the significant instruction x stimulus interaction ($F_{3,114}$ = 3.94; p = 0.010: see main text) the stimulus x group interaction ($F_{6,230}$ = 2.56; p = 0.020) was significant. Post hoc tests revealed that BPD patients reported significantly stronger emotions during erotic (t_{93} = -3.19; p = 0.002) and neutral (t_{93} = -3.28; p = 0.001) stimuli than NPC. BPD patients did not significantly differ from CCP, who did not differ from NPC.

To examine whether the arousal and valence ratings of the stimulus evaluations after scanning (Table S1) differed between groups two stimulus (negative, positive, erotic vs. neutral) x group (BPD, NPC vs. CCP) repeated measures ANOVAs were performed, one involving arousal ratings and the other involving valence ratings. The stimulus evaluations after scanning showed besides a main effect of arousal ($F_{3,80}$ = 74.38; p < 0.001; see main text) a significant valence x group interaction ($F_{6,162}$ = 2.52; p = 0.024). Post hoc tests showed that BPD patients rated the erotic (t_{61} = -2.96; p = 0.004), neutral (t_{61} = -3.85; p < 0.001) and positive (t_{61} = 3.19; p = 0.002) stimuli significantly less pleasant than NPC. CCP rated neutral stimuli significantly less

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pleasant than NPC (t_{54} = 2.59; p = 0.012). Negative stimuli did not differ in valence rating across groups.

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Table S1. Ratings of experienced emotion during scanning and stimulus evaluation after scanning of the three groups.

Experienced emotion during scanning		BPD *n* = 54	NPC *n* = 41	CCP *n* = 24
Neutral, mean (SD)	Look	-2.05 (15.40)	11.61 (20.69)	8.12 (24.43)
	Safe	3.83 (14.78)	15.33 (28.26)	13.17 (19.04)
Negative, mean (SD)	Look	-61.57 (24.08)	-63.81 (23.53)	-69.56 (24.52)
	Safe	-56.40 (27.52)	-52.57 (32.01)	-61.45 (25.61)
Positive, mean (SD)	Look	45.57 (26.30)	51.38 (23.51)	51.58 (28.38)
	Safe	43.45 (25.21)	52.65 (24.83)	50.24 (25.93)
Erotic, mean (SD)	Look	23.08 (36.45)	42.80 (25.75)	32.43 (30.33)
	Safe	24.08 (32.77)	45.10 (28.19)	32.43 (29.12)
Stimulus evaluations after scanning		BPD *n* = 29	NPC *n* = 34	CCP *n* = 22
Arousal, mean (SD)				
Neutral		3.76 (1.02)	3.12 (1.25)	3.73 (0.98)
Negative		5.48 (1.94)	6.21 (1.87)	6.77 (1.57)
Positive		3.62 (1.47)	3.03 (1.95)	3.59 (1.82)
Erotic		4.34 (1.49)	4.15 (1.67)	4.59 (1.53)
Valence, mean (SD)				
Neutral		5.03 (0.73)	5.94 (1.07)	5.27 (0.70)

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Negative	2.24 (1.02)	2.15 (0.86)	1.91 (1.19)
Positive	7.10 (1.05)	7.91 (0.97)	7.59 (0.96)
Erotic	6.24 (1.50)	7.26 (1.24)	6.56 (1.53)

Effect of site

The reported clusters did not show overlap with the significant clusters of group x stimulus x site interaction at lenient significance level of $p < 0.05$ (Fig. S1). Additionally, more detailed analyses within SPSS did not show a significant group x stimulus x site interaction, and the group x stimulus remained significant when site and its interactions were added (Table S2).

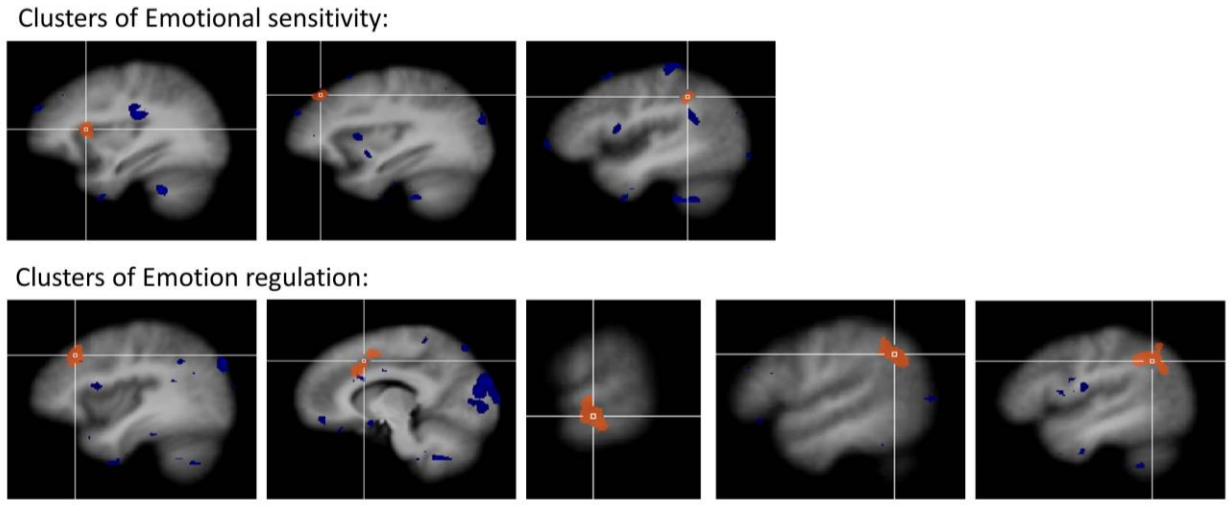

Fig. S1. Overlap of the reported clusters of group x stimulus x site interaction.

