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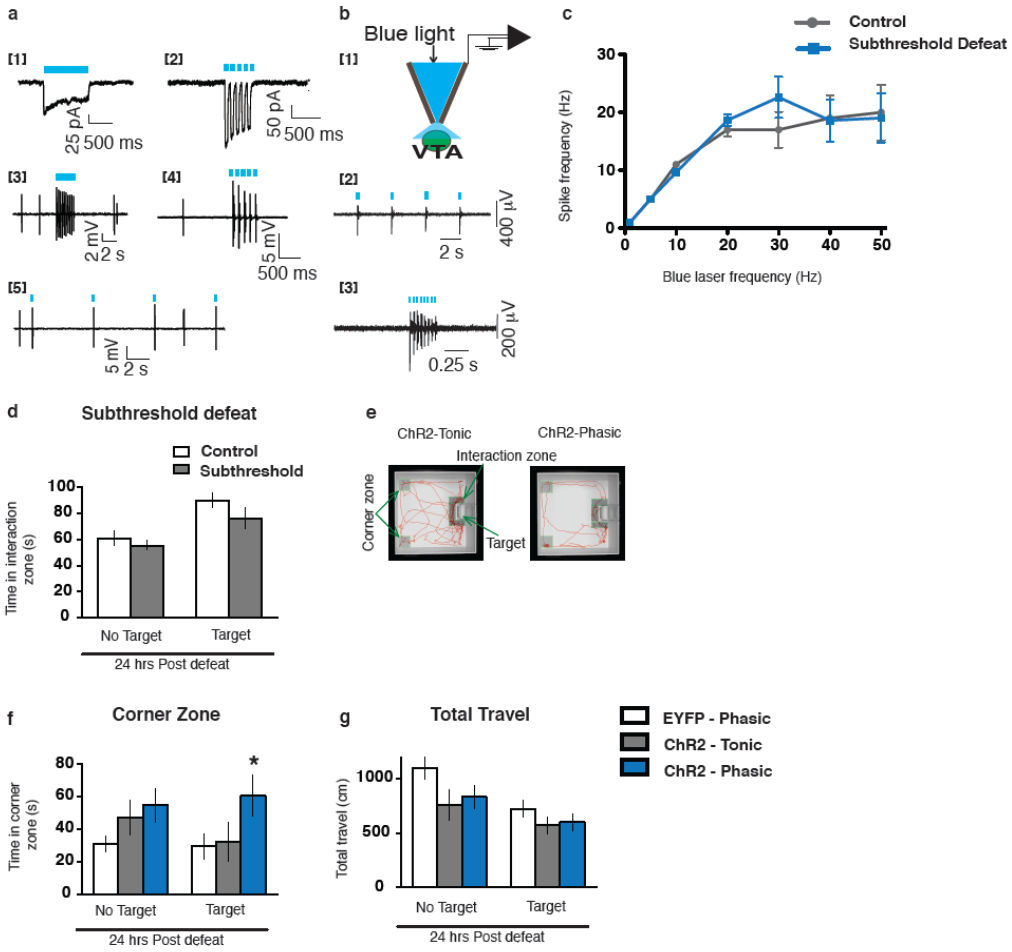
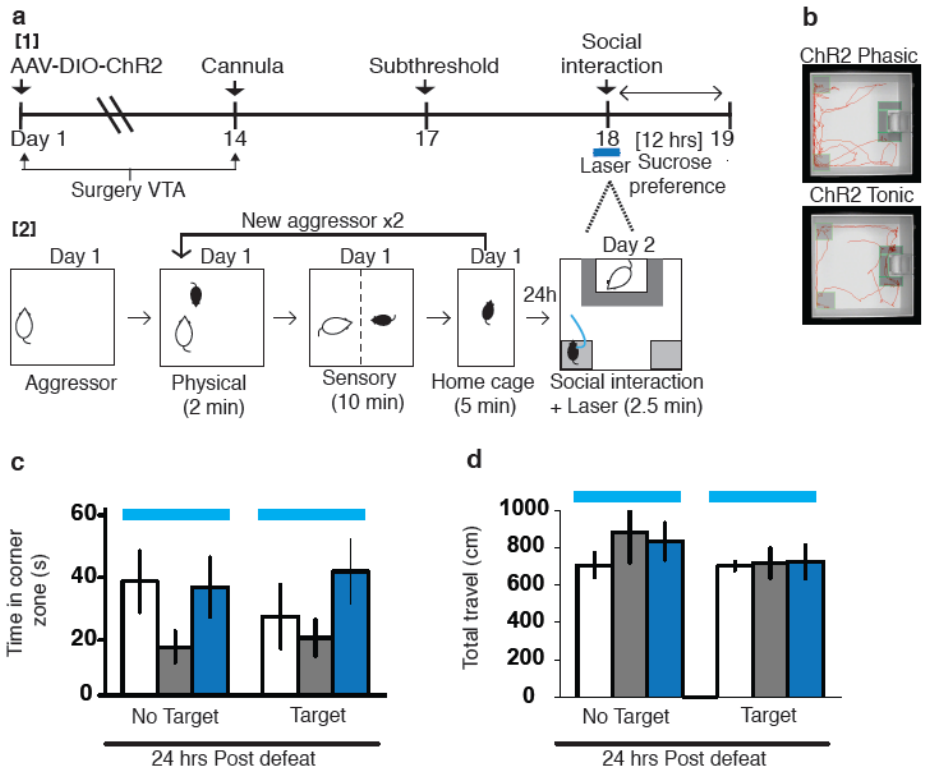
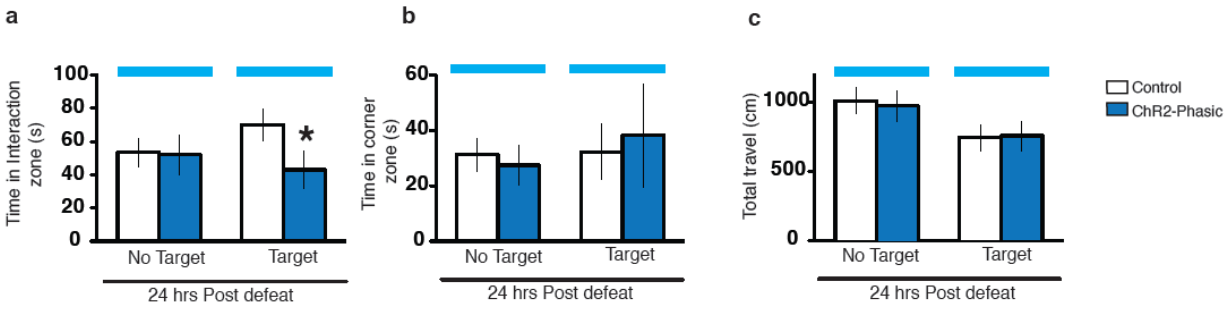
Supplementary figure 1: Functional validation of DIO-ChR2 and optical stimulation during subthreshold social defeat. a1-2, Whole cell, voltage clamp recordings from DIO-ChR2-EYFP infected DA neurons in VTA slices, obtained from TH-Cre mice, show that long duration (1 sec) or five short bursts (20 Hz, 40 ms) of blue light (470 nm) induce temporally precise inward photocurrents. **a3-5,** Cell-attached current-clamp recordings show that long duration (2 sec) or five short bursts (20 Hz, 40 ms) of blue light induce phasic firing while single short light pulses (0.1 Hz) induce tonic firing of VTA DA neurons. **b1**, Schematic showing the optrode (optical electrode) used for *in vivo* delivery of blue light and simultaneous extracellular recording of VTA DA neuron responses in anaesthetized mice. **b2-3,** Extracellular, *in vivo* response of VTA DA neurons to 0.25 Hz and 25 Hz optical stimulation