

A Cell Co-Culture Taste Sensor Using Different Proportions of Caco-2 and SH-SY5Y Cells for Bitterness Detection

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Methods:

1. Image J cell counting

To qualify the ratio of two types of cells, cell counting was performed using ImageJ software for each single-color fluorescence image. The operation steps are as follows, in short:

- 1) Open the image to be counted, and convert it to greyscale before proceeding (Image -> Type->8 bit);
- 2) Inverted image (Edit->Invert);
- 3) To highlight all of the structures you want to count(Image->Adjust-> Threshold); Process->Subtract background with the rolling ball may help if highlighting too many "noise" or background pixels;
- 4) Fill in the gaps in the nucleus (Process->Binary->Fill Holes);
- 5) If particles merged together, cut them apart by adding a 1-pixel thick line (Process->Binary->Watershed);
- 6) Automatic cell counting(Analyze->Analyze Particles).

2. Quantitative reverse transcription-PCR analysis

Total RNA was extracted from Caco-2 cells and SH-SY5Y cells using TaKaRa MiniBEST Universal RNA Extraction Kit (9767), and reverse transcribed with 1 µg RNA using PrimeScript™ 1st Strand cDNA Synthesis Kit (TaKaRa, 6110A) according to the manufacturer's instructions. Real-time PCR was set up using the iQ™ SYBR Green Supermix (Bio-rad, 170-8884), and carried out with the CFX Connect™ Real-Time System. Specific primers were obtained from Sangon Biotech. An endogenous "housekeeping" control gene β -actin was used in the quantification of relative expression levels of targeted genes.

Figure S1:

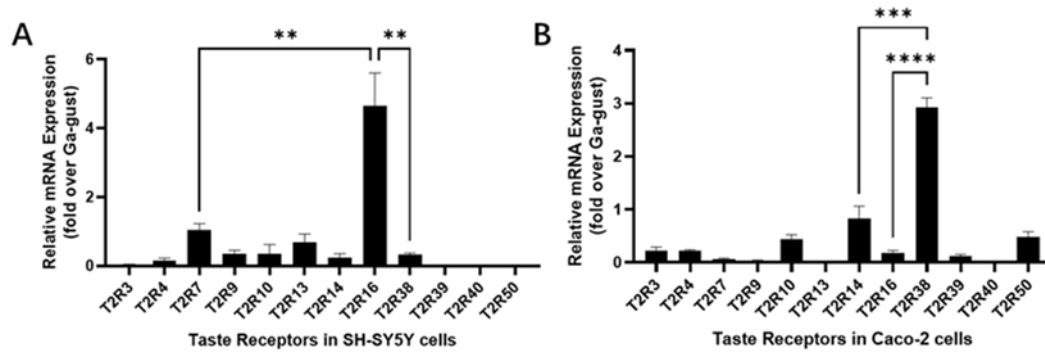


Figure S1 Taste receptors relative mRNA expression in (A) SH-SY5Y cells and (B) Caco-2 cells (n = 3). One-way ANOVA with Tukey's posthoc test, **** $p \leq 0.0001$, *** $p < 0.001$; ** $p < 0.01$.