Supplementary Material

SERT and uncertainty: serotonin transporter expression influences information processing biases for ambiguous aversive cues in mice

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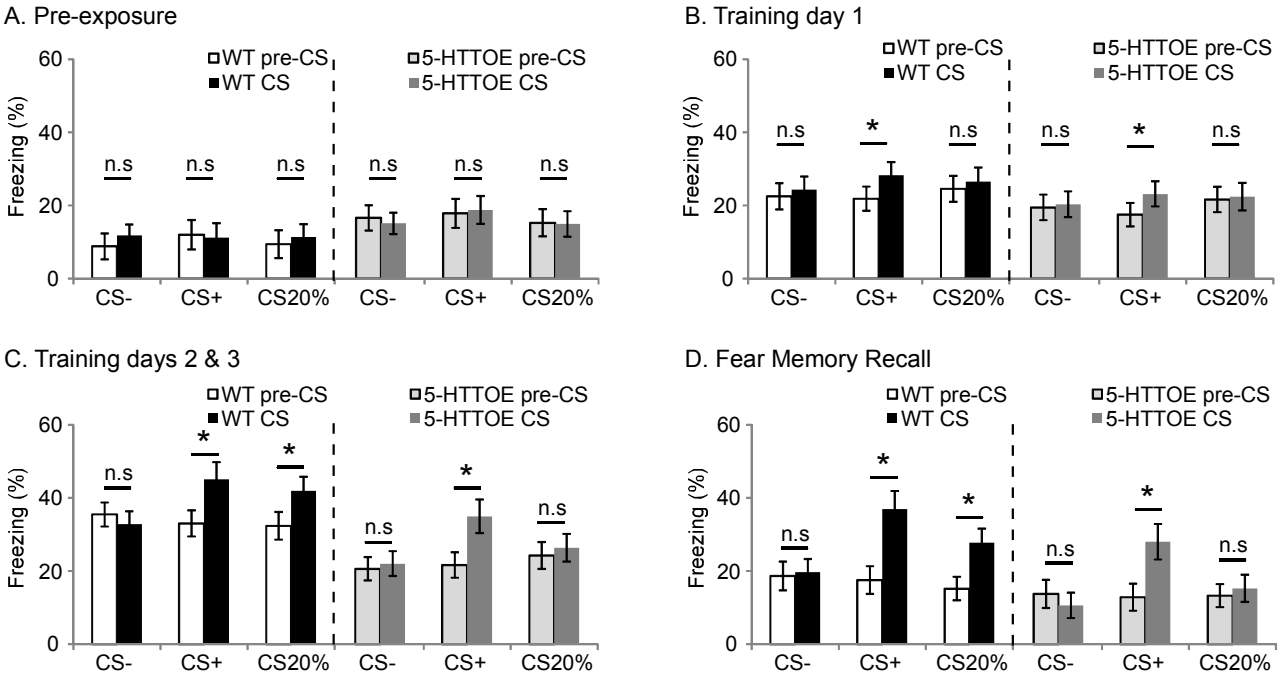
Contents:



Supplementary Figure S1. Raw freezing responses in WT and 5-HTTOE mice showing pre-CS freezing (white bars for WT, light gray for 5-HTTOE) and during-CS freezing (black bars for WT, dark gray for 5-HTTOE) for the three cue types (CS-, CS+, CS20%). A. Responses during pre-exposure. B. Responses during training day 1. C. Responses averaged over training days 2 and 3. D. Responses during the first two trials of the fear memory recall session. *$p < 0.05$; n.s: non-significant.

Supplementary Results

Figure S1 shows raw pre-cue (pre-CS) and cue-evoked (CS) freezing responses for all days of the experiment. We first analysed these freezing responses for the pre-exposure day (ANOVA model: genotype$_2$ × phase $_{(\text{pre-CS,during CS})\ 2}$ × CS type$_3$ × trial$_5$ × S$_{51}$). There was a main effect of trial (F(4,196) = 10.6, P < 0.001), reflecting the increase in immobility over the course of the session, but there were no other main effects or interactions (see Figure S1A).