induces tonic and phasic firing, respectively. **c**, Frequency-response curve of membrane excitability to optical stimulation in VTA DA neurons obtained from mice that went through subthreshold social defeat or non-defeated controls. Light frequencies ranging from 0.1-20 Hz reliably induce the approximately equivalent firing rates in ChR2-expressing VTA DA neurons (i.e., 10 Hz light pulse yields ~10 Hz evoked firing). **d,** Mice that undergo the subthreshold social defeat paradigm do not typically exhibit the defeated phenotype such as social avoidance. These mice spend roughly the same amount of time in the interaction zone, in the presence of a social target, as naïve, undefeated mice. **e**, Representative traces showing mouse behaviour during the social interaction test after having undergone a subthreshold social defeat paradigm combined with optical stimulation 24 hr earlier. **f,** In the absence of a social target, there is no significant difference between EYFP control, ChR2 tonic and ChR2 phasic groups, although there is a trend showing an increase in the time spent in the corner zone in phasic and tonic stimulated mice compared to EYFP control mice. In contrast, in the presence of a target, phasic stimulated mice consistently spent significantly more time in the corner zone compared to EYFP control mice ($F_{2,36} = 4.30$, $p<0.05$; *post-hoc* test: * denotes $p<0.05$; n=9-16). **g,** There was no statistical difference in total travel distances between the 3 groups, though there was a trend for tonic and phasic stimulated mice to travel less compared to EYFP control mice in the absence or presence of a target (no target: $F_{2,36} = 2$, $p>0.05$; target: $F_{2,36} = 0.5$, $p>0.05$). All error bars indicate ± s.e.m.

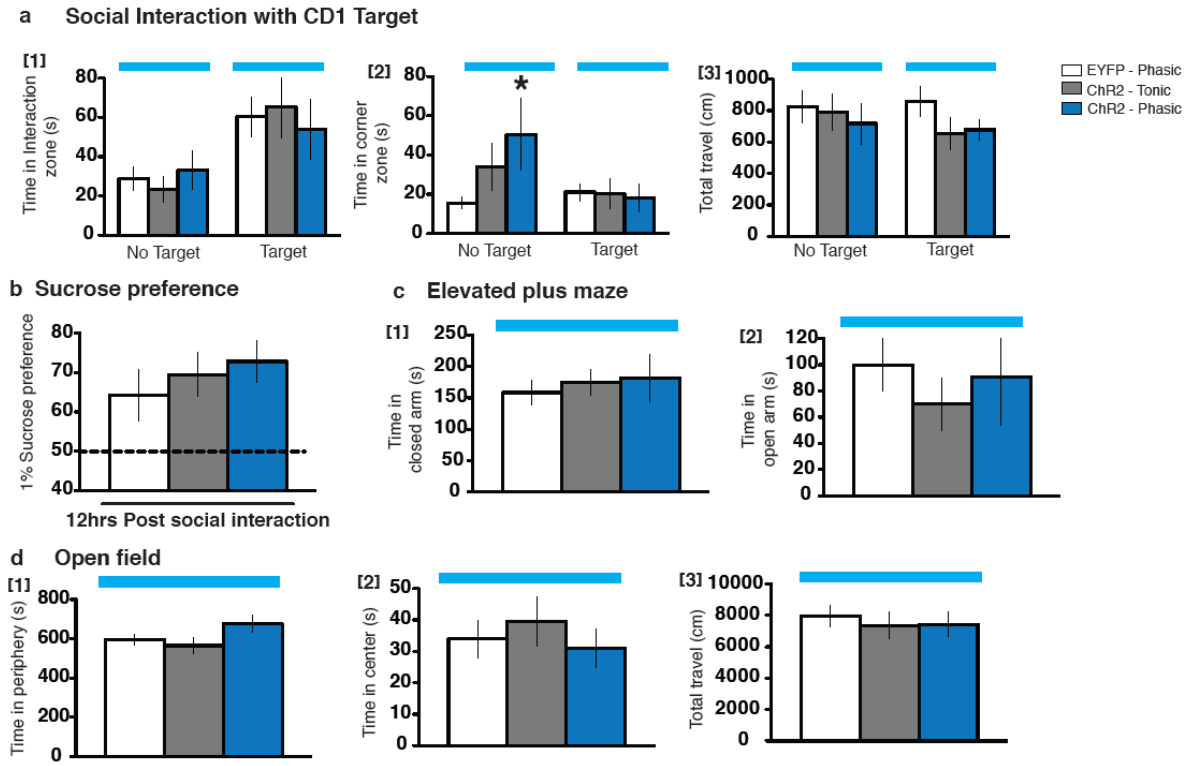




Supplementary figure 2: Stimulation during social interaction test using DIO-ChR2-EYFP in TH-Cre mice. **a1,** Experimental timeline. **a2,** Detailed schematic of the subthreshold defeat paradigm showing optical stimulation during the social interaction test. **b,** Representative traces showing mouse behaviour during the social interaction