Supporting Information:

Social interaction test

In brief, mice were located into a novel arena (80 cm × 31.5 cm) formed of three communicating chambers with central openings permitting free access to all chambers for 5 min. Distance (cm) and time (s) in the differents area was estimated during this time to evaluate locomotor activity and ensure that animals did not have a preference for a particular side of the arena. Following acclimatization, mice were restricted to the central chamber while an unfamiliar mouse located in a small wire cage was placed in one of the external chambers. An identical empty wire cage was placed in other chamber. The unfamiliar mouse was arbitrarily assigned to either the left or right chamber of the arena. The test mouse was allowed to scan the arena/chambers for a further 10 min. Time spent engaging in investigatory behavior with the novel mouse and frequency of investigatory behavior with the novel mouse was evaluated with EthoVision XTsoftware (Noldus Netherlands) in order to observe social approach and preference. All testing happened during the dark phase (21:00–03:00 h) under illumination with red light.

Sociability Index (SI) was defined as the ratio between duration of test mouse in the novel side and duration of test mouse in the familiar side.

Elevated Plus Maze (EPM)

Anxiety is reflected by the avoidance of the open arms of the maze. The EPM is made of dark grey plastic with two opposing closed arms (30 x 5 x15 cm, 10 lux) and two opposing open (30 x 5 cm, 300 lux) linked by a platform (5 x 5 cm, 90 lux), situated 50 cm above the floor and bordered by a black awning. Each trial continued for 5 min and the apparatus was washed before each session.

Behavior was videotaped and the rate of time spent on the open arms was scored by a researcher blind to the animal group. The animals that showed all four paws in the open space were used to calculate this parameter

Western blot analysis for Bax, Bcl-2, inducible nitric oxide synthase (iNOS), IκBα, nuclear factor-©B (NF-kB) and glial fibrillary acidic protein (GFAP)

In brief, brain tissues from each animal were raised in extraction buffer A (0.2 mM phenylmethylsulfonyl floride, 0.15 µM pepstatin A, 1 µM sodium orthovanadate, 20 µM leupeptin), homogenized at the highest setting for 2 min and centrifuged at 1000 *g* for 10 min at 4°C. Supernatants represented the cytosolic fraction. The pellets were resuspended in buffer B (1% Triton X-100, 10 mM Tris-HCl, 150 mM NaCl pH 7.4, 1 mM EGTA, 1 mM EDTA, 0.2 mM

phenylmethylsulfonyl floride, 20 μM leupeptin, and 0.2 mM sodium orthovanadate). After centrifugation at 15,000 *g* for 30 min at 4°C, the supernatants were saved at –80°C. Equivalent amounts of protein (50 μg) for each sample were loaded per lane and electrophoretically separated using 10% denaturing PAGE (SDS-PAGE). The levels of Bax, Bcl-2, iNOS, IκBα, GFAP were quantified in cytosolic fractions. NF-κB p65 was quantified in nuclear fractions. The filters were blocked with 5% (w/v) non-fat dried milk in phosphate-buffered saline (PBS), for 40 min at room temperature (RT), and probed with one of the following primary antibodies: anti-Bax, anti-Bcl-2, anti-iNOS, anti-NF-κB p65, anti-IκBα antibody, anti-GFAP antibody in 5% non-fat dried milk/0.1% Tween 20/PBS at 4°C overnight. All antibodies were purchased from Santa Cruz Biotechnology and diluted 1:500. Membranes were incubated with peroxidase-conjugated goat anti-rabbit IgG or peroxidase-conjugated bovine anti-mouse IgG secondary antibody (1:2000, Jackson ImmunoResearch Laboratories, West Grove, PA, USA) for 1 h at RT. To establish that blots were loaded with equal amounts of protein lysates, they were furthermore probed with a β-actin antibody (1:2000, Sigma-Aldrich, Milan, Italy). Membranes were stripped by agitation with 3% glycine, pH 2 , blocked in 5% non-fat dried milk/PBS for 1 h at RT and probed for 2 h at RT with anti-β-actin antibody. Membranes were stripped and blocked as above and probed for 2 h at RT with anti-β-actin antibody. The proteins expression were quantified by densitometry with Gel Logic 2200 PRO software and standardized to β-actin levels. Images of blot signals (8 bit/600 dpi resolution) were imported to analysis software (Image Quant TL, v2003). A preparation of commercially available molecular weight markers (10 to 250 kDa) was used to calculate molecular weight positions and as reference concentrations for each molecular weight.

