Title

The effect of acute stress on the balance between goal-directed and habit networks in obsessive-compulsive disorder.

Authors

AL van der Straten MD[1-3], W van Leeuwen MD[1-3], D Denys MD PhD[1-3], HJF van Marle MD PhD[1,4], GA van Wingen PhD[1-3]

Affiliations

[1] Amsterdam UMC, University of Amsterdam, Department of Psychiatry, Amsterdam Neuroscience, Amsterdam, Netherlands

[2] Amsterdam Brain and Cognition, University of Amsterdam, Amsterdam, Netherlands

[3] Spinoza Centre for Neuroimaging, Royal Netherlands Academy for Arts and Sciences, Amsterdam, Netherlands.

[4] Amsterdam UMC, Vrije Universiteit Amsterdam, Department of Psychiatry, Amsterdam Neuroscience, Amsterdam, Netherlands

Contact information

Anouk van der Straten, MD

Amsterdam UMC, University of Amsterdam

Department of Psychiatry,

Meibergdreef 5

1105 AZ Amsterdam

The Netherlands

Tel: +31 20 8913600

a.l.vanderstraten@amc.nl

Abstract

Introduction

The classical cognitive-behavioral theory of obsessive-compulsive disorder (OCD) holds that compulsions (repetitive behaviors) are performed to reduce distress that is evoked by obsessions (intrusive thoughts), whereas a recent neuroscience-inspired theory suggests that compulsivity results from a disbalance between goal-directed and habit-related neural networks. Stress exacerbates OCD symptoms in patients but also shifts goal-directed behavior towards habitual behavior in healthy volunteers. To bridge these theories, we investigated whether acute stress would influence the balance between goal-directed and habit networks in patients with OCD.

Methods

Twenty-three OCD patients and twenty-three healthy controls participated in a controlled stress induction paradigm using the socially evaluated cold-pressor test in a crossover design. Stress responses were evaluated through cortisol, blood pressure and heart rate measurements and anxiety ratings, while brain activity was measured with resting-state functional magnetic resonance imaging. A seed-to-voxel analysis was performed for the caudate nucleus and posterior putamen as hubs of the goal-directed and habit network, respectively.

Results

Stress induction increased blood pressure and psychological stress measures across groups and resulted in blunted cortisol responses in patients. Furthermore, acute stress increased connectivity between the caudate nucleus and precuneus in patients compared to healthy controls, which was positively correlated with compulsivity but not obsession severity. The posterior putamen showed no significant group differences in stress-induced connectivity.

Conclusion

Acute stress increases the interaction between the goal-directed and default mode network in OCD, which is associated with compulsivity severity. These findings suggest that stress-related compulsivity involves increased recruitment of the goal-directed network.

Introduction

Obsessive-compulsive disorder (OCD) is a debilitating psychiatric disorder with an estimated lifetime prevalence of 1-3% [1]. Patients with OCD suffer from obsessions, defined by recurrent intrusive anxious thoughts or images, and/or compulsions, defined by repetitive behaviors or mental acts. Although the patients are usually aware of the senselessness of the compulsions they are performing, they are unable to inhibit their behaviors. Currently, two apparently opposing theories have been postulated to explain compulsivity in OCD. The classic cognitive-behavioral theory proposes that performing compulsions is primarily a strategy to reduce distress that is evoked by obsessions [2]. This theory provides an explanation of OCD symptoms that forms the basis of current cognitive-behavioral therapy. However, the variety in phenomenology shows that the relationship between obsessionality and compulsivity is more complex [3]. Recent neuroscientific studies suggest that patients with OCD show a general impairment in goal-directed behavior, resulting in an overreliance on habits [4, 5]. Goal-directed behavior consists of actions that are performed to achieve a desired goal. When actions are performed on a regular basis, habits are formed to facilitate actions that do not require planning or organization. This leads to greater efficiency, but often at the cost of behavioral flexibility. Along these lines, the overreliance on (maladaptive) habitual behavior is thought to contribute to the development of compulsivity in OCD [6]. Both rodent and human studies have pointed towards the involvement of the posterior putamen and the caudate nucleus in the habit and goal-directed network, respectively [6-10]. Both structures are part of the corticostriatal circuitry, the dysfunction of which is seen as the neuroanatomical basis of OCD [11, 12]. The recent focus on the habit hypothesis became apparent in the discussion around the new DSM-5, in which OCD moved out of the chapter ‘anxiety disorders’ to obtain its own chapter: i.e. ‘obsessive–compulsive and related disorders’ [13].

The habit theory however, does not account for the typical clinical observation that psychological stress exacerbates OCD symptoms [14]. Furthermore, recent studies in healthy humans have shown that acute stress reduces prefrontal cortex-related goal-directed control and thereby induces a bias towards reliance on habitual behavior [15, 16]. Integrating these findings leads to the hypothesis that stress may reduce goal-directed control and induce a shift towards habits in patients, thereby contributing to the compulsivity

seen in OCD [6, 17]. However, this novel model that connects the critical role of stress as posited by the classical cognitive-behavioral theory with the neuroscience-inspired habit theory of OCD remains to be tested. In this study, we investigated the effect of controlled stress induction on resting-state functional connectivity of both the caudate nucleus (as major hub within the goal-directed network), and the posterior putamen (as major hub of the habit network) comparing OCD patients with matched healthy controls. We hypothesized that in OCD patients, acute stress would reduce connectivity within the goal-directed network (i.e. between the caudate nucleus and the ventromedial prefrontal cortex) and/or increase connectivity within the habit network (i.e. between the posterior putamen and supplementary motor area).

