Supplemental Methods

*Inhibition of Erk activity by U0126.* To establish the dependence of fear extinction on Erk signaling, mice were injected into the dorsal hippocampus with 1 µg/0.25 µl/site of U0126, an inhibitor of Erk phosphorylation by the mitogen activated and extracellular signal-regulated kinase. The drug was injected i.h. immediately after every extinction test.

*Effect of stronger training on fear conditioning and extinction.* To increase the strength of the conditioning memory, we employed: a. a group exposed to three 2-s shocks of 0.7 mA at 1-min intervals during a 3-min contextual exposure; or b. a group exposed to a single 2-s, 1.5 mA footshock at the end of a 3 min contextual exposure.
Legends for Supplemental Figures

Figure S1. Behavioral and molecular responses of cFos-LacZ mice to conditioning and extinction. A, cFos-LacZ mice did not differ from their wild type littermates or C57BL/6 mice in their ability to acquire and extinguish contextual fear. B, Injections of FDG did not affect acquisition and extinction of contextual fear in cFos-LacZ mice. C, I.h. injections of U0126 (0.5 µg/0.25 nl/site) immediately after individual extinction tests prevented extinction in cFos-LacZ mice (*p < 0.05, **p < 0.01 vs vehicle). D, The number of cFos and pErk cells in response to conditioning and extinction, respectively, did not differ among cFos-LacZ, wild type and C57BL/6 mice. The number of FDG+ cells was lower but not statistically significant.

Figure S2. Differences in FDG and pErk co-localization between the hippocampal CA1 area and parietal cortex. A, Hippocampus. Left column: pErk (top), FDG (middle) and composite (bottom) micrographs of CA1. Right: upper and lower rows: lack of detectable co-localization of FDG and pErk signals. Clear nuclear FDG signals and nuclear FDG signals overlayed by pErk+ passing fibers are marked. B, Parietal cortex. Left column: pErk (top), FDG (middle) and composite (bottom) micrographs of the parietal cortex adjacent to the hippocampus shown above. Right: upper and lower rows: clear co-localization of FDG and pErk signals. Labels: dashed squares, areas in the low magnification micrographs (left) selected to capture the high magnification micrographs (right); green stars, FDG+ cells; yellow stars, pErk+/FDG+ cells; yellow arrow, overlays of pErk+ fibres and FGD puncta.
Figure S3. Levels of cFos and pErk after stronger training conditions.  

A, Mice trained with one or three training trials (3 footshocks spaced between 1 min contextual exposures) exhibited similar levels of freezing after training and during extinction ($F_{4,70} = 0.105, p = 0.98$).  

B, Mice trained with a stronger shock (one 1.5 mA footshock delivered after a 3-min contextual exposure) exhibited similar levels of freezing after training but impaired extinction ($F_{4,70} = 3.91, p < 0.05$) when compared to the 0.7 mA group, indicating stronger fear memory.  

C, Mice trained with 1.5 mA footshock had similar cFos levels in the hippocampus after training ($t_{8} = 0.27, p = 0.73$) and reduced pErk levels on E5 ($t_{8} = 4.11, p < 0.05$) when compared to the 0.7 mA group. (*$p < 0.05$).