**Structural Hb definition**

The habenula was manually created bycoloring individual voxels using MRICron. In the geometric method, the habenula outline is created by defining three points, A, B and C.

****

**Figure S1. Structural Hb definition**

a, geometric method. In the geometric method, the habenula outline is created by defining three points, A, B and C. b, Examples of habenula (HB) masks created in native space for a representative participant. The first column shows the image intensity obtained for the habenula at 3T with the MP2Rage (without overlaid HB masks). The second column shows the masks created based on the geometric method.

**ROI definition of functional connectivity**

The seeds for the left and right habenula proposed by Kim et al. (Kim et al., 2016) were used in this study (Figure S2), which were thought to be suitable for habenula seed region selection in functional MRI applications. We selected several other regions of interest (ROI) for exploratory analysis: the bilateral Gpi, medial prefrontal cortex (mPFC), dorsomedial prefrontal cortex (dmPFC), ventromedial prefrontal cortex (vmPFC), Hippocampus, amygdala, PAG, VTA, and NAcc. Here, the masks of bilateral mPFC, vmPFC, dmPFC, hippocampus, amygdala, and NAcc were obtained using the WFU\_PickAtlas software package (<http://fmri.wfubmc.edu/software/>PickAtlas) and extracted in the AAL altas. The mask of bilateral Gpi as defined based on the Atlas of the basal ganglia (Keuken et al., 2014)([www.nitrc.org/projects/atag](http://www.nitrc.org/projects/atag)). The masks of VTA and PAG as defined based on the AAN atlas (Edlow et al., 2012) ([http://nmr.mgh.harvard.edu/martinos/ resources/aan-atlas](http://nmr.mgh.harvard.edu/martinos/%20resources/aan-atlas)).

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**Figure S2. The habenula regions of interest of functional analysis.**

Anatomical regions of interest were used for the left (red) and right habenula (blue). R, right.

**Comparison of demographic and clinical variables between unmedicated and medicated OCD**

Two sample *t* tests or Chi-square test were used to compare the demographic and clinical characteristics between unmedicated and medicated OCD.

Unmedicated OCD and medicated OCD patients showed no significant difference in demographic variables. Medicated OCD patients scored higher in obsession and Y-BOCS total. No other significant difference was found in other clinical variables.

Table S2 Comparison of demographic and clinical variables between unmedicated and medicated OCD

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
|  | Unmedicated (n=46) | Medicated (n=34) | **(χ2/*t*)** | ***p*** |
| **Age (years)** | 22.63±4.45 | 22.41±4.16 | 0.22 | 0.824 |
| **Gender(male/female)** | 25/21 | 18/16 | 0.02 | 0.901 |
| **Education (years)** | 14.32±2.07 | 14.59±2.84 | -0.50 | 0.620 |
| **BDI** | 20.89±8.00 | 20.71±12.10 | 0.08 | 0.935 |
| **TAI** | 57.74±7.38 | 56.79±9.28 | 0.51 | 0.612 |
| **SAI** | 55.34±12.95 | 53.35±12.08 | 0.70 | 0.487 |
| **OCI-R** | 32.00±11.86 | 31.76±12.28 | 0.09 | 0.929 |
| washing | 5.04±3.61 | 6.29±3.54 | -1.54 | 0.127 |
| checking | 5.96±3.48 | 5.50±2.86 | 0.62 | 0.535 |
| ordering | 5.50±3.20 | 5.06±2.78 | 0.64 | 0.522 |
| hoarding | 3.85±3.02 | 3.61±2.59 | 0.37 | 0.712 |
| obsessing | 7.02±2.62 | 7.47±2.98 | -0.72 | 0.477 |
| mental neutralizing | 4.63±3.23 | 3.82±2.81 | 1.17 | 0.247 |
| **Y-BOCS** | 19.40±6.13 | 18.10±5.63 | **-2.05\*** | **0.043** |
| obsession | 9.17±3.28 | 9.59±3.53 | -0.72 | 0.472 |
| compulsion | 10.12±8.50 | 8.50±4.72 | **-2.25\*** | **0.027** |

***Note.*** OCD, Obsessive-compulsive disorder; BDI, the score of the Beck Depression Scale; SAI, the score of the state anxiety scale; TAI, the score of the trait anxiety scale; OCI-R, the score of the Obsessive-Compulsive Inventory-Revised; Y-BOCS, the score of the Yale–Brown Obsessive– Compulsive Scale. \* Significance level of 0.05.