Methods

Participants

Twenty-five OCD patients were recruited from the outpatient clinic for anxiety disorders at the psychiatry department of the Amsterdam UMC and through advertisement at a Dutch patient organization website. Inclusion criteria were (1) being aged 18-65 years; (2) having a diagnosis of OCD according to the DSM-IV; (3) having a Yale-Brown Obsessive-Compulsive Scale (Y-BOCS) score of 12 or higher; (4) having provided written informed consent and being willing and able to understand, participate and comply with the study requirements. Exclusion criteria were (1) having a current major depressive disorder, bipolar disorder, psychotic disorder or alcohol or substance dependence as assessed with the Mini-International Neuropsychiatric Interview (M.I.N.I. [18]); (2) having a history of major head trauma or neurological disease; (3) any MRI contraindications; (4) a self-reported inability or unease to stop smoking 3 hours prior to testing; (5) having any endocrinological disorder or regularly using corticosteroids; (6) undergoing a current treatment with antipsychotic medication; (7) using benzodiazepines or recreational drugs 72 hours prior to each test session or using alcohol within 24 hours before each measurement; and (8) having an irregular sleep/wake rhythm. In addition, twenty-five healthy controls without a current or past psychiatric diagnosis as assessed with the M.I.N.I. and matched according to age, sex, and educational level, were recruited through flyers and online advertisements. All participants were assessed on anxiety, depressive and obsessive-compulsive symptoms, using the Hamilton Anxiety Rating Scale (HAM-A) [19], the Hamilton Depression Rating Scale (HDRS) [20] and the YBOCS [21], respectively. In addition, the

presence of psychiatric co-morbidities was assessed with the M.I.N.I. [18] and the Structured Interview for DSM-IV Personality (SIDP-IV) [22]. This study was approved by the Ethical Committee of the Academic Medical Center in Amsterdam (METC 2014_168) and all participants provided written informed consent.

Design and measurements

The study used a cross-over design with counterbalanced order of stress induction vs. neutral condition across participants, with the two sessions separated by a 1 week interval. All the sessions took place in the afternoon to control for the normal daily fluctuations of cortisol. During the stress session, participants were exposed to the socially evaluated cold-pressor test (SECPT). This is a relatively short, extensively studied procedure which is known to induce a reliable physiological and subjective stress response through a minimally invasive method [23]. First, participants had to hold their right hand and wrist in ice water (1-3°C) for three minutes while being videotaped and monitored by an unfamiliar investigator. This unfamiliar person was instructed to wear a white lab coat and act unsympathetically, thereby inducing social stress. Afterwards, participants were asked to count backwards from 2059 in steps of 17. In case the participant made a mistake they were told to start over. During the control session, participants had to immerse their right hand in warm water (35-37°C) without being videotaped or monitored by an unfamiliar investigator. Following the warm water procedure, they were asked to count to 600 in steps of 10. The physiological stress response was examined by measuring blood pressure and heart rate at five time points: at baseline, once during the water immersion procedure, after the math task, before the scanning session (45 minutes before the resting-state scan), and after the scanning session (40 minutes after the resting-state scan), see Figure 1. In addition, cortisol was measured through saliva sampling at four time points: at baseline, after the math task, right before the resting-state scan, and after the scanning session. Moreover, to assess the subjective stress response, participants were asked to rate their anxiety level by filling out the State-Trait Anxiety Inventory (STAI [24]) at four time points: prior to the water procedure, after the math task, right before the resting-state scan, and after the scanning session. Furthermore, participants had to rate how difficult, unpleasant, and stressful they experienced the stress procedure right after the math task using a subjective stress level questionnaire with a VAS scale [23].

Because this study was part of a larger set of experiments, the resting-state scan was conducted approximately 65 minutes after the stress induction procedure. At the end of the stress session there was a short debriefing, in which the participants were told that the purpose of the test was to create stress. For an overview of the study design and measurements, see Figure 1.

Data acquisition

MR imaging was performed on a 3.0 T Philips MRI scanner (Philips, Best, The Netherlands), using a 32-channel SENSE head coil. The scanning protocol included a high-resolution T1-weighted MRI (sequence parameters: repetition time=6.9ms; echo time=3.1ms; flip angle=8°; 150 sagittal slices; voxel size=1.1mm isotropic, total duration=179s). For fMRI, multi-echo echoplanar imaging (EPI) was used to acquire T2*-weighted MRI volumes with blood oxygen level-dependent (BOLD) contrast (sequence parameters: repetition time=2375ms; number of echos=3, echo time=9, 26 and 44ms; flip angle=76°; field of view=224mm x 122mm x 224mm; voxel size=3.0 mm isotropic; 37 slices; 200 volumes with a total duration of 8 minutes). Multi-echo EPI reduces signal dropout and distortion while enhancing BOLD contrast sensitivity [25]. Three dummy scans were acquired to allow for equilibration of the signal. Head immobilization was established using foam pads inside the coil. Participants were instructed to stay awake and relax with their eyes open while thinking of anything that came to their mind.

Data analysis

Demographical and clinical data were analysed with SPSS software (version 24.0, Chicago, IL, USA). Group differences were tested using Mann-Whitney U tests for continues not normally distributed variables (age, Y-BOCS score, HAM-A, HDRS, mean FD) and $\chi 2$ tests for gender, handedness and education. The physiological stress measures (blood pressure, heart rate and cortisol) and anxiety levels (STAI) were analysed using mixed model analysis of variance (ANOVA) with the factors group (patients, healthy controls), condition (stress, control) and time (five time points for blood pressure and heart rate, four for anxiety), corrected for the order of sessions. No time factor was used for cortisol, as we calculated the area under the curve with respect to the ground (AUCg) to assess overall exposure to cortisol during the session [26]. We additionally compared baseline cortisol values between the conditions to ensure that

the results were not due to significant differences at baseline. Baseline cortisol and AUCg values were log-transformed before testing because of a non-normal distribution. Each question of the subjective stress level questionnaire was entered in a mixed model ANOVA with the factors factors group (patients, healthy controls), condition (stress, control) and question type (how difficult, how stressful, how unpleasant), corrected for the order of sessions. When the assumption of sphericity was violated the ANOVA results were corrected with a Greenhouse Geisser correction. The analyses were followed by post-hoc paired-t-testing in case of significance, with Bonferroni correction for multiple comparisons.