test. **c,** In the absence of a social target, though not significant, there was a trend showing tonic stimulated mice spending slightly less time in the corner zone compared to control and phasic stimulated mice. In the presence of a target, there is a trend showing phasic stimulated mice spending more time in the corner zone compared to EYFP and tonic stimulated mice (no target: $F_{2,22} = 1.4$; target: $F_{2,22} = 1.3$, $p>0.05$ n=7-11). **d,** There was no difference in total travel distance during the social interaction test between the three groups (no target: $F_{2,22} = 0.5$; target: $F_{2,22} = 0.02$, $p>0.05$ n=7-11). All error bars indicate ± s.e.m.

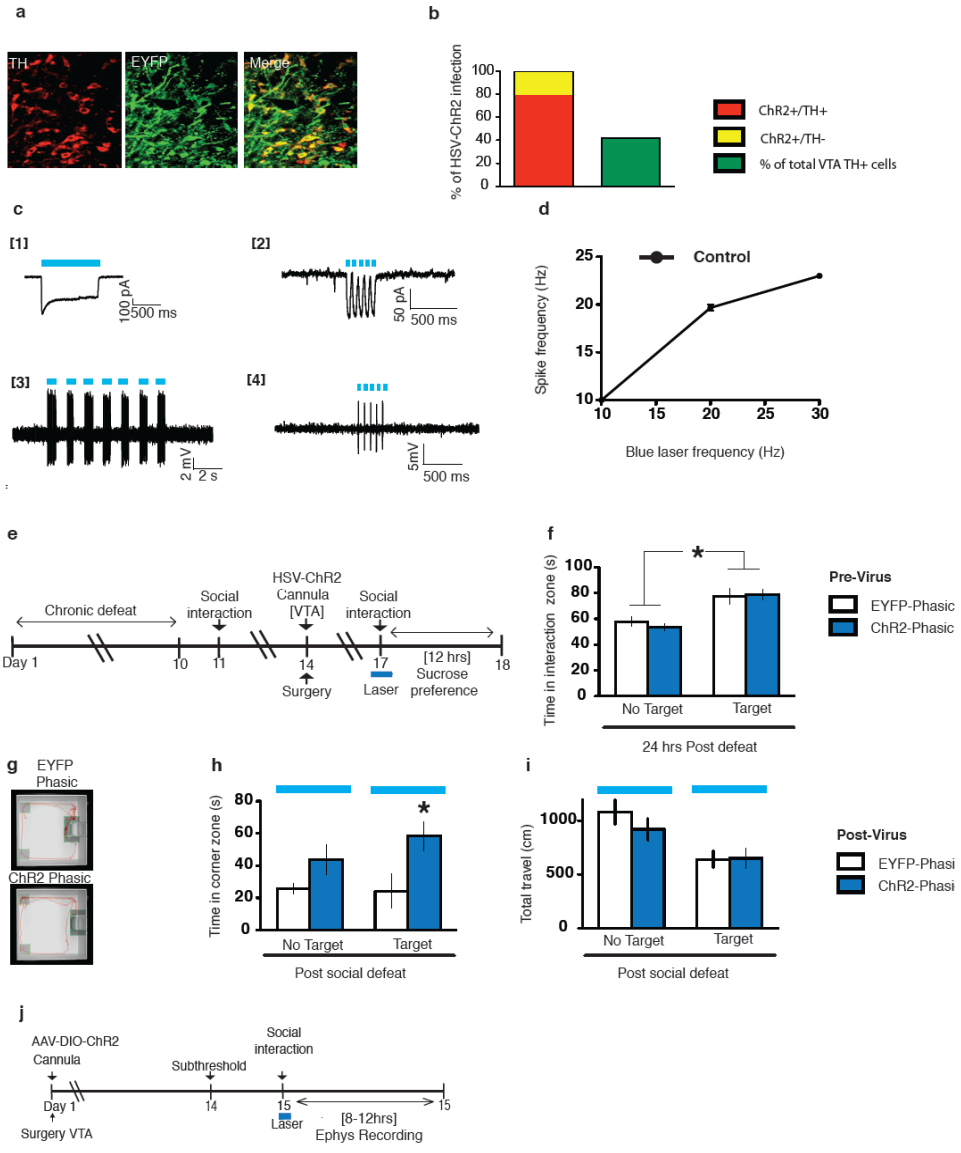


Supplementary figure 3: Phasic opto-stimulation of VTA DA neurons induces social avoidance during the social interaction test even when the social target is a C57 mouse. a, In the presence of a C57 social target, phasic (20 Hz) stimulated mice, that had previously undergone subththreshold social defeat, spent significant less time in the interaction zone compared