**Medication status effect on MRI results**

Covariance analyses were used to compare the Hb volume between unmedicated and medicated OCD with age, gender, and education as covariables. For whole-brain functional connectivity analysis, two sample *t* test was performed to determine significant difference between unmedicated and medicated OCD, controlling the age, gender, and education. The statistical parametric maps were thresholded at the voxel-level familywise error-corrected (FWE) *p* < 0.05, with an initial voxel-wise threshold of *p* < 0.001 by SPM12. Then we ran small volume correction (SVC) with the hippocampus and dmPFC (FWE*p* <0.05). The medication status (unmedicated vs. medicated) was included as a covariate in DCM within OCD group. The medication status effect was reported if there was a posterior probability > 0.95.

There was no significant medication status effect on the grey matter volume (Table S2), functional connectivity of the Hb-hippocampus and effective connectivity of habenula (Hb).

**Table S3 Comparison of GMV in habenula between unmedicated and medicated OCD**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **GMV** | **Unmedicated (n=46)** | **Medicated (n=34)** | **F** | ***p*** |
| **left Hb** | 37.76±17.50 | 33.05±14.28 | 1.55 | 0.218 |
| **right Hb** | 34.69±16.71 | 32.83±17.59 | 0.36 | 0.548 |

***Notes.***GMV, gray matter volume.

**Replication analysis**

***Participants of replication dataset***

The MRI data of replication dataset were obtained from the cooperative research group in the South China Normal University. Thirty-eight OCD patients were recruited from outpatient clinics affiliated with the Second Xiangya Hospital of Central South University in Changsha, Hunan, China. All patients fulfilled met DSM-V criteria for OCD and were diagnosed by two experienced psychiatrists using the Structured Clinical Interview for DSM-IV. The inclusion criteria for the OCD group were (1) no other psychiatric diagnoses meeting the DSM-V criteria except for OCD; (2) aged 16-35 years old; (3) right-handed; (4) having at least nine years of education. Thirty-four healthy controls (HCs) were also recruited from community matched for age, gender, and years of education. The inclusion criteria for HCs were (1) having no current or history of any psychiatric disorders; (2) aged 16-35 years old; (3) right-handed; (4) having at least nine years of education. Participants were excluded if they had: (1) a history of major medical or neurological problems (e.g., hypothyroidism, seizure disorder, or brain injury); (2) MRI contraindication; (3) being pregnant, lactating, or preparing for pregnancy; (4) inability to cooperate with the MRI procedure. One healthy control was excluded from the analyses due to excessive head motion in any direction more than 3.0 mm or/and 3.0。.

***Measurements of replication dataset***

*The Center for Epidemiologic Studies Depression Scale (CES-D)* assessed the depression level (Park & Yu, 2021), and *The State Anxiety Inventory (SAI)* assessed the anxiety level of all participants (Knowles & Olatunji, 2020; Spielberger et al., 1970). *The Yale–Brown Obsessive–Compulsive Scale (Y-BOCS)* assessed the severity of obsessive-compulsive symptoms of patients with OCD (Cohen, 1988; Lee et al., 2018).

***MRI acquisition of replication dataset***

All participants were scanned on a Siemens Skyra 3-T MRI system (Siemens Magnetom Skyra 3.0-T scanner) with foam padding to minimize head motion. The three-dimensional T1-weighted scans were acquired with a T1 weighted gradient echo sequence (T1-weighted Image, T1WI). The parameters were as follows: repetition time (TR) = 1900 ms, echo time (TE) = 2.01 ms, flip angle (FA) = 9◦,slice thickness = 1.0 mm, voxel size = 1 × 1 × 1 mm3, field of view (FOV) = 256 × 256 mm2, slice numbers = 176. The resting-state functional images were acquired with echo planar imaging sequence with the following parameters: TR = 2000 ms; TE = 30 ms, FA = 80◦, voxel size = 4 × 4 × 4 mm3, matrix = 64 × 64, FOV = 256 × 256 mm2, slice thickness = 4.0 mm, slice numbers = 32, volume =216.

***Same analyses were conducted in the replication dataset.***

***Results of the replication dataset***

*Demographic and clinic characteristics of HC and OCD*

There was no significant difference in age, gender and years of education between OCD and HC group. OCD patients had higher scores of CES-D and SAI than HCs (see Table S4).