MRI analysis

Functional MRI data were preprocessed using SPM12 (www.fil.ion.ucl.ac.uk/spm/) and the CONN toolbox v18 (http://www.nitrc.org/projects/conn) in Matlab version 2014b (http://www.mathworks.com). First, realignment was performed and the three echoes were combined using in-house software to optimize BOLD contrast sensitivity and minimize signal dropout and distortion [25]. This was followed by slice-time correction. Artifacts were detected for scrubbing using the ART-based functional outlier detection. Volumes were considered outliers if they exceeded the global z signal threshold of 9 or the motion threshold of 2mm. Following co-registration of the mean EPI to the T1-weighted structural scan, functional images were normalized to Montreal Neurological Institute (MNI) space and smoothed with a 3D Gaussian kernel of 6mm at FWHM. Participants with realignment parameters exceeding 3mm translation on the x-, y-, or z-axis were excluded from the analysis, resulting in the exclusion of two patients. One healthy control was excluded due to an incidental neurological finding and one control was excluded because of insufficient brain coverage due to inaccurate setting of the field of view. This led to the inclusion of twenty-three patients and twenty-three healthy controls for further analysis.

After preprocessing, six principal components were derived from white matter and ventricles and they were included as nuisance parameters using CompCor [27]. Motion-related artefacts were accounted for by regressing out realignment parameters. In addition, volumes that exceeded the threshold of global signal change (i.e. Z-value=9) or motion (i.e. 2mm) were considered an outlier and were censored using scrubbing. Based on our hypothesis, we selected four seeds (i.e. left and right caudate for the goal-directed network, left and right posterior putamen for the habit network) for further analysis using the

Harvard-Oxford atlas. The putamen was delineated at y=2 as described in previous research [8], resulting in the region y≤2 for the posterior putamen. Because the caudate nucleus and posterior putamen are located closely to each other, the timeseries from the ipsilateral seed were regressed out to ensure that the results were specific for the selected seeds and did not reflect striatal connectivity in general. The resulting residual time series were then band-pass filtered at 0.008-0.09 Hz and were used to create individual connectivity maps through bivariate correlation and Fisher's r-to-z transformation [28]. We performed a seed-to-voxel analysis for the selected seeds (i.e. left and right caudate, left and right posterior putamen) to investigate whether there were group differences in stress-induced connectivity.

Connectivity maps were entered into a second-level group (patient, healthy control) X condition (stress, control) mixed model ANOVA, corrected for age, gender and the order of sessions (i.e. stress induction at first or second session). Post-hoc analyses were performed in case of significance to determine the direction of the interaction effect. In addition, we analyzed group differences during the control visit with a two sample t-test to assess whether patients showed altered connectivity without the presence of stress, again corrected for age, gender and the order of sessions. Voxel-wise statistical tests were family wise error (FWE) rate corrected (two-sided; $P<0.05$) for multiple comparisons across the whole brain at cluster level using an initial height threshold of $P<0.001$. Because we were most interested in connectivity within both hypothesized networks, we performed a small volume correction for the regions targeted by our seeds, i.e. the supplementary motor area (SMA) for the posterior putamen and the ventromedial prefrontal cortex (vmPFC) for the caudate nucleus. We used the WFU Pickatlas toolbox in SPM12 to define the SMA ROI according to the anatomical automatic labeling (AAL) atlas. For the goal-directed ROI, we selected coordinates of the vmPFC that have shown to be active during goal-directed behavior in an instrumental discrimination task using a sphere with 10mm radius [29]. Movement during scanning was investigated by calculating the mean framewise displacement (FD) for each scanning session, and was compared between groups with a Mann-Whitney U test. To further investigate whether the imaging results were linked to the severity and type of symptoms, ratings on the obsession and compulsion scale of the YBOCS were correlated with the connectivity results extracted from the significant clusters using a Pearson correlation in SPSS.

Results

Demographic and clinical data

Demographic and clinical data are presented in Table 1. There were no significant group differences in age, sex distribution, level of education and movement during scanning. As expected, ratings on the Y-BOCS, HAM-A and HDRS were significantly higher in patients than in controls. Patients had various current comorbid disorders, including panic disorder (n=2), agoraphobia (n=1), hypochondria (n=1), generalized anxiety disorder (n=1), social anxiety disorder (n=1), past major depressive disorder (n=6) and past alcohol abuse (n=1). In addition, three patients matched the criteria for a personality disorder, i.e. obsessive-compulsive, antisocial and avoidant personality disorder. Furthermore, nineteen patients used a stabile dose of medication during the two visits, including fluoxetine, (es)citalopram, sertraline, paroxetine, clomipramine or venlafaxine.

Stress response

Blood pressure and heart rate were analyzed using a mixed model ANOVA with the factors group (patients, healthy controls), condition (stress, control) and time (five time points), corrected for the order of sessions. For systolic blood pressure, this analysis revealed a main effect of condition with higher systolic blood pressure in the stress condition ($F(1,35)=4.64$, $p=0.038$, $\eta_p^2=0.12$), a main effect of time ($F(4,140) = 3.20$, $p=0.015$, $\eta_p^2=0.08$) and a condition X time interaction ($F(4,140)=8.91$, $p<0.001$, $\eta_p^2=0.20$). The condition X time interaction was due to higher systolic blood pressure after the math task during the stress condition compared to the control condition across groups ($t(45)=6.35$, $p<0.001$). The analysis of the diastolic blood pressure revealed a main effect of condition with higher diastolic blood pressure in the stress condition ($F(1,34)=4.24$, $p=0.047$, $\eta_p^2=0.11$). The analysis of the heart rate did not show any significant results. For cortisol, the analysis of baseline level showed no significant effects. We then calculated the AUCg and entered these values into an interaction analysis with the factors group (patients, healthy controls) and condition (stress, control). The analysis revealed a group X condition interaction ($F(1,43)=6.36$, $p=0.015$, $\eta_p^2=0.13$). Post-hoc testing showed significantly higher cortisol levels in the stress condition than the control condition in healthy controls ($t(22)=3.83$, $p=0.001$) but not in patients. The analysis of subjective anxiety levels, as measured with the STAI, revealed a main effect of condition with

