Cell Reports, Volume *22*

Supplemental Information

Cooperative CRF and α1 Adrenergic Signaling in the VTA Promotes NMDA Plasticity and Drives Social Stress Enhancement of Cocaine Conditioning

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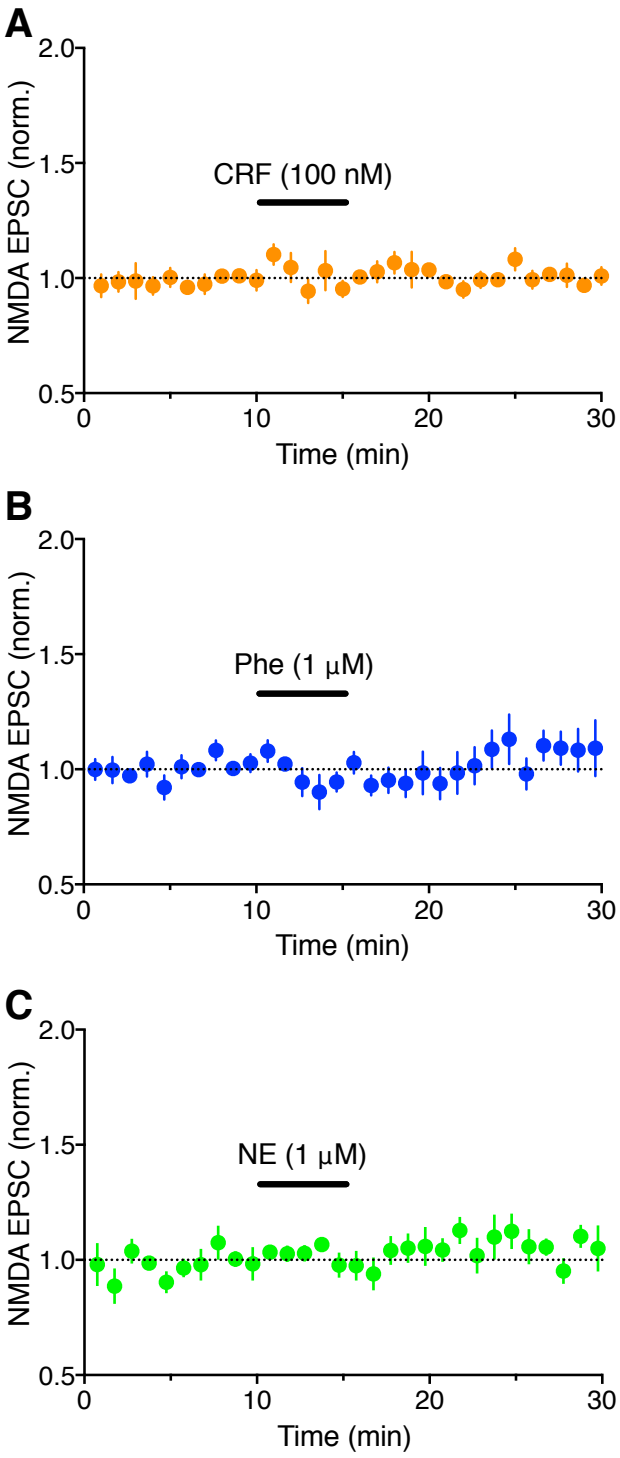

Figure S1. CRF, phenylephrine, and NE do not affect NMDA transmission, Related to Figures 1-3

Summary time graphs showing that CRF (A: n = 5), phenylephrine (B: n = 5), and NE (C: n = 7) have no measurable effect on NMDA EPSCs. The EPSC amplitude in CRF, phenylephrine, and NE was not different from baseline EPSC amplitude (two-tailed paired t-test).

Data are presented as mean ± SEM.

