Supplementary Information

1. **Schematic representation of the experimental design**
2. **Long Trace protocol decreased freezing to tone-cue**
3. **Lack of fear generalization**

1. Schematic presentation of the experimental design. Please note that the length of the elements does not correlate with time length.

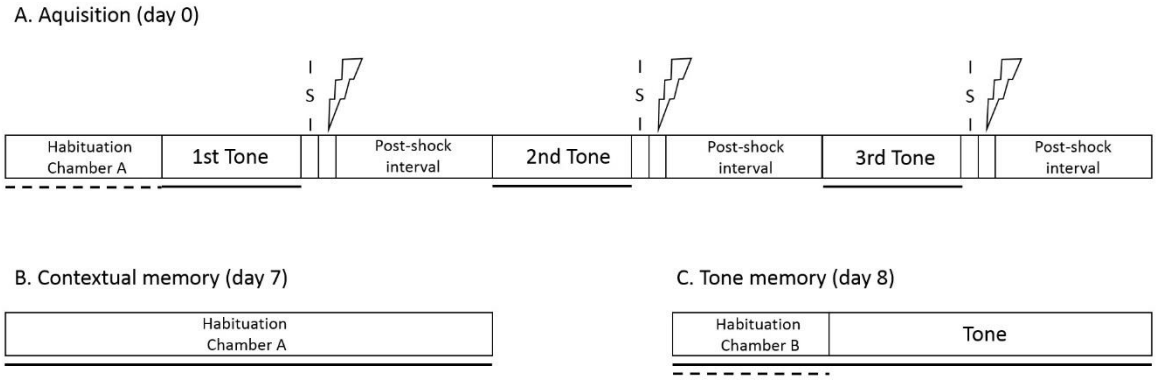

Figure S1. (A) Acquisition of pairing in Chamber A. Habituation and tone lasted 300-s and 15-s, respectively, in every experiment. Duration of ISI, and duration or intensity of the foot shock (flash) varied in the different experiments according to the main text. Analysis of (B) contextual memory in chamber A without the tone at day 7 and (C) tone memory in chamber B at day 8. The underlying lines indicate the elements of analysis for fear acquisition and memory retrieval (solid lines) and fear generalization (in Fig S2, dashed lines).

2. Merging the data of the 1 mA/30-s ISI groups from experiments III and IV revealed that cued memory was slightly, but significantly, reduced. However, these subtle effects suggest that a longer ISI uncouples the tone-cue from the US, leaving the contextual stimuli as the primary correlate to the US, as previously shown (see main text).

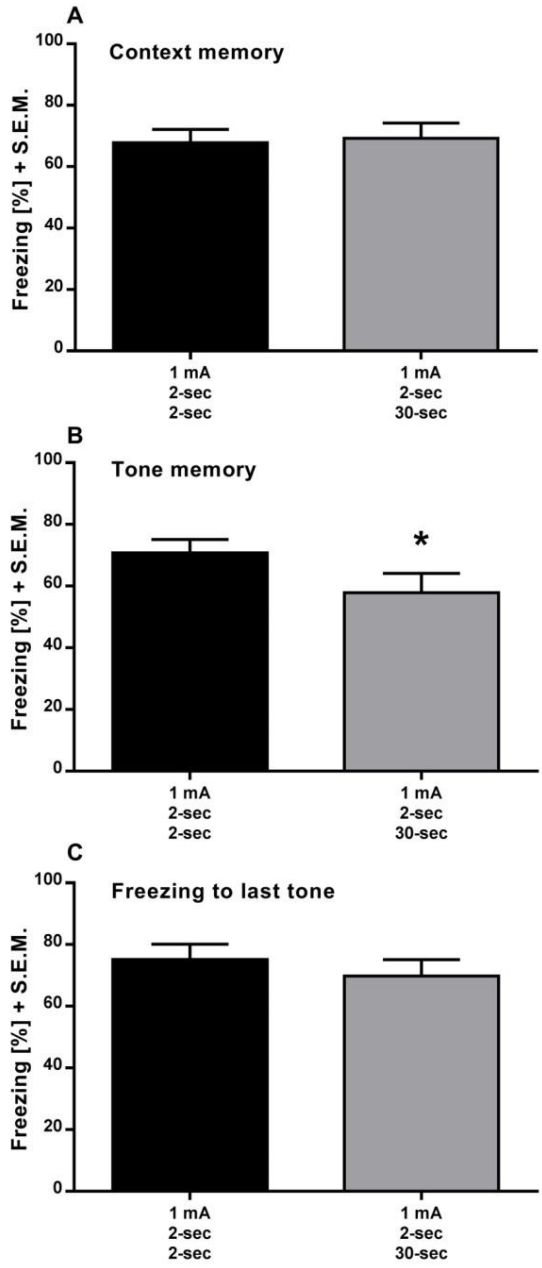

Figure S2: Long Trace protocol decreased freezing to tone-cue but left contextual memory unaffected when animal number per group is increased by merging control (black bar) and 30-s ISI groups of experiments III and IV. (A) The 30-s ISI group froze comparably to the learned context seven days after acquisition but (B) significantly less to the learned tone eight days after acquisition (unpaired one-tailed t-test; P=0.0495). (C) Freezing to the last tone during acquisition.

3. Fear generalization was not detected since the freezing level during the habituation phase in the novel context at day 8 is statistically comparable to the freezing level during habituation phase at day 0 (prior to acquisition). The time window of each analysis is two minutes.

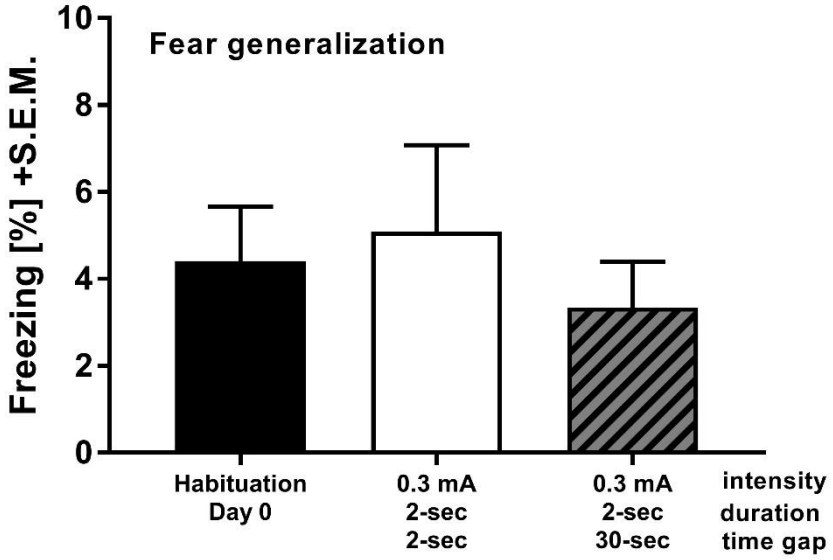

Figure S3: Acquisition with foot shock of an intensity of 0.3 mA did not lead to fear generalization in a novel context. We compared the freezing level of rats during the habituation phase at day 0 before acquisition (black bar) with the freezing level of the same rats that have received USs of 0.3 mA in a novel context shortly before presenting the tone-cue at day 8 (white or striped bar). One-Way ANOVA revealed no differences between the groups ($F_{(2,29)}$=0.276, P=0.7608).