higher anxiety levels in the stress condition ($F(1,40)=5.20$, $p=0.028$, $\eta_p^2=0.12$), a main effect of time ($F(2.08,83.12)=4.45$, $p=0.014$, $\eta_p^2=0.10$) and a condition X time interaction ($F(2.61,104.28)=12.31$, $p<0.001$, $\eta_p^2=0.24$). In addition, there was a significant main effect of group ($F(1,40)=46.3$, $p<0.001$, $\eta_p^2=0.54$), indicating that patients reported higher anxiety levels than healthy controls overall. Post-hoc paired t-testing showed that participants in both groups reported significantly higher anxiety levels in the stress condition compared to the control condition after the stress procedure ($t(45) = 7.17$, $p<0.001$), with a trend towards significance during scanning ($t(43) = 2.22$, $p=0.032$). The analysis of the subjective stress level questionnaire showed a main effect of condition with higher ratings in the stress condition ($F(1,42)=105.99$, $p<0.001$, $\eta_p^2=0.72$). Analyzing the specific subscales showed that participants in both groups rated the stress condition as more difficult ($t(44)=12.09$, $p<0.001$), more unpleasant ($t(44)=13.02$, $p<0.001$), and more stressful ($t(44)=10.8$, $p<0.001$) than the control condition. Together, these results show that the stress condition successfully increased blood pressure and psychological stress measures in both groups, and that group differences were only present for the stress-induced cortisol response, and anxiety levels (independent of stress induction).

Seed-to-voxel analysis

To assess the effects of stress induction on these networks, we performed a mixed model ANOVA with the factors group (patients, healthy controls) and condition (stress, control), corrected for the order of sessions. This analysis revealed a significant interaction for the goal-directed seeds (for significant clusters, see Table 2. Compared to healthy controls, patients showed increased connectivity between the left and right caudate nuclei and precuneus after stress induction (Figure 3). This interaction effect was mainly driven by decreased connectivity after stress in healthy controls between the right caudate nucleus and precuneus (MNI: -10, -62, 40, cluster size=209, p(FWE)<0.001) and the left caudate nucleus and precuneus (MNI: -4, -66, 44, cluster size=126, p(FWE)=0.004). The increase in connectivity after stress in patients was not significant. We found no significant clusters in the vmPFC using a small volume correction. For the habit seeds, we found no significant group differences in connectivity after stress between the posterior putamen and the SMA, nor with other regions of the brain. Furthermore, there was no main effect of stress across patients and controls in any of the seeds.

We further investigated whether the connectivity results were linked to the severity and type of symptoms, by correlating the extracted values from the significant clusters of the stress versus control contrast with the ratings on the obsession and compulsion scale of the Y-BOCS in patients only. This showed a significant correlation between the connectivity of the right caudate nucleus and precuneus and ratings on the compulsion scale of the Y-BOCS ($r=0.49$, $n=23$, $p=0.017$, see Figure 4), indicating that patients who are more compulsive show a larger increase in connectivity after acute stress. There were no significant correlations between the imaging results and the rating on the obsession scale of the Y-BOCS.

We then analyzed group differences during the control session with a two sample t-test to see whether patients showed altered connectivity without the presence of stress, corrected for age, gender and the order of sessions. Compared to healthy controls, patients showed significantly lower connectivity between the caudate nucleus and lateral occipital cortex, the angular gyrus, the middle and superior frontal gyrus, the precuneus cortex, the posterior cingulate gyrus and the frontal pole ($p(FWE)<0.05$, for significant clusters see Table 3. Both the left and right posterior putamen showed no significant differences in baseline connectivity between patients and controls.

Discussion

This is the first study investigating the effects of acute stress on intrinsic functional connectivity in patients with OCD, which aimed to test the hypothesis that stress induces a shift from the goal-directed towards the habit network. Using a seed-to-voxel resting-state analysis, we found that stress increased coupling between the seeds involved in the goal-directed network (i.e. the left and right caudate nucleus) and the precuneus in patients compared to healthy controls. This result was positively correlated with the severity of compulsive symptoms. Stress did not have an effect on functional connectivity between the seeds involved in the habit network (i.e. the left and right posterior putamen) and the rest of the brain, including the supplementary motor areas. Measures of blood pressure and subjective anxiety confirmed successful stress induction across groups, and showed significantly lower cortisol responses in patients compared to healthy controls. Together, these results suggest that stress-induced compulsivity is primarily associated with increased connectivity of the goal-directed network. The habit theory of OCD is centered around the

balance between the goal-directed and habit networks, but ignores the influence of distress. Distress however, is a central aspect of the cognitive-behavioral theory of OCD. Our results therefore link the apparent contradictory theories of OCD and suggest that both factors contribute to the pathophysiology of OCD.

The results were however not fully in line with our hypothesis, as stress did not affect intrinsic connectivity within the habit network, and we did not observe reduced connectivity of the goal-directed network with the ventromedial prefrontal cortex. Instead, the results point towards altered connectivity between the caudate nucleus and the precuneus, an area that is part of the default mode network (DMN). The precuneus is involved in self-consciousness and self-referential processing, defined as the process of relating information to the self [30, 31]. When individuals are not involved in an attention demanding or goal-directed task, the DMN is activated and self-referential processing is believed to predominate [32]. Compared to healthy controls, patients showed decreased baseline connectivity between the goal-directed network and the precuneus. After stress, healthy controls showed uncoupling of the goal-directed network and DMN, which implies that stress normally induces upscaling of the goal-directed network at the cost of the DMN. In contrast, OCD patients did not show a decrease in connectivity between the caudate nucleus and precuneus after stress, suggesting that patients might be stuck in self-referential processing at the cost of goal-directed behavior. Almost all OCD symptoms can be narrowed down to the underlying desire for absolute certainty and maintaining control, therefore obsessions usually develop in domains where there is no absolute level of certainty, such as sexuality, sickness and danger [3]. Unfortunately patients do not base the level of certainty on external reality, but on feelings of anxiousness and unrealistic thoughts. This causes them to continuously focus on themselves, resulting in a mental loop of self-referential processing. The lack of downscaling of the DMN that was seen in patients might thus be the result of compulsive self-referential processing under stress. The stress-induced connectivity between the right caudate nucleus and the precuneus was positively correlated with the compulsion scale of the Y-BOCS, indicating that patients who are prone to compulsive behavior are more likely to show an increase in connectivity after stress. This further confirms the assumption that our results can be interpreted as stress-induced self-referential processing at the cost of goal-directed control, resulting in mental compulsions. Remarkably, we found no stress induced coupling between the posterior putamen and SMA.