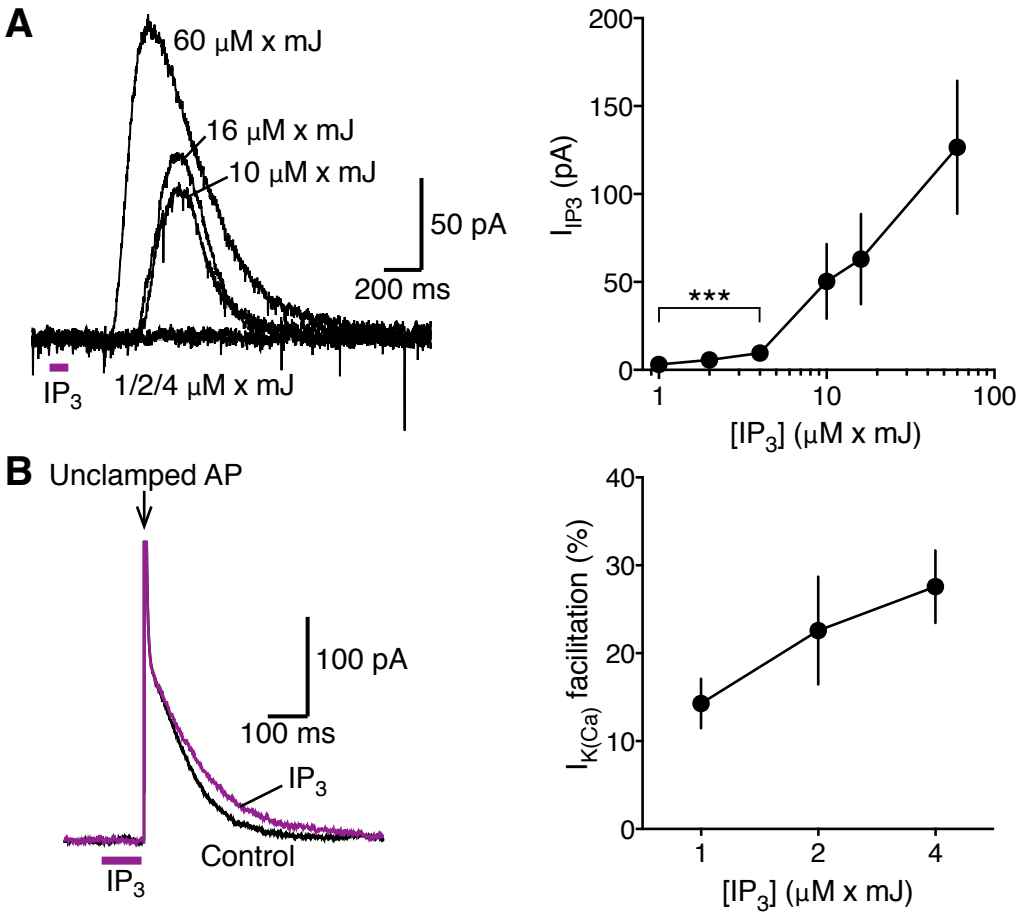

Figure S2. Concentration dependence of IP_3 responses, Related to Figure 1

(A) Example traces and summary graph depicting the concentration dependence of IP_3-evoked SK currents (I_{IP3}). Data were obtained from 7 cells, where six different IP_3 concentrations (1, 2, 4, 10, 16, and 60 μM x mJ; photolytically applied for 100 ms) were tested in each cell ($F_{5,30} = 3.42$, $p < 0.05$, repeated measures one-way ANOVA). ***$p < 0.001$ vs 60 μM x mJ (Bonferroni post hoc test).

(B) Example traces (using 4 μM x mJ IP_3) and summary graph illustrating facilitation of AP-evoked $I_{K(Ca)}$ caused by low levels of IP_3 (1, 2, and 4 μM x mJ; n = 14, 7, and 7, respectively; $F_{2,25} = 3.03$, $p = 0.067$, one-way ANOVA). Note the relatively long latency (~200-400 ms) following application of higher concentrations of IP_3 (10, 16, and 60 μM x mJ) to evoke measureable I_{IP3}, which reflects the time required to engage the regenerative IP_3R-mediated Ca^{2+}-induced Ca^{2+} release process. In contrast, IP_3 effect on AP-evoked $I_{K(Ca)}$ occurs with no latency, as rapid Ca^{2+} influx triggered by APs initiates the Ca^{2+}-induced Ca^{2+} release process, which can be augmented by low levels of IP_3.

Data are presented as mean ± SEM.