**Table S4 Differences of demographic and clinical variables between OCD and HC groups**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
|  | **OCD(n=38)** | **HC(n=34)** | **(χ2/*t*)** | ***p*** |
| **Age(years)** | 21.84±4.69 | 20.24±2.02 | 1.91 | 0.061 |
| **Gender (male/female）** | 25/13 | 16/18 | 3.86 | 0.145 |
| **Education (years)** | 13.61±2.07 | 13.91±1.60 | -0.70 | 0.487 |
| **CES-D** | 46.68±11.58 | 34.20±8.48 | 5.17\*\*\* | <0.001 |
| **SAI** | 46.24±10.50 | 34.19±7.48 | 5.56\*\*\* | <0.001 |
| **Y-BOCS** | 19.32±5.64 | - | - | - |
| obsession | 10.21±2.52 | - | - | - |
| compulsion | 9.09±3.75 | - | - | - |

***Note.*** OCD, Obsessive-compulsive disorder; HC, healthy control; CES-D, the score of the Center for Epidemiologic Studies Depression Scale; SAI, the score of the state anxiety scale; Y-BOCS, Yale–Brown Obsessive–Compulsive Scale. \*\*\* Significance level of 0.001.

*Volumetric analysis of the habenular nucleus*

The volumes of left Hb (HCs = 44.70 ± 23.06 mm3; OCD = 29.08 ± 22.53 mm3; F=6.09, *p* = 0.016) and right Hb (HCs = 48.21 ± 18.11 mm3; OCD = 34.99 ± 18.11 mm3; F=10.04, *p* = 0.002) were significantly smaller in OCD patients than in HCs (see Table S5). After controlling for the total intracranial volume (TIV) of each subject, the differences remained (F*left*=4.09, *pleft*=0.047; F*right*=8.37, *pright*=0.005).

**Table S5 Comparison of gray matter volume of habenula in the replication dataset**

|  |  |  |  |
| --- | --- | --- | --- |
| **GMV** | **OCD(n=38)** | **HC(n=34)** | **F** |
| **left Hb** | 29.08±22.53 | 44.70±23.06 | 6.09\* |
| **right Hb** | 34.99±21.16 | 48.21±18.11 | 10.04\*\* |

***Notes.***GMV, gray matter volume. \*Significance level of 0.05; \*\*Significance level of 0.01.

*Functional connectivity of the habenula*

ROI analysis showed that patients with OCD showed significantly increased FC of right Hb-right hippocampus ([26, -126, -18], voxels=53, T=4.80, *p*=0.001, SVC) (see Figure S3). No other significant difference in FC between the OCD group and HCs was detected.

**Figure S3** **Group difference in functional connectivity of the right Hb by ROI analysis**

Enhanced resting-state functional connectivity (rsFC) of the right habenula-right hippocampus were found in patients with obsessive-compulsive disorder (OCD) when compared to healthy controls. The connectivity values were obtained from small-volume correction analyses. The color bar represents the T value. ***Abbreviations****:* L, left;R, right.

*Effective connectivity of the habenula*

The fully connected model between right habenula and right hippocampus was established in the DCM analysis. The winning model at the group level were presented in Figure S4. The between-group comparisons revealed that patients with OCD had an increased effective connectivity from the right hippocampus to the right habenula and increased self-inhibition of the right habenula (see Figure S4-(3)).

图片包含 背景图案

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**Figure S****4 The DCM models**

**(1)** The fully connected model between the habenula and hippocampus was established in the DCM analysis. (2) The DCM model in healthy controls. Blue one-way arrows and values are inhibitory modulation. Blue bidirectional arrows and values are self-inhibition (3) The DCM model in patients with OCD. Blue one-way arrows and values are inhibitory modulation. Orange one-way arrows indicate decreased inhibition. The red dots represent the significant group difference between healthy controls and patients with OCD. The between-group comparisons revealed that patients with OCD had an increased effective connectivity from the right hippocampus to the right habenula and increased self-inhibition of the right habenula. These models were thresholded at 0.95 posterior probability after exhaustive Bayesian Model Reduction (BMR) and Bayesian Model Averaging (BMA). ***Abbreviations***:OCD, obsessive-compulsive disorder; HC, healthy controls; HPC, hippocampus; Hb, habenula.