to control (EYFP and tonic combined) mice (t_{13} = 1.8, * $p<0.05$; one-tailed t-test, n=6-9). **b,** In the presence of target, phasic stimulated mice tended to spend more time in the corner zone compared to control mice (no target: t_{13} = 0.4; target: t_{13} = 0.3 $p>0.05$). **c,** There was no difference in total travel between control and phasic stimulated mice (no target: t_{13} = 0.2; target: t_{13} = 0.1 $p>0.05$). All error bars indicate ± s.e.m.

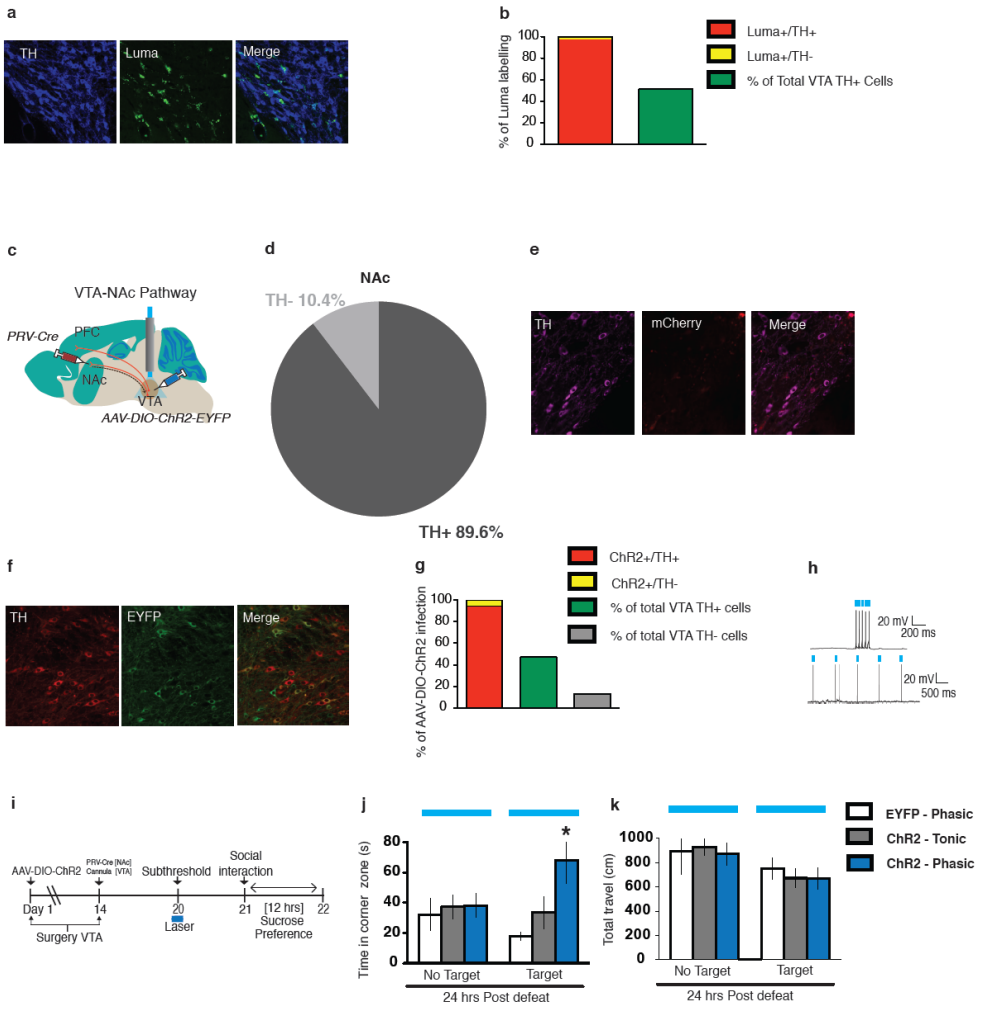


Supplementary figure 4: Optical stimulation of VTA DA neurons of stress naïve mice during a social interaction, open field, or elevated plus maze test does not affect baseline behaviours. **a1,** In the presence of a social target, stress naïve (non-defeated) mice that received either phasic (20 Hz) or tonic (0.5 Hz) optical stimulation of VTA DA neurons spent the same amount of time in the interaction zone as control groups. **a2,** While in the absence of a social target, phasic stimulated mice spent significantly longer time in the corner zone compared to EYFP control ($F_{2,24} = 2.50$, $p<0.05$; *post-hoc* test: * denotes $p<0.05$; n=8-11), in the presence of a target there was no difference in time spent in the corner zone between the three groups. **a3,** There was no significant difference in the total travel distances of phasic and tonic stimulated mice compared to the EYFP control group ($F_{2, 24} = 1.61$, $p>0.05$; n=8-11). **b,** There was no difference in 1% sucrose preference between the 3 groups ($F_{2, 23} = 1.61$, $p>0.05$; n=8-9). **c1-2,** There was no significant difference in time spent in the open or closed arm of the elevated plus maze between these 3 groups of mice that received phasic or tonic optical stimulation during the test (open arm: $F_{2, 25} = 0.3$; closed arm: $F_{2, 25} = 0.2$, $p>0.05$; n=6-14). **d1-3,** Optical stimulation during the open field behaviour test did not induce any difference between control EYFP, tonic, and phasic stimulation groups (periphery: $F_{2, 25} = 2$; middle: $F_{2, 25} = 0.4$; total travel: $F_{2, 25} = 0.2$, $p>0.05$ n=6-14). All error bars indicate ± s.e.m.