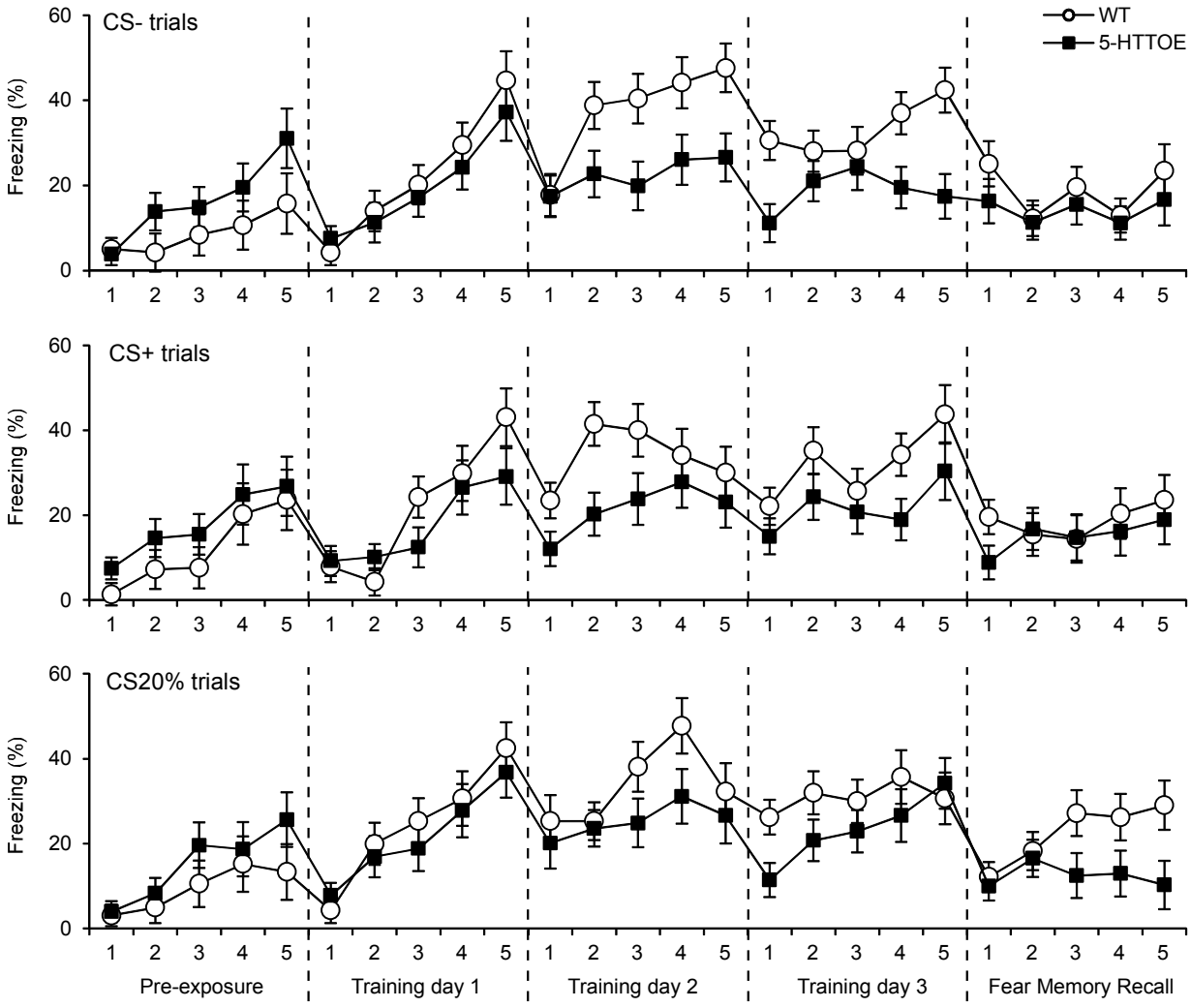
Performing the same analysis on training day 1 revealed main effects of phase (F(1,49) = 10.6, P =0.002) and trial (F(4,196) = 48.2, P < 0.001), and an interaction between phase and CS type (F(2,98) = 3.8, P = 0.025). Simple main effects analysis of the effect of phase within each CS type revealed higher freezing during CS+ trials compared to the pre-CS+ period (F(1,49) = 12.1; P = 0.001), and this effect was equivalent in both genotypes, whereas there was no effect of phase for the CS- or CS20% trials (F(1,49) < 1.2; P > 0.25). There were no other main effects or interactions. These data are summarized in Figure S1B.

