Supplemental materials

Methods and materials

1. Strain and biological origin

Candidate *Bifidobacterium* strains were provided by the Culture Collection of Food Microorganisms at Jiangnan University (Wuxi, China). This study involves no intrusive human intervention procedures. Fecal samples are collected with written informed consent for research purpose.

Table S1 Strains and biological origin

Number	Species	Biological Origin	Donor age	Donor gender
CCFM687	*B. longum* subsp. *infantis*	Feces of infant	< 24 hours	Unknow
CCFM753	*B. longum* subsp. *infantis*	Feces of infant	< 1 year	Male
CCFM681	*B. longum* subsp. *longum*	Feces of infant	< 24 hours	Unknow
CCFM688	*B. longum* subsp. *longum*	Feces of infant	< 24 hours	Unknow
CCFM758	*B. longum* subsp. *longum*	Feces of infant	< 1 month	Female
CCFM760	*B. longum* subsp. *longum*	Feces of infant	< 2 years	Female
CCFM686	*B. longum* subsp. *longum*	Feces of infant	< 24 hours	Unknow
FSDJN6M3	*B. longum*	Feces of infant	< 24 hours	Male
FJSWXJ38M1	*B. longum*	Feces of long-lived people	91 years	Female
HAN4-25	*B. longum*	Feces of long-lived people	96 years	Female
HUB2-25	*B. longum*	Feces of infant	< 6 months	Male
GX17-A9	*B. longum*	Feces of long-lived people	90 years	Male
FSHHK25M1	*B. longum*	Feces of long-lived people	86 years	Female
FSHHK27M1	*B. longum*	Feces of long-lived people	86 years	Female

2. Protocols of chronic unpredictable mild stress (CUMS)

The CUMS procedure is a classical modeling method that widely used for inducing anxiety and depression, and validated by many independent research group with lots of published articles. The stressors used in this study has no fatal or disabling risk. More importantly, each stressor is used no more than three times. Although the stressors cause temporary discomfort to animals, they recover quickly when the stress stopped.

a) Forced swimming
   1. Fill a glass tank 22 cm deep with water at 23 ± 2 °C
   2. Place the animal in the glass tank for 10 min.
   3. Return the animal to a clean and dry cage with fresh bedding in order to avoid chills and colds.

b) Restraint

1. Place the animal in a 50 mL plastic tube; adjust it with plastic tape on the outside so the animal is unable to move. The tube must have a hole at the far end to allow regular breathing. To place the animal in the plastic tube, it is necessary to place the head of the animal close to the entrance, after which it should enter by itself.
2. Wait for 1 h. Although the plastic tube should be sufficient to prevent the animal coming out is desirable to leave the immobilizer inside the cage.
3. Return the animal to its cage. The best way to extract the animal is to make a sudden movement downwards, dropping it into the cage. Try to avoid pulling from the tail.

c) Water deprivation
   1. Remove the bottle of water from the cage during 24 h. If the animal house is supervised by staff, indicate that the water/food must not be replaced in that cage.
   2. Place the bottle of water back after the time point is reached.

d) Isolation
   1. Place the animal alone in a new cage.
   2. Return the animal to the cage with their mates.

e) Food deprivation
   1. Remove the food from the cage for 24 h.
   2. Place the food back.

f) Wet bedding
   1. Place the animal in a new cage with 200ml water per 100g bedding for 24 h.
   2. Return the animal to the cage with their mates.

g) No bedding
   1. Place the animal in a new cage without bedding for 24 h.
   2. Return the animal to the cage with their mates.

h) Tilt cages
   1. Tilt the cages at 45° for 24 h.
   2. Return the cage to the normal state.

i) Clip tail
   1. The plastic clamp is clipped at 2 cm from the end of tail, sustain 1 min each time, total 3 times a day.
   2. Return the animal to the cage with their mates.

j) Continuous illumination
   1. Transfer the cages into another house in a continuous illumination with white light. The illumination intensity is 500 lux.
   2. After exposed for 24h, return the cage to the normal state.

3. Protocols of behavioral tests

2.1 Forced Swim Test (FST)

Using a glass cylinder with 30 cm with 23-25℃ water. In the first day, all mice are forced to swim for 15 minutes for training. The next day, place mice in the same environment for 10 minutes. Locomotor activity is monitored using a video tracking system (EthoVision pro, Noldus Inc., Leesburg, VA). EthoVision software is used to calculate the immobility time of each animal during

the test period. Immobility is defined as the motionless situation except floating motions required to keep the head above the water.

2.2 Tail Suspension Test (TST)

Tape the tip of mice tail with adhesive tape (2 cm from tail tip) to a suspension bar (30 cm high from the floor). Locomotor activity is monitored by the video tracking system for 6 min. The duration of immobility was scored. Immobility is defined as the absence of voluntary or escape-orientated movement.

2.3 Elevated Plus Maze (EPM)

Mice were placed in the center area of the maze facing the open arms, animals were allowed to move freely for 6 min and their behavior was recorded by a video tracking system. The entries into light and dark arms, and the time spent in light arms are scored.

2.4 Dark/Light Box Test

The apparatus consists of a box (50×21×25cm) divided into a dark and an illuminated compartment (half for each). Mice were placed in the dark chamber and facing the door to the light initially. Recorded the time spent in the light chamber and entries into light chamber.

