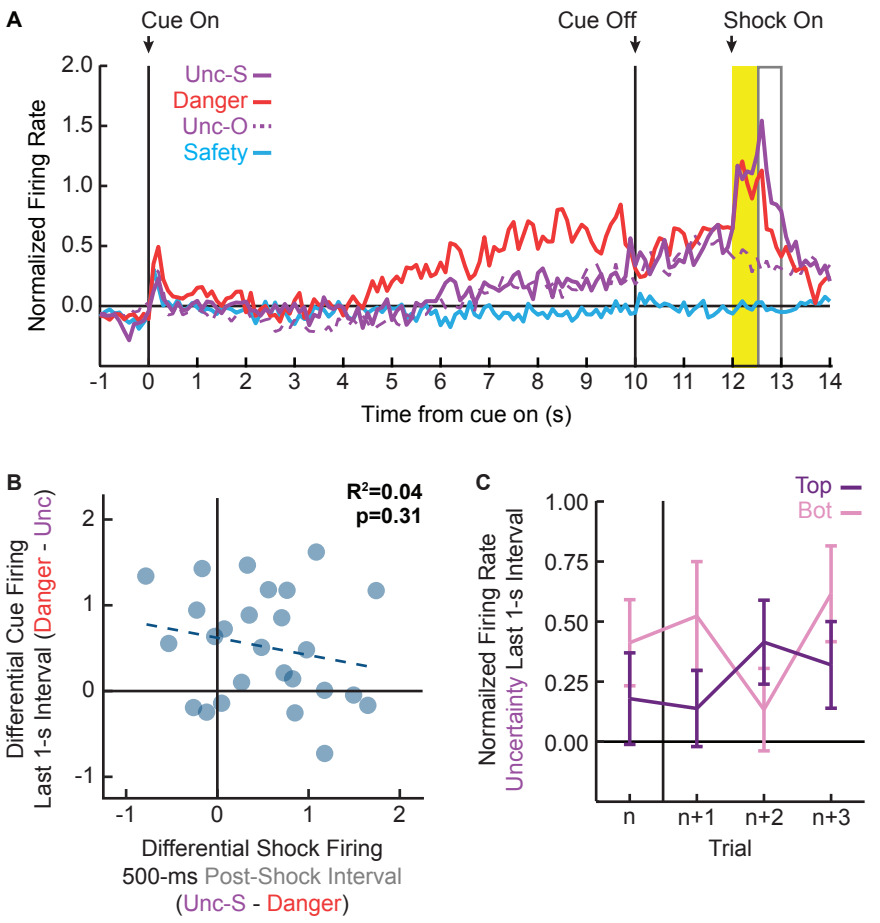

Figure S1. Cue firing and cue updating by shock-responsive neurons. (A) Mean normalized firing rate is plotted for each trial type (danger, red; uncertainty, purple; safety, blue) over the cue presentation period for the 26 shock-responsive neurons. The shock-responsive population showed non-selective firing at cue onset that gave way to selective firing over cue presentations. In support, ANOVA for normalized firing rate in the 1-s pre-cue and 10-s cue period (500 ms bins) found main effects of trial-type, bin and the trial-type x bin interaction (all $F > 5.55$, all $p < 0.00001$). **(B)** Differential firing to foot shock on uncertainty-shock trials over danger trials had no predictive relationship to differential firing to the danger vs uncertainty cue. **(C)** Unlike fear to uncertainty on subsequent trials (Fig. 3A), single trials with high +PE activity did not increase subsequent firing to the uncertainty cue. ANCOVA for normalized firing to uncertainty using trial 'n' firing as a covariate failed to find a PE firing x cue firing interaction ($F_{(3,201)} = 0.49$, $p = 0.688$) Thus, vlPAG shock-responsive neurons did not update their own cue firing.

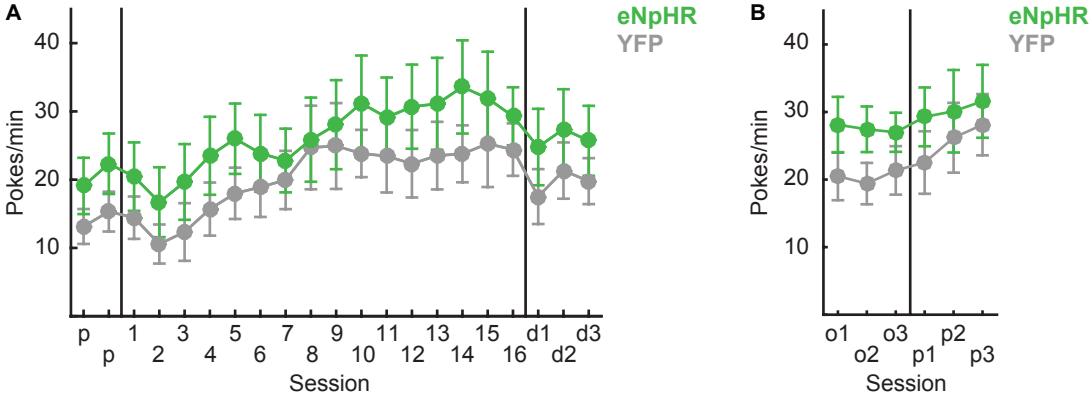

Figure S2. Baseline nose poke rates. (A) Mean ± SEM baseline nose poke rates for YFP (gray) and eNpHR rats (green) are plotted for the two pre-exposure (p), 16 discrimination (1-16), and 3 'dummy' tethred-only sessions (d1-d3). YFP and eNpHR rats did not differ in baseline nose pokes during these sessions, but males poked at higher rates than females ($F(1,9) = 6.20$, $p < 0.05$). **(B)** Mean ± SEM baseline nose poke rates are plotted for the three illumination (o1-o3) and 3 post-illumination untethered sessions (p1-p3) for YFP (gray) and eNpHR (green) rats. Again, no group differences in baseline nose poke rates were found, but there was a trend toward significance for higher rates in males compared to females ($F(1,9) = 4.88$, $p = 0.055$).