An analysis spanning training days 2 and 3 (ANOVA model: genotype$_2$ × phase $_{(\text{preCS,during CS})\ 2}$ × day$_2$ ×CS type$_3$ × trial$_5$ × S$_{51}$) revealed a three-way interaction between genotype, phase, and CS type (F(2,98) = 3.1, P = 0.048). Simple main effects analysis in the 5-HTTOE mice revealed a main effect of phase in CS+ trials (F(1,49) = 16.9, P < 0.001) but not in CS- or CS20% trials (F (1,49) < 1.8, P = 0.2). In contrast, in WT mice there was a main effect of phase in both the CS+ (F(1,49) = 13.4, P = 0.001) and CS20% trials (F(1,49) = 35.5, P < 0.001) but not in the CS- trials (F(1,49) = 1.4, P = 0.2). These data are summarized in Figure S1C.

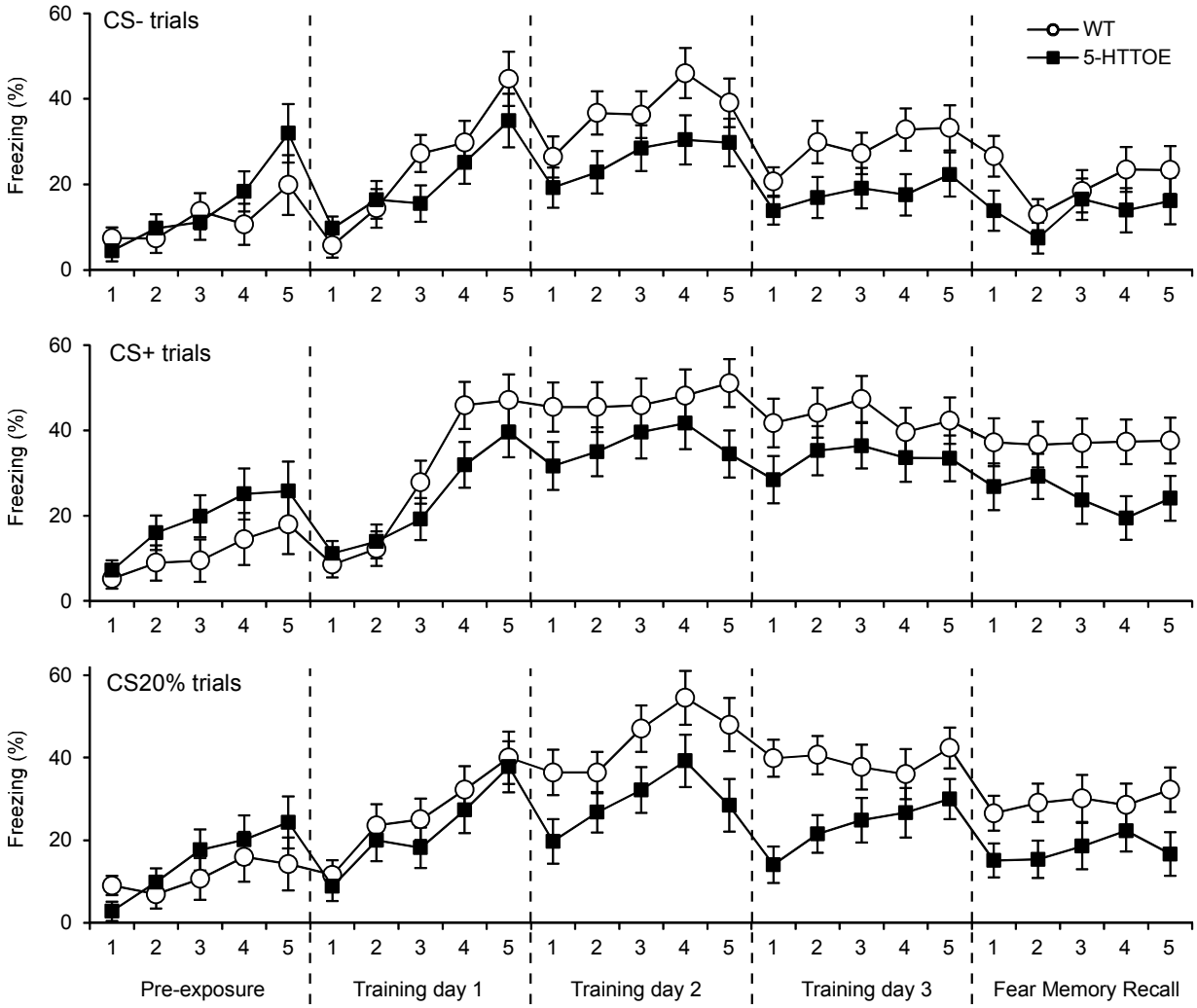
Finally, we analysed pre-CS and CS freezing levels for the first two trials of each type during the fear memory recall day (ANOVA model: genotype$_2$ × phase $_{(\text{preCS,during CS})\ 2}$ × CS type$_3$ ×