Immunohistochemical localization of chymase, tryptase, tumor necrosis factor alpha (TNF-α) and interleukin-1beta (IL-1β)

Brain tissues were fixed in 10% (w/v) PBS-buffered formaldehyde and 8 μm sections prepared from paraffin-embedded tissues. After deparaffinization, endogenous peroxidase was raised with 0.3% (v/v) hydrogen peroxide in 60% (v/v) methanol for 30 min. Sections are permeabilized with 0.1% (w/v) Triton X-100 in PBS for 20 min. Non-specific adsorption was maimed by incubating all sections in 2% (v/v) normal goat serum in PBS for 20 min. Endogenous biotin or avidin binding sites were stopped by sequential incubation for 15 min with avidin and biotin (Vector Laboratories). Sections were washed with PBS and incubated with secondary antibody. Specific labeling was individualized with a biotin-conjugated goat anti-rabbit IgG and avidin-biotin peroxidase complex (Vectastatin, ABC elite kit, Vector Laboratories). To check the binding specificity for chymase, tryptase, TNF-α and anti-IL-1β parallel sections was incubated with only primary or secondary

antibody; no positive staining was found. Images captured (n=5 photos from each sample gathered from all animals in each experimental group) were quantitatively evaluated for gaps in immunoreactivity by computer-assisted color image analysis (Leica QWin V3, Cambridge, UK). All analyses were carried out without knowledge of the treatments.

Golgi staining

Blocks of brain tissue were puted into solutions A and B, for 2 weeks in the dark at RT. The blocks were transferred to solution C (4°C), frozen on dry ice and saved at -70°C until sectioning. Cryostat sections (100 µm) were cut at -25°C and mounted onto gelatinized slides. Only sections stained dark with Golgi impregnation throughout were selected for analysis. Neurons chosen for tracing follow the subsequent criteria: (1) completely impregnated with Golgi stain; (2) unobscured by other impregnated neurons or precipitant; (3) 70% of the dendritic tree was evident within the plane of focus; and (4) dentate granule neurons must have been situated in the external one-half of the granule cell layer in the dentate gyrus. Spines were numbered under oil (X100), using light microscopy (Axostar Plus equipped with Axio-Cam MRc, Zeiss), and the total visible dendritic length calculated with an imaging computer program (Axio-Vision, Zeiss). Spine density was measured referring to the length of the dendrite.

Bromodeoxyuridine (BrdU) and Doublecortin (DCX) immunohistochemistry

To label newly cells we used incorporation of the thymidine analogue BrdU into DNA in the S-phase of the cell cycle.

BrdU was dispensed intraperitoneally at 150 mg/kg in saline. Mice were sacrificed 2 h after injection; anesthetized with sodium pentobarbital (30 mg/kg) and transcardially perfused with cold saline, followed by 4% cold paraformaldehyde in PBS. All brains were post-bounded overnight in 4% paraformaldehyde at 4°C, cryoprotected in 30% sucrose and stored at 4°C. Serial sections were clipping through the entire hippocampus using a cryostat and saved in PBS.

Immunohistochemistry was executed in the following steps: incubation for 2h in 1:1 formamide/2X SSC at 65°C, rinse in 2X SSC for 5 min, incubation in 2N HCl at 37°C for 30 min, and rinse in 0.1 M boric acidcfor 10 min, pH 8.5, 2 h incubation in 0.1 M PBS with 0.3% Triton X-100, and 5% normal donkey serum. Sections were incubated overnight at 4°C with primary antibodies for DCX (goat; 1:500; Santa Cruz Biotechnology), BrdU (mouse; 1:500; Santa Cruz Biotechnology). After secondary antibody incubation, sections were developed using Vector ABC kit and diaminobenzidine. All sections were looked by light microscopy (Axostar Plus equipped with Axio-Cam MRc; Zeiss) and scanned via an imaging computer program (Axio-Vision; Zeiss,

Hallbergmoos, Germany). Two major observations in stereological analyses are that no tagged cells be numbered twice and that the area numbered be consistent in each section. Every sixth section throughout the entire hippocampus was treated for BrdU and DCX immunohistochemistry. Using this spacing assures that the same neuron will not be numbered in two sections. All labeled cells in the dentate gyrus (granule cell layer) and hilus were numbered in each section by an experimenter obscured to the research code. To discriminate single cells within clusters, all counts were carried out at 400X and 1000X under a light microscope, missing cells in the outermost focal plane. A cell was numbered as being in the subgranular zone (SGZ) of the dentate gyrus if it was movingor in the SGZ. Cells that were situated more than two cells away from the SGZ were booked as hilar. The total number of marked cells per section was calculated and multiplied by six to obtain the total number of positive cells per dentate gyrus. ANOVA and post hoc Tukey tests were performed on these totals with Bonferroni correction for multiple comparisons.