Supplementary Online Content


eAppendix. eMethods

eReferences

eFigure. SPECT template with striatal region of interest

This supplementary material has been provided by the authors to give readers additional information about their work.
Exclusion criteria

Exclusion criteria were: (1) past or present history of neurological disorder (e.g. epilepsy, meningitis, structural brain damage); (2) abnormal ECG; (3) past or present history of substance abuse, psychosis, bipolar disorder or ADHD; (4) current major depression or use of antidepressant medication; (5) past or present use of medication known to affect D2/3 receptor binding (including all antipsychotics and methylphenidate); (6) history of use of any illicit drug other than cannabis; (7) past year radiation exposure for research purposes; (8) family history (first-degree) of psychotic disorder; (9) positive urine drug screen prior to SPECT; and (10) for females, a positive urine pregnancy test or breast feeding.

Additionally, individuals with SHI were excluded if they had attended a primary or secondary school in which they actively learned or used Dutch Sign Language, because being part of a signing deaf community could be protective against social exclusion. Control subjects were excluded if they had any form of impairment that could lead to social exclusion (e.g. wheelchair user).

Screening

Before the scanning day, subjects were interviewed using an adapted version of the Comprehensive Assessment of Symptoms and History (CASH) by a trained psychologist to screen for the presence of a personal and family history of psychiatric disorders. Handedness was established using the Edinburgh Handedness Inventory. A physician took the person’s medical history and performed a brief physical examination.

In control subjects, hearing was tested with an AC40 audiometer (Interacoustics, Assens, Denmark) using an automated Hughson-Westlake procedure. Subjects with SHI were tested by an audiometrician at the audiology department of the Academic Medical Center of the University of Amsterdam. Unaided pure-tone perception measurements were acquired to calculate Fletcher Indices (FI), as well as free field speech-in-noise measurements with hearing aids to quantify impairment in daily life situations. In addition, subjects with SHI were interviewed about the history of their hearing loss.

For SHI subjects, a high-resolution T2-weighted Magnetic Resonance Imaging (MRI) scan was acquired to exclude major brain lesions or abnormalities. For 4 SHI subjects with an implanted hearing device (1 Bone Anchored Hearing Aid (BAHA), 3 Cochlear Implants (CI)) no MRI scan could be acquired, but their pre-operative medical history contained no indication of brain lesions. While placement of a CI requires surgery under anesthesia, brain lesions as a result of surgery are unlikely since the implant is placed in the mastoid and inner ear, and only interfaces with the auditory nerve in the cochlea. Importantly, the pertinent subjects did not report post-operative neurological complications.

SPECT acquisition

Participants underwent two [123I]IBZM SPECT scans on the same day: before and after intravenous administration of dexamphetamine. Participants were asked to fast from midnight until the end of the second SPECT scan (no smoking, food, caffeine or alcohol). Participants were offered a small breakfast (1-4 slices of bread or equivalent) 3-4 hours before scanning. One day prior to the SPECT scans and on the day of imaging, participants were given potassium iodide to prevent free radioactive iodine from entering the thyroid. All participants received approximately 80 MBq [123I]IBZM (specific activity > 200 MBq/nmol; radiochemical purity > 95%) as an intravenous bolus, followed by continuous infusion of 20 MBq/hour for the duration of the experiment (300 minutes). To induce a state of sustained binding equilibrium after 120 minutes, the bolus to hourly infusion ratio was approximately 4.0. Scans were performed using a 12-detector single-slice brain-dedicated scanner (Neurofocus 810, Inc., Medfield, © 2013 American Medical Association. All rights reserved.
Massachusetts, USA) with a full-width at half maximum (FWHM) resolution of 6.5 mm, throughout the 20 cm field-of-view. After positioning of the subjects with the head parallel to the orbitomeatal line, axial slices parallel and upward from the orbitomeatal line to the vertex were acquired in 5-mm steps (300 sec scanning time per slice). The energy window was set at 135–190 keV. SPECT data were acquired for approximately 60 minutes per scan, starting from 120 minutes and 240 minutes after the initiation of $[^{123}I]IBZM$ administration. Approximately 15 minutes after the first SPECT scan, dexamphetamine sulphate (0.3 mg/kg body weight; Spruyt Hillen BV, IJsselstein, The Netherlands) was administered intravenously over 2 minutes.

**SPECT analysis**

Attenuation correction of all images was performed as earlier described. Images were reconstructed in 3D mode. A SPECT template was computed from all scans (76 scans from 38 subjects) with the DARTEL procedure in the SPM8 package running on Matlab for Windows (Mathworks, Natick, MA). Scans were normalized to the same space as the template by applying the individual flow fields generated in the DARTEL procedure. A region-of-interest (ROI) analysis was performed with a fixed ROI for the striatum (STR) generated from the SPECT template by selecting all voxels with signal intensities exceeding a threshold of half the peak value (eFigure). The occipital cortex (OCC) was selected as a reference region to estimate the concentration of free and non-specifically bound $[^{123}I]IBZM$ and drawn manually on the SPECT template using itk-SNAP software (version 2.1, PICSL, University of Pennsylvania). The non-displaceable binding potential (BPND) was calculated as the ratio of specific to non-specific activity (Formula 1).

**Formula 1:**

$$BP_{ND} = \frac{\text{Activity}_{STR} - \text{Activity}_{OCC}}{\text{Activity}_{OCC}}$$

**eReferences**


eFigure: SPECT template with striatal region of interest

Transversal (left), sagittal (top right) and coronal (bottom right) cross sections of the DARTEL generated template of all 76 SPECT scans. Superimposed in red: standard striatal region of interest generated by thresholding the template at 0.5 of the peak value.