$trial_2 \times S_{51}$). This analysis revealed main effects of phase ($F(1,49) = 44.2$, $P < 0.001$) and CS type ($F(2,98) = 6.1$, $P = 0.003$), as well as interactions between CS type and phase ($F(2,98) = 11.6$, $P < 0.001$) and genotype and phase ($F(1,49) = 7.2$, $P = 0.01$). Although the three-way interaction between genotype, CS type, and phase was not significant ($F(2,98) = 0.5$, $P = 0.6$), simple main effects analysis were informative about how 5-HTTOE and WT mice changed their freezing responses in the presence of the cues. 5-HTTOE mice increased their freezing to the CS+ cue (effect of phase for CS+: $F(1,49) = 11.0$, $P = 0.002$) but not the CS20% cue (effect of phase for CS20%: $F(1,49) = 0.6$, $P = 0.5$). In contrast, WT mice increased their freezing to both the CS+ cue (effect of phase for CS+: $F(1,49) = 17.3$, $P < 0.001$) and the CS20% cue (effect of phase for CS20%: $F(1,49) = 22.4$, $P < 0.001$). These data are summarized in Figure S1D.

Raw freezing responses from the pre-CS and during-CS periods for all trials are shown in Figure S2 and S3, respectively. Note that raw freezing responses were lower in 5-HTTOE mice versus WTs during both pre-CS and CS periods during training days 2 and 3. The key point is that the change in freezing from pre-CS to during-CS was equivalent in WT and 5-HTTOE mice for CS- and CS+ trials but was significantly greater in WT than 5-HTTOE mice for CS20% trials, as shown in Figure S1C.



Supplementary Figure S2. Raw pre-CS freezing responses in WT and 5-HTTOE mice across all trials of the experiment for CS- (upper panel), CS+ (middle panel), and CS20% (lower panel).



Supplementary Figure S3. Raw cue-evoked freezing responses in WT and 5-HTTOE mice across all trials of the experiment for CS- (upper panel), CS+ (middle panel), and CS20% (lower panel).