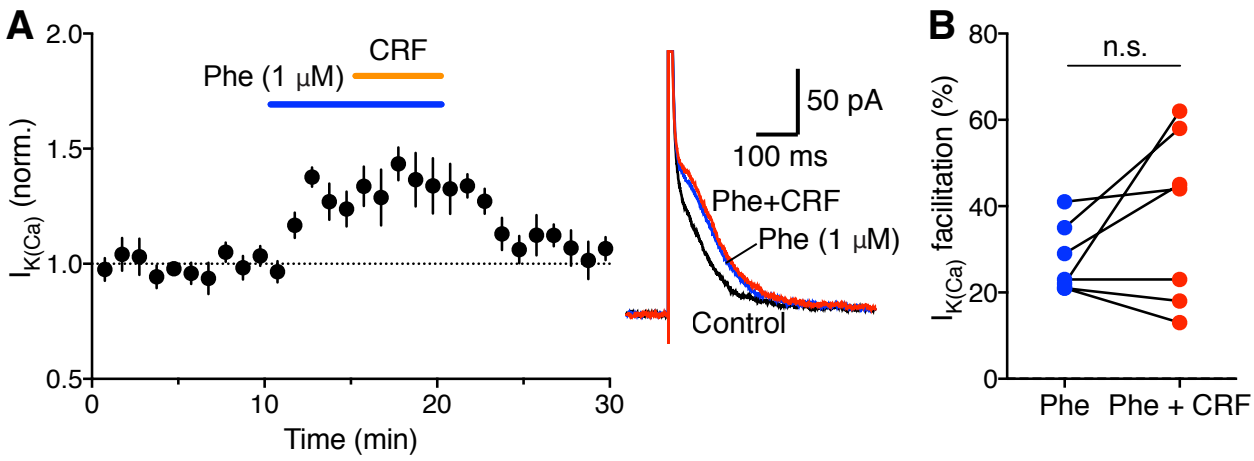

Figure S3. CRF does not enhance the effects of high-concentration phenylephrine, Related to Figure 4

(A) Summary time graph (left) and example traces (right) showing that CRF does not have significant effect on AP-evoked $I_{K(Ca)}$ facilitated by a high concentration (1 μM) of phenylephrine (n = 9).

(B) Graph plotting the magnitude of $I_{K(Ca)}$ facilitation caused by phenylephrine (1 μM) alone and by CRF and phenylephrine in individual cells ($t_6 = 1.57$, $p = 0.17$, two-tailed paired t-test). Data are presented as mean ± SEM.

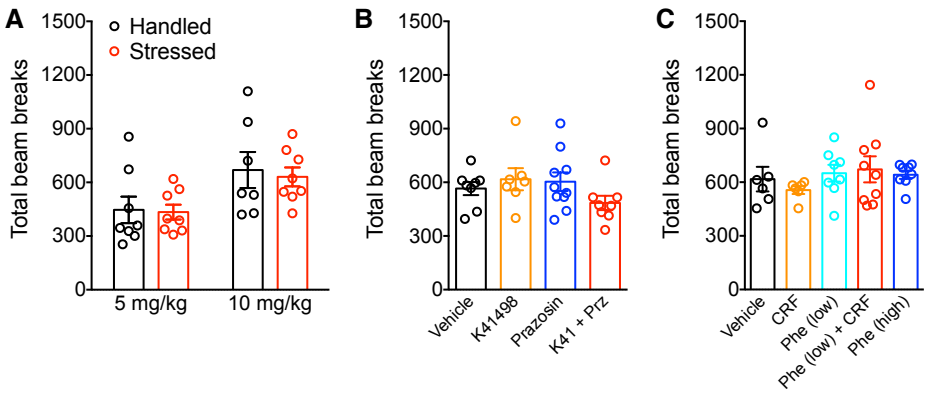

Figure S4. Social defeat stress and VTA microinjections do not affect locomotor activity during cocaine conditioning, Related to Figures 6 and 7

(A) Locomotor activity during cocaine conditioning of rats subjected to social defeat stress 10 min prior to conditioning ($F_{1,27}$ = 0.035, p = 0.85; two-way ANOVA; n = 7-8 rats).

(B) Locomotor activity during cocaine conditioning of rats subjected to social defeat stress and administered various antagonists into the VTA prior to stress ($F_{3,29}$ = 1.45, p = 0.25; one-way ANOVA; n = 7-9 rats).

(C) Locomotor activity during cocaine conditioning of rats administered various agonists into the VTA prior to conditioning ($F_{4,32}$ = 0.62, p = 0.65; one-way ANOVA; n = 6-9 rats).

Data are presented as mean ± SEM.

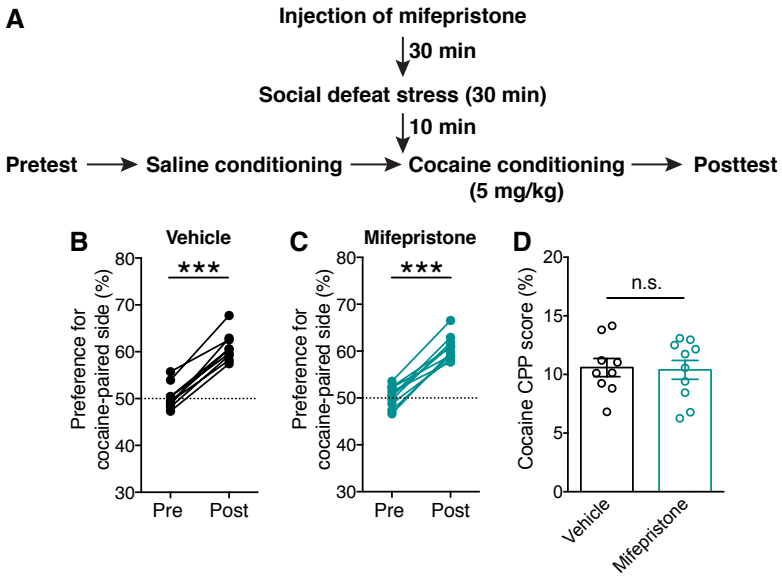

Figure S5. Mifepristone administration does not prevent social defeat stress enhancement of cocaine conditioning, Related to Figure 7

(A) Experimental timeline for testing the effects of mifepristone injections on defeat stress-induced enhancement of cocaine conditioning.