This leads to the speculation that compulsions are more strongly related to impaired goal-directed control than to overdependence on habits. Our null finding could be due to the study design, as the motor circuit is not probed during resting-state. Future research is needed to see whether these results are confirmed using a task that requires goal-directed behavior for further understanding of the pathophysiology of OCD.

In addition to the group differences in connectivity, patients also showed lower stress-induced cortisol levels compared to healthy controls. This blunted cortisol response could be due to adaptation of stress circuits as a consequence of chronic exposure to stress. This mechanism is seen across multiple psychiatric disorders including post-traumatic stress disorder, major depressive disorder and panic disorder [33]. Similarly, previous studies have also shown a non-response of cortisol during stressful exposure and response prevention therapy in OCD [34, 35].

Without the presence of stress, the goal-directed seeds showed extensive group differences in connectivity with frontal areas, whereas the habit seeds showed no differences in connectivity. This is consistent with the hypothesis that patients already show an impairment in the goal-directed network, regardless of the presence of induced stress [4]. The finding of reduced connectivity between the caudate and these cortical regions is inconsistent with corticostriatal hyperconnectivity that is often described in OCD [36-38], but has previously been demonstrated in research conducted in unmedicated patients [39].

This study has several limitations. Resting-state fMRI is a promising technique for investigating the functional architecture of the brain, but the results provide no information on the direction of the connectivity. Therefore, we can only speculate on causal relationships between significant nodes [40]. Second, the resting-state scan was not conducted immediately following the stress induction which makes it difficult to determine whether the participants were still in acute stress or already in the recovery phase. This study was part of a larger trial, therefore the resting-state scan was conducted approximately 65 minutes after the SECPT. This is of importance because the effects of stress-related neuromodulators can vary over time. Catecholaminergic and dopaminergic systems are activated directly after a stressor, while the slower genomic effects of corticosteroids can continue for several hours [41]. Participants still rated their anxiety level to be higher before the resting-state scan started in the stress visit compared to the control visit, implicating that the stress response was prolonged during the scanning session. Third, the majority of our patients were using medication and had various comorbid disorders which could have

influenced the imaging results. Despite these limitations, our results show that acute stress influences functional connectivity of the goal-directed network in OCD. These findings provide first insight into the effects of stress on the balance between the goal-directed and habit network in OCD, implicating that stress-induced compulsivity is predominantly associated with increased connectivity of the goal-directed network.

References

1. Ruscio, A.M., et al., *The epidemiology of obsessive-compulsive disorder in the National Comorbidity Survey Replication.* Mol Psychiatry, 2010. **15**(1): p. 53-63.
2. Salkovskis, P.M., *Obsessional-compulsive problems: a cognitive-behavioural analysis.* Behav Res Ther, 1985. **23**(5): p. 571-83.
3. Denys, D., *Obsessionality & compulsivity: a phenomenology of obsessive-compulsive disorder.* Philos Ethics Humanit Med, 2011. **6**: p. 3.
4. Gillan, C.M., et al., *Disruption in the balance between goal-directed behavior and habit learning in obsessive-compulsive disorder.* Am J Psychiatry, 2011. **168**(7): p. 718-26.
5. Gillan, C.M., et al., *Enhanced avoidance habits in obsessive-compulsive disorder.* Biol Psychiatry, 2014. **75**(8): p. 631-8.
6. Gillan, C.M., et al., *The role of habit in compulsivity.* Eur Neuropsychopharmacol, 2016. **26**(5): p. 828-40.
7. Balleine, B.W. and J.P. O'Doherty, *Human and rodent homologies in action control: corticostriatal determinants of goal-directed and habitual action.* Neuropsychopharmacology, 2010. **35**(1): p. 48-69.
8. de Wit, S., et al., *Corticostriatal connectivity underlies individual differences in the balance between habitual and goal-directed action control.* J Neurosci, 2012. **32**(35): p. 12066-75.
9. Alexander, G.E., M.D. Crutcher, and M.R. DeLong, *Basal ganglia-thalamocortical circuits: parallel substrates for motor, oculomotor, "prefrontal" and "limbic" functions.* Prog Brain Res, 1990. **85**: p. 119-46.
10. Watson, P., G. van Wingen, and S. de Wit, *Conflicted between Goal-Directed and Habitual Control, an fMRI Investigation.* eNeuro, 2018. **5**(4).
11. Radua, J., et al., *Meta-analytical comparison of voxel-based morphometry studies in obsessive-compulsive disorder vs other anxiety disorders.* Arch Gen Psychiatry, 2010. **67**(7): p. 701-11.
12. Milad, M.R. and S.L. Rauch, *Obsessive-compulsive disorder: beyond segregated cortico-striatal pathways.* Trends Cogn Sci, 2012. **16**(1): p. 43-51.
13. American Psychiatric Association, *Diagnostic and statistical manual of mental disorders (5th ed.)*. 2013, Arlington, VA: American Psychiatric Publishing.
14. Rachman, S., *A cognitive theory of obsessions: elaborations.* Behav Res Ther, 1998. **36**(4): p. 385-401.
15. Schwabe, L. and O.T. Wolf, *Stress prompts habit behavior in humans.* J Neurosci, 2009. **29**(22): p. 7191-8.
16. Otto, A.R., et al., *Working-memory capacity protects model-based learning from stress.* Proc Natl Acad Sci U S A, 2013. **110**(52): p. 20941-6.
17. Adams, T.G., et al., *The role of stress in the pathogenesis and maintenance of obsessive-compulsive disorder.* Chronic Stress (Thousand Oaks), 2018. **2**.
18. Sheehan, D.V., et al., *The Mini-International Neuropsychiatric Interview (M.I.N.I.): the development and validation of a structured diagnostic psychiatric interview for DSM-IV and ICD-10.* J Clin Psychiatry, 1998. **59 Suppl 20**: p. 22-33;quiz 34-57.
19. Hamilton, M., *The assessment of anxiety states by rating.* Br J Med Psychol, 1959. **32**(1): p. 50-5.
20. Hamilton, M., *A rating scale for depression.* J Neurol Neurosurg Psychiatry, 1960. **23**: p. 56-62.
21. Goodman, W.K., et al., *The Yale-Brown Obsessive Compulsive Scale. I. Development, use, and reliability.* Arch Gen Psychiatry, 1989. **46**(11): p. 1006-11.
22. Pfohl, B.B., N.; Zimmerman, M.; Stangl, D.;, *Structured interview for DSM-IV personality (SIDP-IV)*. 1997, Washington (DC): American Psychiatric Association.
23. Schwabe, L., L. Haddad, and H. Schachinger, *HPA axis activation by a socially evaluated cold-pressor test.* Psychoneuroendocrinology, 2008. **33**(6): p. 890-5.
24. D Spielberger, C., et al., *Manual for the State-Trait Anxiety Inventory (Form Y1 – Y2)*. Vol. IV. 1983.
25. Poser, B.A., et al., *BOLD contrast sensitivity enhancement and artifact reduction with multiecho EPI: parallel-acquired inhomogeneity-desensitized fMRI.* Magn Reson Med, 2006. **55**(6): p. 1227-35.