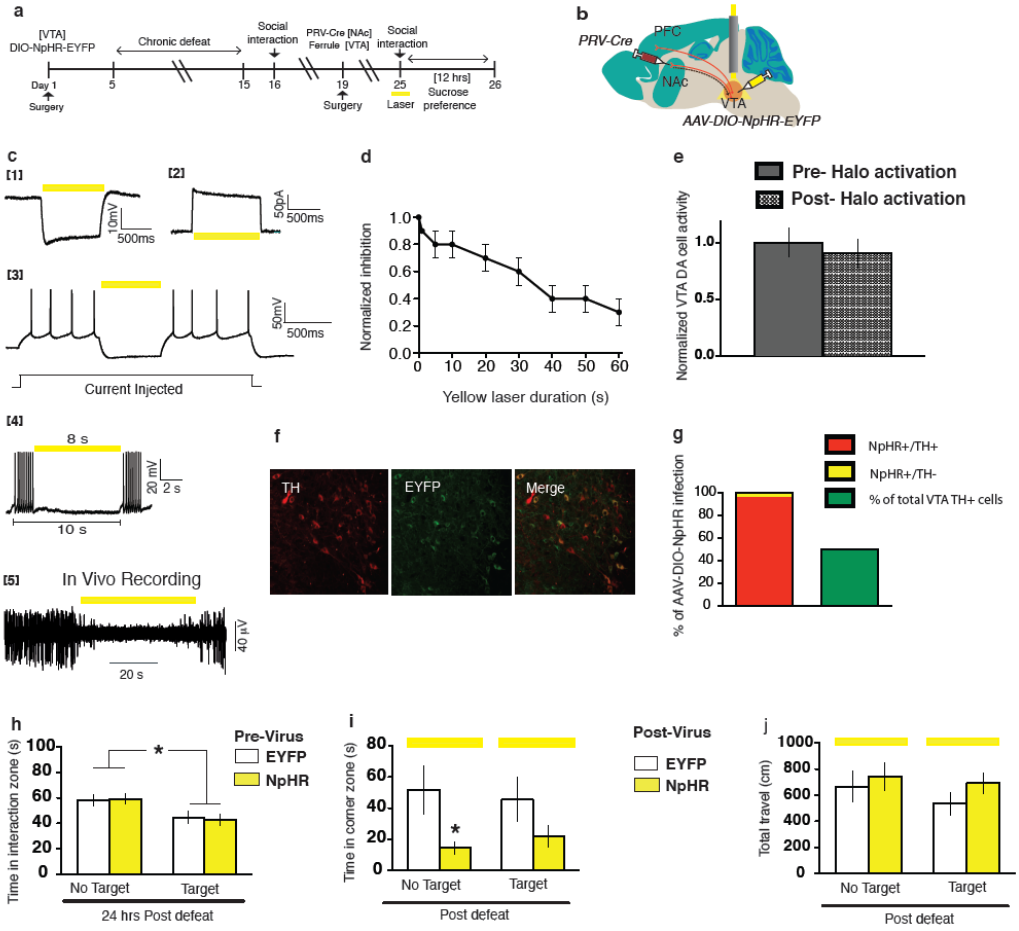
Supplementary figure 5: HSV-ChR2 validation and stimulation during social interaction. a, Confocal images showing co-expression of HSV-ChR2-EYFP in TH+ DA cells. **b,** Quantification of HSV-ChR2-EYFP infection shows that 80±2% of ChR2-expressing neurons were TH+ while 20±2% were TH-. These TH+ ChR2-expressing neurons were 42±5% of total VTA TH+ cells (n=2 sections from n=5 animals). **c1-2**, Whole cell, voltage clamp recordings from HSV-Chr2-EYFP infected VTA DA neurons in slices show that long duration (1 sec) or five short bursts (20 Hz, 40 ms) of blue light (470 nm) induce temporally precise inward photocurrents. **c3-4,** Cell-attached current-clamp recordings show that multiple 1 sec duration or five short bursts (20 Hz, 40 ms) of blue light induced phasic firing. **d**, Frequency-response curve of membrane response to optical stimulation in VTA DA neurons expressing HSV-ChR2-EYFP from control, non-defeated mice. **e,** Experimental timeline for testing if phasic optical stimulation of VTA neurons converts previously resilient mice to susceptible mice. **f,** Both groups of resilient mice (Pre-EYFP and Pre-ChR2 injection) spent significantly more time in the interaction zone in the presence of social target during the 1st social interaction test ($F_{3,54}$ = 9, p<0.001; post-hoc: * denotes p<0.05; n=13-16). **g,** Representative traces: top, mice injected with control HSV-EYFP virus; bottom, mice injected with HSV-ChR2-EYFP virus. After repeated social defeat stress, resilient mice were identified and half were injected with each vector. Both groups were then given phasic stimulation during the second social interaction test. **h,** In the presence of a social target, previously resilient mice injected with HSV-ChR2-EYFP and exposed to phasic stimulation during the 2nd social interaction test spent significantly longer time in the corner zone compared to resilient mice injected with HSV-EYFP (t_{17} = 2.45, * p<0.05; two tailed t-test, n=11-18). **i,** There was no difference in total travel distance during the social interaction test between the three groups (no target: t_{17} = 1.1; target: t_{17} = 0.01; p>0.05; n=11-18). **j**, Timeline of experiments measuring changes in intrinsic membrane properties of VTA DA neurons following optical stimulation during the social interaction test. All error bars indicate ± s.e.m.