(B–C) Changes in the preference for the cocaine-paired side (conditioned with 5 mg/kg cocaine) in socially defeated rats that received systemic injection of vehicle (B) or mifepristone (C) (B: $t_8 = 13.6$, $p < 0.001$; C: $t_9 = 12.94$, $p < 0.001$; two-tailed paired t-test; n = 9-10 rats).

(D) Summary graph demonstrating independence of glucocorticoid receptors for stress-induced enhancement of cocaine conditioning ($t_{17} = 0.17$, $p = 0.86$; two-tailed unpaired t-test).

Data are presented as mean ± SEM.

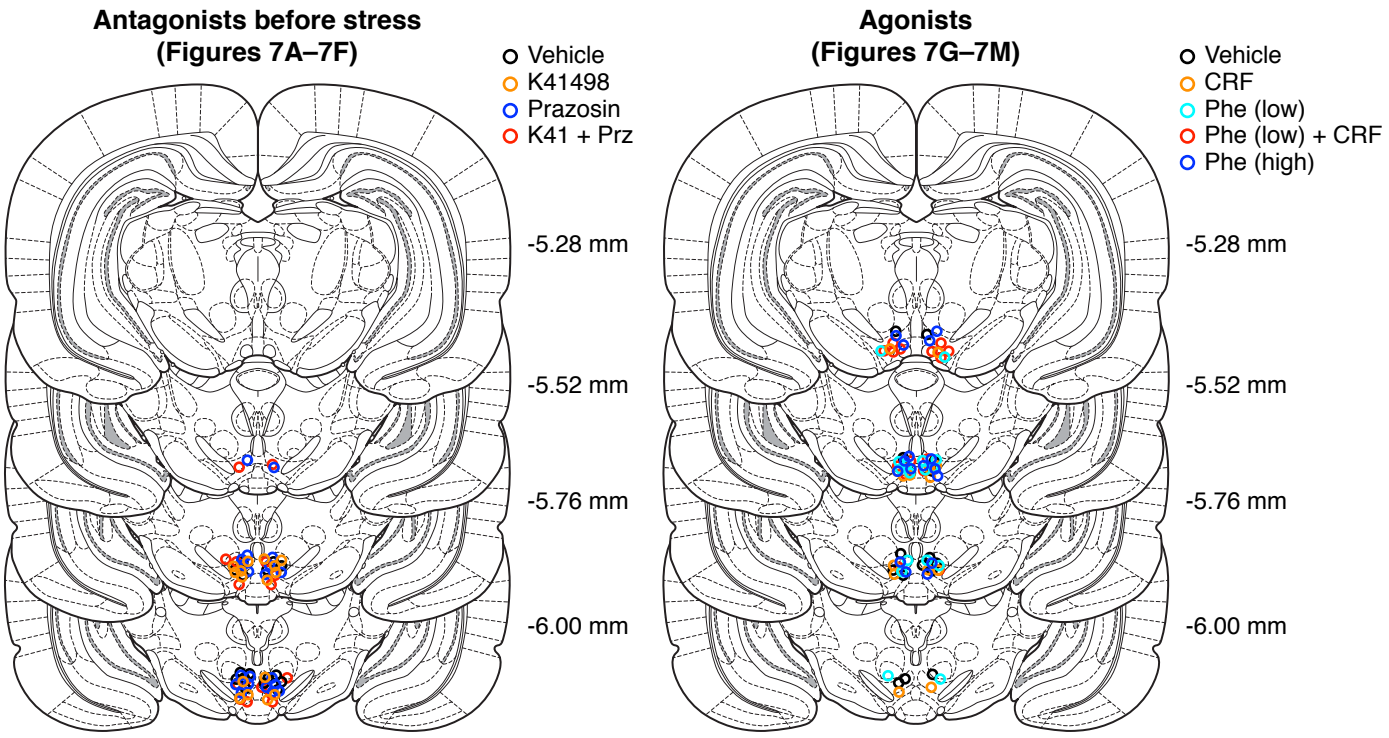

Figure S6. Cannula locations in the VTA, Related to Figure 7

Approximate locations (mm from bregma) of cannula tips for intra-VTA microinjection experiments in Figure 7.