26. Pruessner, J.C., et al., *Two formulas for computation of the area under the curve represent measures of total hormone concentration versus time-dependent change.* Psychoneuroendocrinology, 2003. **28**(7): p. 916-31.
27. Behzadi, Y., et al., *A component based noise correction method (CompCor) for BOLD and perfusion based fMRI.* Neuroimage, 2007. **37**(1): p. 90-101.
28. Whitfield-Gabrieli, S. and A. Nieto-Castanon, *Conn: a functional connectivity toolbox for correlated and anticorrelated brain networks.* Brain Connect, 2012. **2**(3): p. 125-41.
29. de Wit, S., et al., *Differential engagement of the ventromedial prefrontal cortex by goal-directed and habitual behavior toward food pictures in humans.* J Neurosci, 2009. **29**(36): p. 11330-8.
30. Cavanna, A.E. and M.R. Trimble, *The precuneus: a review of its functional anatomy and behavioural correlates.* Brain, 2006. **129**(Pt 3): p. 564-83.
31. Northoff, G., et al., *Self-referential processing in our brain - A meta-analysis of imaging studies on the self.* Neuroimage, 2006. **31**(1): p. 440-457.
32. Buckner, R.L., J.R. Andrews-Hanna, and D.L. Schacter, *The brain's default network: anatomy, function, and relevance to disease.* Ann N Y Acad Sci, 2008. **1124**: p. 1-38.
33. Wichmann, S., et al., *Cortisol stress response in post-traumatic stress disorder, panic disorder, and major depressive disorder patients.* Psychoneuroendocrinology, 2017. **83**: p. 135-141.
34. Gustafsson, P.E., et al., *Diurnal cortisol levels and cortisol response in youths with obsessive-compulsive disorder.* Neuropsychobiology, 2008. **57**(1-2): p. 14-21.
35. Kellner, M., et al., *Non-response of cortisol during stressful exposure therapy in patients with obsessive-compulsive disorder--preliminary results.* Psychiatry Res, 2012. **199**(2): p. 111-4.
36. Harrison, B.J., et al., *Altered corticostriatal functional connectivity in obsessive-compulsive disorder.* Arch Gen Psychiatry, 2009. **66**(11): p. 1189-200.
37. Harrison, B.J., et al., *Brain corticostriatal systems and the major clinical symptom dimensions of obsessive-compulsive disorder.* Biol Psychiatry, 2013. **73**(4): p. 321-8.
38. Fitzgerald, K.D., et al., *Developmental alterations of frontal-striatal-thalamic connectivity in obsessive-compulsive disorder.* J Am Acad Child Adolesc Psychiatry, 2011. **50**(9): p. 938-948 e3.
39. Posner, J., et al., *Reduced functional connectivity within the limbic cortico-striato-thalamo-cortical loop in unmedicated adults with obsessive-compulsive disorder.* Hum Brain Mapp, 2014. **35**(6): p. 2852-60.
40. Cole, D.M., S.M. Smith, and C.F. Beckmann, *Advances and pitfalls in the analysis and interpretation of resting-state FMRI data.* Front Syst Neurosci, 2010. **4**: p. 8.
41. Hermans, E.J., et al., *Dynamic adaptation of large-scale brain networks in response to acute stressors.* Trends Neurosci, 2014. **37**(6): p. 304-14.

Acknowledgements

This work is supported by ZonMw grants no. 165.610.002 and 016.156.318.

Tables

TABLE 1; Demographic and clinical data presented as the mean ± SD.

	Patients (n=23)	Controls (n=23)	P-value
Age (years)	33.48 ± 2.02	33.52 ± 3.07	p=0.605[1]
Gender (male/female)	10/13	12/11	p=0.555[2]
Handedness (left/right)	2/21	4/19	p=0.381[2]
Highest education (n)	3/5/7/3/5	0/4/10/4/5	p=0.436[2]
– lower professional	3	0	
– secondary school	5	4	
– medium professional	7	10	
– higher professional	8	9	
Y-BOCS:			
– total	22.57 ± 1.24	0.13 ± 0.10	p<0.001[1*]
– obsessions	11.61 ± 0.64	0.13 ± 0.10	
– compulsions	10.52 ± 0.84	0.00 ± 0.00	
HAM-A	11.83 ± 1.57	1.39 ± 1.64	p<0.001[1*]
HDRS	8.17 ± 1.24	0.65 ± 0.19	p<0.001[1*]
Medication status (yes/no)	19/4	0/23	
Framewise displacement (mm)			
– control visit	0.14 ± 0.02	0.12 ± 0.01	p=0.538[1]
– stress visit	0.16 ± 0.03	0.12 ± 0.01	p=0.070[1]

* Significant difference between groups, $p \leq 0.05$, tested with [1] Mann-Whitney U test or [2] chi-square test. Scales: Yale-Brown Obsessive-Compulsive Scale (Y-BOCS), Hamilton Anxiety Rating Scale (HAM-A), Hamilton Depression Rating Scale (HDRS).