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Table S2. Detailed analyses of site.

	Stimulus x Group x Site		Stimulus x Group	
	F	*p*	*F*	*p*
Emotional Sensitivity				
Anterior insula	0.63	0.535	14.46	< 0.001
Dorsolateral prefrontal cortex	1.86	0.162	13.13	< 0.001
Temporoparietal junction	0.57	0.570	10.80	0.001
Emotion Regulation				
Dorsolateral prefrontal cortex	1.08	0.343	14.12	< 0.001
Dorsal anterior cingulate cortex	0.68	0.510	19.12	< 0.001
Middle temporal gyrus	0.45	0.641	17.79	< 0.001
Inferior parietal lobe, right	0.87	0.423	14.09	< 0.001
Inferior parietal lobe, left	0.32	0.726	15.88	< 0.001

Exploratory analyses of BPD patients compared to both control groups when viewing positive versus neutral stimuli

The whole brain random effects ANOVA F-map: stimulus (positive-look vs. neutral-look) x group (BPD vs. NPC) resulted in five clusters, which were anatomically identified at the left amygdala, left middle occipital gyrus and the cerebellum (Table S3). Simple effects showed higher activity in the amygdala and middle occipital gyrus for NPC when presented with positive

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compared to neutral stimuli. No differences were found for the comparison of BPD with CCP, also when corrected for the significant difference of medication between BPD and CCP.

Exploratory analyses of BPD patients compared to both control groups when viewing erotic versus neutral stimuli

The whole brain random effects ANOVA F-map: stimulus (erotic-look vs. neutral-look) x group (BPD vs. NPC) resulted in two clusters, which were anatomically identified at the medial prefrontal cortex and temporal pole (Table S3). In the medial prefrontal cortex, simple effects showed higher activity in the NPC for erotic versus neutral stimuli. Furthermore, no differences were found for the comparison of BPD and CCP. In the temporal pole, simple effects showed less activity for BPD patients compared to NPC when presented with neutral stimuli. Additionally, a significant difference was found between BPD and CCP ($F_{1,77} = 5.11$; $p = 0.007$). These results did hold when corrected for medication.

Exploratory analyses of BPD patients compared to both control groups during the safe versus look conditions when presented with positive stimuli

The whole brain random effects ANOVA F-map: stimulus (positive-safe vs. positive-look) x group (BPD vs. NPC) resulted in three clusters, which were anatomically identified at the lingual gyrus and cerebellum (Table S3). Simple effects showed in NPC higher activity during the safe compared to the look condition in the lingual gyrus. Moreover, as a significant

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difference was found for the comparison with CCP ($F_{1,77} = 4.34$; $p = 0.041$), these results point to be specific for BPD. Results remained the same when controlled for activity during neutral stimuli for the comparison with NPC and medication within the BPD group. However, for the comparison with CCP the results did not hold significance when corrected for activity during neutral stimuli and when medication was added as a covariate.

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Table S3. Significant brain areas of between BPD and NPC comparison for additional whole brain random effects ANOVAs.

Brain area	L/R	BA	Cluster size	Talairach peak voxel			*F*	*p*
			mm^3	x	y	z	df = 1, 95	
Emotional Sensitivity								
Look Positive versus Look Neutral, cluster threshold = 15								
Amygdala	L		430	-24	-10	-11	11.20	0.001
Middle occipital gyrus	R	19	658	18	-85	16	12.69	0.001
Cerebellum	R		1796	12	-88	-30	15.14	< 0.001
Cerebellum	R		459	12	-64	-41	8.05	0.006*
Cerebellum	L		854	-12	-73	-17	14.11	< 0.001
Look Erotic versus Look Neutral, cluster threshold = 11								
Medial frontal gyrus	L	10	408	-3	59	7	14.24	< 0.001
Superior temporal gyrus, temporal pole	R	38	443	18	11	-36	9.43	0.003*
Emotion Regulation								
Safe Positive versus Look Positive, cluster threshold = 13								
Lingual gyrus	L	17	365	-18	-88	-2	16.13	< 0.001

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Cerebellum	R	546	9	-88	-29	17.73	< 0.001
Cerebellum	R	1273	30	-76	-38	1.08	0.301*

L = Left; R = Right; BA = Brodmann area.
Thresholded at $p < 0.005$ and cluster size.
* Possibly affected by site.

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