Supplementary Materials

Methods

1. Animals

Adult (6-8 weeks) male C57BL/6J (Guangdong Medical Laboratory Animal Center, Guangdong, China) mice were used in the experiments. They were provided with food and water *ad libitum*, and housed under a 12-h light/dark cycle (lights on at 7:00 a.m.). All behavioral testing was conducted during the light phase of the cycle. The study was approved by the ethics committees at the Shenzhen Institutes of Advanced Technology, Chinese Academy of Sciences.

2. Visual stimuli

In the behavioral experiment, four types of looming stimuli were used, i.e., the normal looming (no-change, Disk), the topologically changing looming (TP-change, Ring), and two non-topologically changing looming (non-TP-change, Square and S). The Disk, Square and S stimuli had the same TP (no hole), thus the shape transformations between them had non-TP changes. The Ring stimuli (one hole) were topologically different from the other three, hence a TP-change looming. The S-like figure was specifically designed to control the figure area and other potential confounding factors, e.g., spatial frequency and perimeter length. Therefore, the main difference between the S (and the Square) and the TP-change (Ring) stimulus was the hole in the ring, i.e., the topological property (TP).

In the first c-fos experiment (see Fig. 1D-E), visual stimulus-evoked c-fos activation in the SC was analyzed. Mice were given no-change and TP-change looming stimuli as described in the behavioral experiment.

During the second c-fos test (see Fig. 2), mice were exposed to non-looming shape-changing stimuli. These stimuli did not change in size over time and included both TP and non-TP changes. During the TP-change stimulus, the overhead shape transformed back and forth between a disk and a ring. During the non-TP change stimulus, the overhead shape transformed back and forth between a disk and a square. The area of the shape was kept invariant during both the TP and non-TP changing transformations. The visual stimuli were programmed in Matlab (Mathworks, Natick, USA) using Psychtoolbox.

3. Looming behavior test

Mice were placed in a closed plexiglass box (40 cm long, 40 cm wide, and 30 cm high) with a dark sheltered nest in one of the corners and an LCD monitor in the ceiling. The LCD was used to present the visual stimuli (see Figure 1A). Mouse behaviors and responses were recorded using an infrared camera. The mice were allowed to be familiarized with the box for 15 min the day before the test. In addition, each mouse was given 5 min to freely explore the box on the day of the test. The stimulus was triggered by the experimenter when the mouse entered an area located on the opposite side of the box with respect to the corner that housed the sheltered nest. The following parameters were measured and used to analyze mice's looming-evoked defensive behavior: (i) flight latency (ms), i.e., the time passed from onset of looming stimulus to

onset of escape to the nest, (ii) time to the nest (ms), i.e., the time taken to escape to the nest, and (iii) time in the nest, i.e., the time that a mouse spent in the nest after being exposed to a stimulus. Data analysis was performed using Adobe Premiere. During the behavioral experiment, each mouse was presented with four different stimuli. Each stimulus was present once in a randomized order. Each mouse had a unique ear tag with a numerical identifier.

4. Histology

The mice were transcardially perfused with 4% paraformaldehyde in PBS 1.5 h after the behavioral experiments ended. During perfusion, the mice were under anesthesia with sodium pentobarbital (80 mg/kg BW). Their fixed brains were cryoprotected using 30% sucrose in PBS and cut into 30 μm slices using a cryostat. Immunohistochemistry was performed to map c-fos activation in the brain. Antibody staining was performed on single-well floating tissue slices. The slices were incubated for 48 h in primary antibodies at 4°C followed by overnight incubation with secondary antibodies at 4°C. Primary antibody rabbit anti-c-fos (2250, Cell Signaling Technology; 1:500) and secondary antibody Alexa fluor 488 goat anti-rabbit (Jackson, 1:200) were used. For counterstaining, slices were incubated for 10 min with 4′, 6-diamidin-2-phenylindol (DAPI, 0.4 μg/mL, Sigma). All images were captured using either a Zeiss LSM 880 confocal microscope or an Olympus VS120 virtual microscopy-slide scanning system. C-fos staining within the SC was manually counted by an individual experimenter blind to the experimental groups.