2.5 Step Down Test (SDT)

Electric shock training phase: The mouse was placed on the platform and the electric grid was energized (0.2 mA). When the mouse jumped off the platform, it was subjected to a single electric shock stimulus, resulting in nociceptive memory. The training time was three minutes. If the mice did not jump off the platform within 3 minutes, they were driven off by humans. All mice returned to the original cage after the training. 24h after training phase is the memory reproduction phase: Put the mouse on the platform for a test of 3 minutes. This test will not be energized. Recorded the latency of jumping off the platform.

4. Microbial analysis

Microbiota-related analyses were conducted by QIIME and R (version 3.5.0) software. Alpha Diversity is a quantitative measure that reflects how many different types (such as species) there are in a dataset, and simultaneously takes into account how evenly the basic entities (such as individuals) are distributed among those types. Shannon index is a simple mathematical measure for assessing species richness, which accounts for both abundance and evenness of the species present. The PD whole tree index is calculated based on the branch lengths of the phylogenetic tree as a measure of phylogenetic diversity. Linear Discriminant Analysis Effect Size (LEfSe) and Phylogenetic Investigation of Communities by Reconstruction of Unobserved States (PICRUSt) were performed online (http://huttenhower.sph.harvard.edu/galaxy). LEfSe is a method for discovering high-dimensional biological markers and determines the features (organisms, clades, operational taxonomic units, genes, or functions) most likely to explain differences between classes

by coupling standard tests for statistical significance with additional tests encoding biological consistency and effect relevance. PICRUSt is a bioinformatics software package designed to predict metagenome functional content from marker gene (e.g., 16S rRNA) surveys and full genomes.

Supplementary results

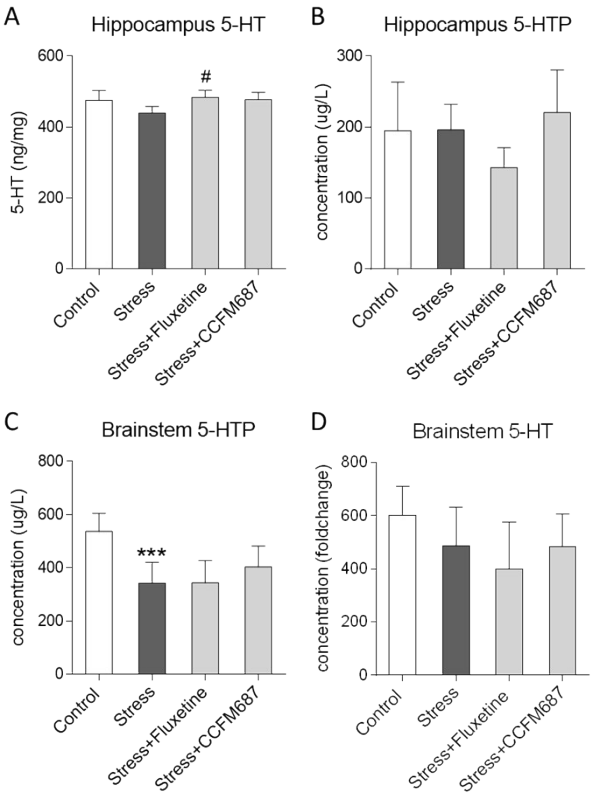

Figure. S1 5-HTP and 5-HT distribution in hippocampus and brainstem.

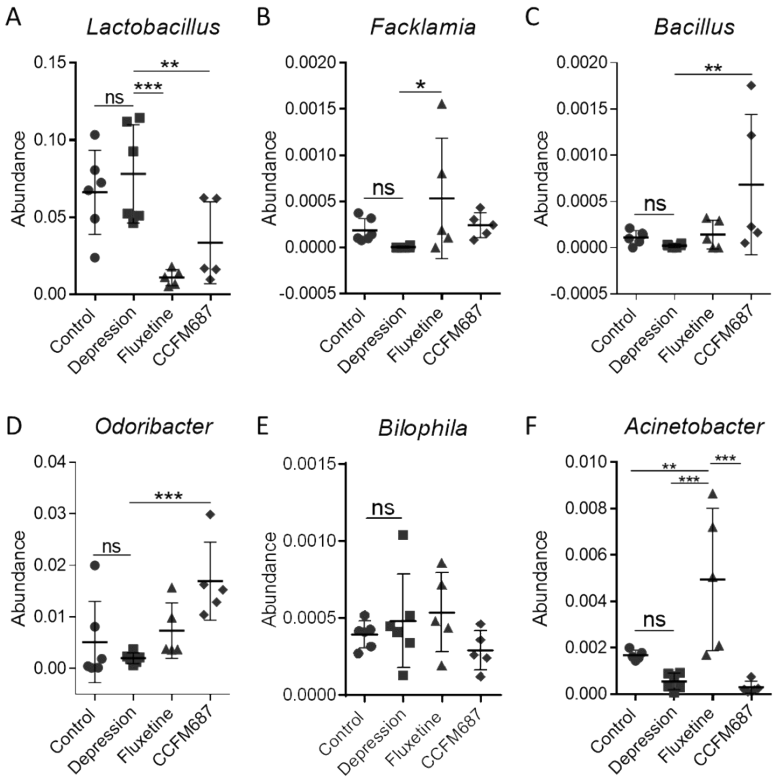

Figure. S2 Microbial distribution at genus level.