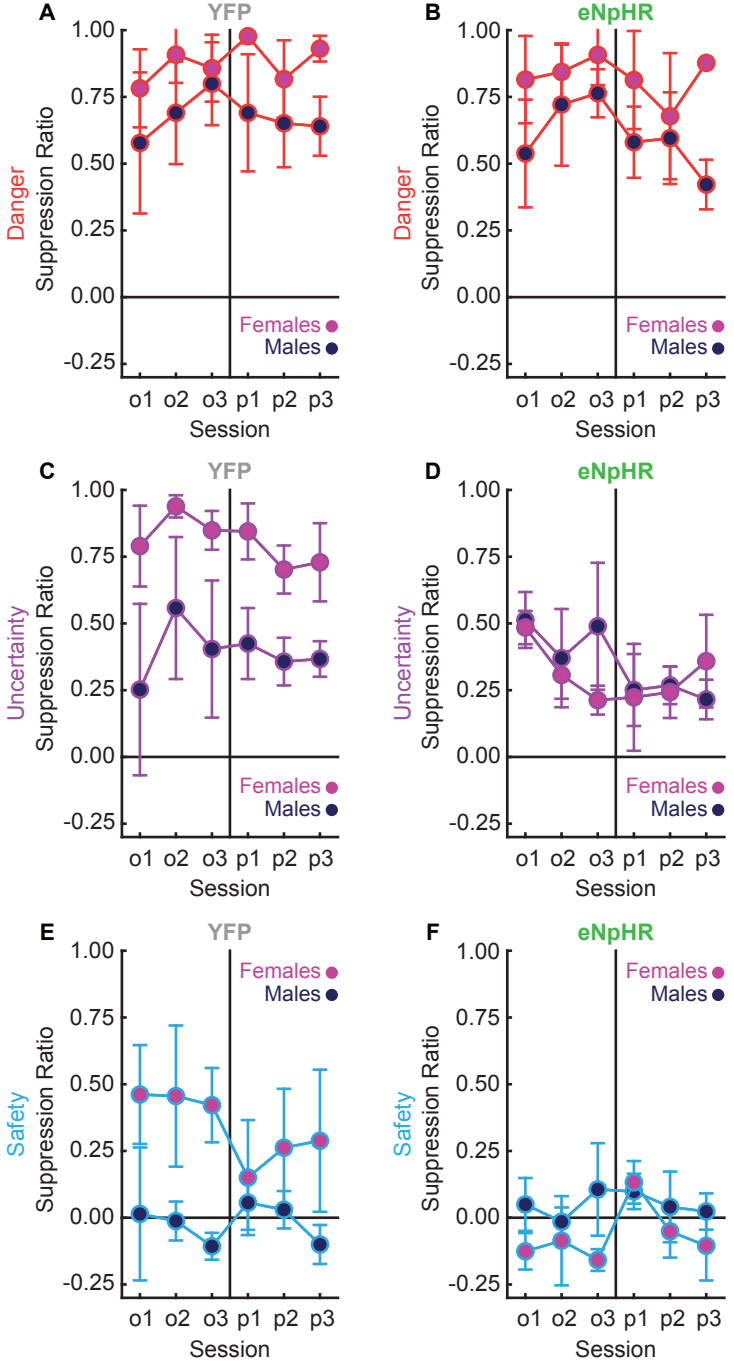

Figure S3. Optogenetics fear discrimination by sex. Mean ± SEM suppression ratio to the danger cue for YFP **(A)** and eNpHR **(B)** female (pink) and male (dark blue) rats are plotted for the 3 sessions of optogenetic manipulation (o1-o3) and 3 post-manipulation sessions (p1-p3). For the danger cue, there was no main effect of or interactions with group (all $F < 0.628$, $p > 0.05$).

Mean ± SEM suppression ratio to the uncertainty cue for YFP **(C)** and eNpHR **(D)** female (pink) and male (dark blue) rats are plotted for the 3 sessions of optogenetic manipulation (o1-o3) and 3 post-manipulation sessions (p1-p3). For the uncertainty cue, a main effect of group ($F_{1,9} = 9.68$, $p < 0.05$, $\eta^2_p = 0.52$, power = 0.79) and group x sex interaction reached significance ($F_{1,9} = 6.90$, $p < 0.05$, $\eta^2_p = 0.43$, power = 0.65), but there was no other interactions with group (all $F < 2.19$, $p > 0.05$).

Mean ± SEM suppression ratio to the safety cue for YFP **(E)** and eNpHR **(F)** female (pink) and male (dark blue) rats are plotted for the 3 sessions of optogenetic manipulation (o1-o3) and 3 post-manipulation sessions (p1-p3). For the safety cue, a session x group x sex interaction reached significance ($F_{5,45} = 2.59$, $p < 0.05$, $\eta^2_p = 0.22$, power = 0.75), but there was no main effect of or other interactions with group (all $F < 3.75$, $p > 0.05$).