TABLE 2; Significant clusters in baseline connectivity comparison between patients and controls.

Seed	MNI coordinates			Cluster size	P-value	Region	BA
	x	y	z		(FWE)		
Healthy controls > OCD patients (control condition only)							
Caudate nucleus L	-46	-60	+36	477	<0.001	lateral occipital cortex L, angular gyrus L	39
	-34	+24	+50	357	<0.001	mid. and sup. frontal gyrus L	8
	-6	-64	+38	267	<0.001	precuneus cortex	7
	-4	-40	+26	243	<0.001	posterior cingulate gyrus	26
	+14	+64	+22	183	0.002	frontal pole R	10
	+28	+18	+40	106	0.037	mid. and sup. frontal gyrus R	9
Caudate nucleus R	-30	+16	+58	321	<0.001	mid. and sup. frontal gyrus L	8
	-2	-66	+30	204	<0.001	precuneus cortex	7
	-4	-48	+26	196	0.001	posterior cingulate gyrus	30
	-38	-58	+30	190	0.002	lateral occipital cortex L, angular gyrus L	39
	-12	+58	+20	161	0.004	frontal pole L	10
Posterior putamen L					N.S.		
Posterior putamen R					N.S.		

TABLE 3; Significant clusters from the seed-to-voxel group X condition interaction analysis.

Seed	MNI coordinates			Cluster size	P-value	Region	BA
	x	y	z				
OCD patients > healthy controls (stress > control condition)							
Caudate nucleus L	-6	-62	+38	92	0.038	precuneus	7
Caudate nucleus R	-4	-66	+42	467	<0.001	precuneus	7
Posterior putamen L					no clusters		
Posterior putamen R					no clusters		

FIGURE 1;

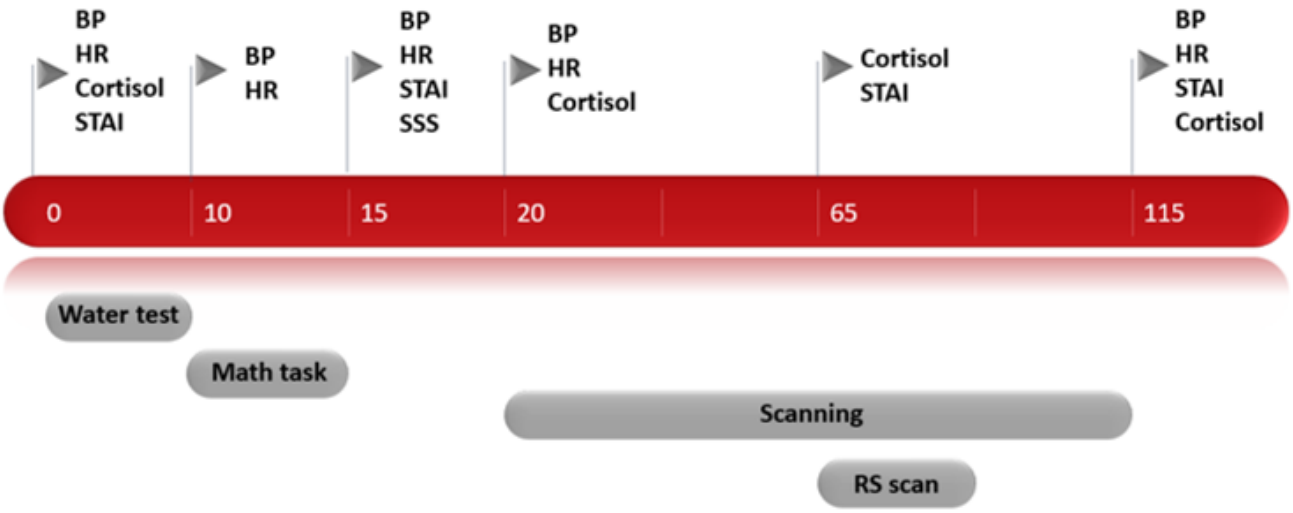


Figure 1. Timeline of experimental procedure and measurements. *Abbreviations: blood pressure (BP), heart rate (HR), State-Trait Anxiety Inventory (STAI), subjective stress scale (SSS) and resting-state (RS) scan.*

FIGURE 2; Physiological and subjective stress response, time points: baseline (0 min), after water test (10 min), after math task (15 min), before scanning session (20 min), before resting-state scan (65 min) and after scanning (115 min) reported as the mean with standard error.

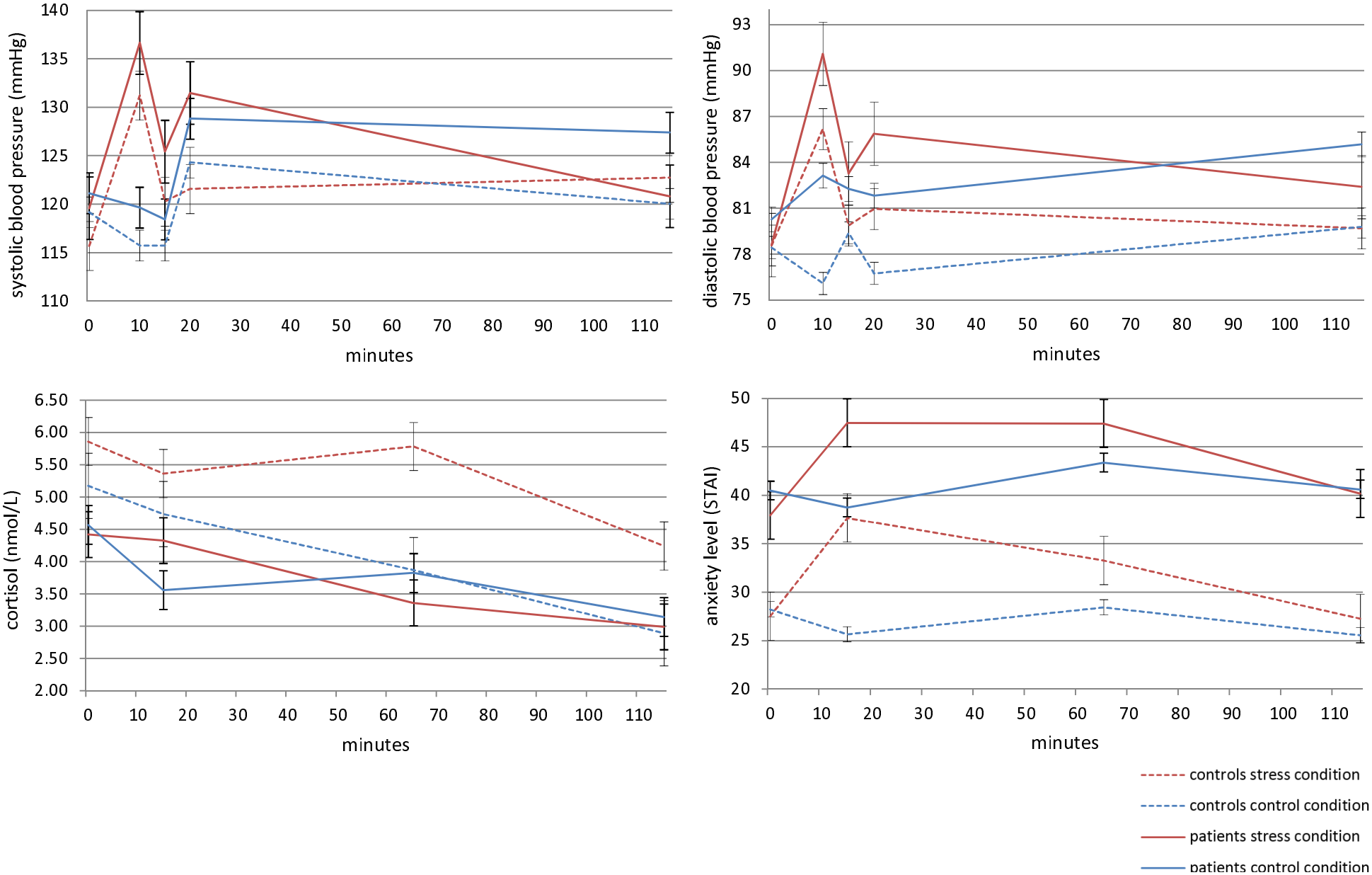
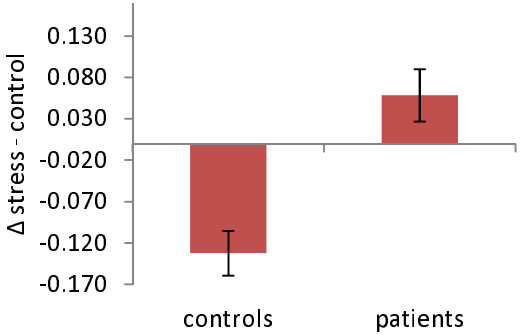


FIGURE 3; Stress effects in patients compared to healthy controls with the selected seeds, significant connectivity results (thresholded at p>0.001 for illustrational purposes) and contrast estimates extracted from the significant clusters.

Seed region: left caudate nucleus; OCD patients > healthy controls (stress > control condition)



Seed region: right caudate nucleus; OCD patients > healthy controls (stress > control condition)

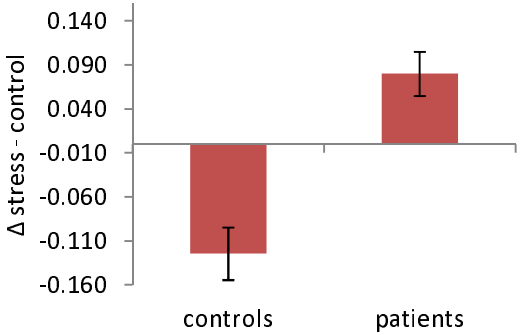
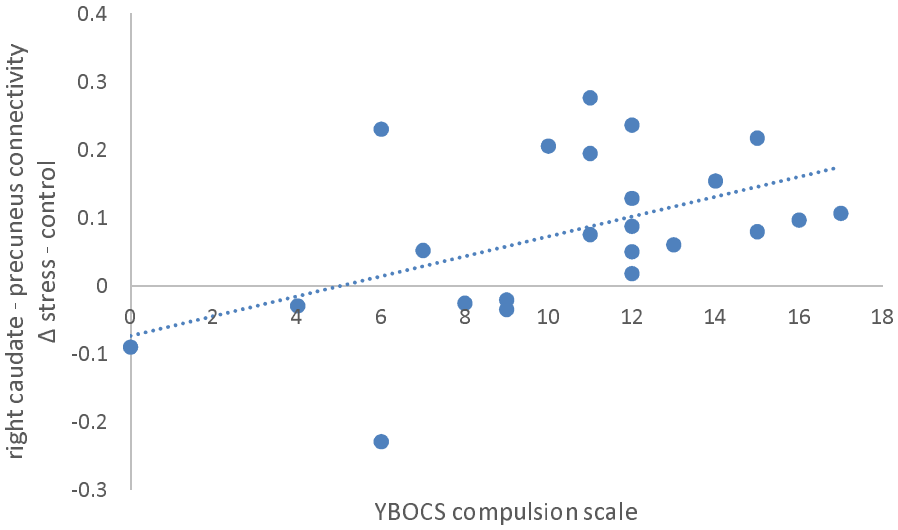



FIGURE 4; Correlation between the connectivity of the right caudate nucleus and precuneus and ratings on the compulsion scale of the Y-BOCS.