Supplementary figure 6: Validation of retrobead labeling, PRV-Cre infection, and DIO-ChR2 expression in the VTA-NAc pathway followed by ChR2 stimulation of this pathway during the social interaction test. a, Confocal images showing distribution of green retrogradely transported retrobeads, lumafluors, in the VTA. **b**, Quantification of lumafluor labeling shows that 98±1% of lumafluor labeled cells were TH+ and 2±7% were TH-. These TH+ lumafluor-labeled neurons were 51±4% of total VTA TH+ neurons (n=2-5 sections from n=5 animals). Fewer than 1% of labeled neurons project to both NAc and mPFC (data not shown). **c**, Schematic showing PRV-Cre injected into NAc and AAV-DIO-ChR2-EYFP injected into VTA so that ChR2 expresses specifically in VTA-NAc neurons. **d**, Quantification in the VTA of neuronal expression of Cre following PRV-Cre injection into the NAc, shows that 90±1% of retrograde Cre-expressing VTA cells were TH+ neurons, whereas the rest of them were TH-. **e**, Confocal images showing colocalization of TH+ DA cells with retrogradely labeled PRV-Cre from NAc. **f**, Confocal images showing expression of DIO-ChR2-EYFP in DA cells projecting to NAc in C57 mice injected with PRV-Cre in the NAc. **g**, Quantification of DIO-ChR2-EYFP infection in PRV-Cre injected mice shows that 95±0.3% of ChR2-expressing neurons were TH+ and 5±0.2% were TH-. These TH+ ChR2-expressing neurons were 47±2% of total VTA TH+ neurons, while 13±3% of total VTA TH- neurons expressed ChR2, which is about 2% of total VTA neurons (n=3-5 sections from n=4-6 animals). **h,** Phasic (20 Hz) and tonic (0.5 Hz) light stimulation induced spiking in VTA-NAc neurons expressing ChR2. **i**, Timeline of projection specific surgery and behavioural experiments with PRV-Cre and ChR2. **j,** Mice receiving phasic activation of VTA-NAc neurons during the social interaction test, in the presence of a social target, spent significantly more time in the corner zone, compared to EYFP control mice ($F_{2,20}$ = 3.67, p<0.05; *post-hoc* test: * p<0.05; n=5-10). **k,** There was no difference in total travel distance during optical stimulation of VTA-NAc neurons during the social interaction test between the three groups (no target: $F_{2,20}$ = 0.1; target: $F_{2,20}$ = 0.2, p>0.05; n=5-10). All error bars indicate ± s.e.m.

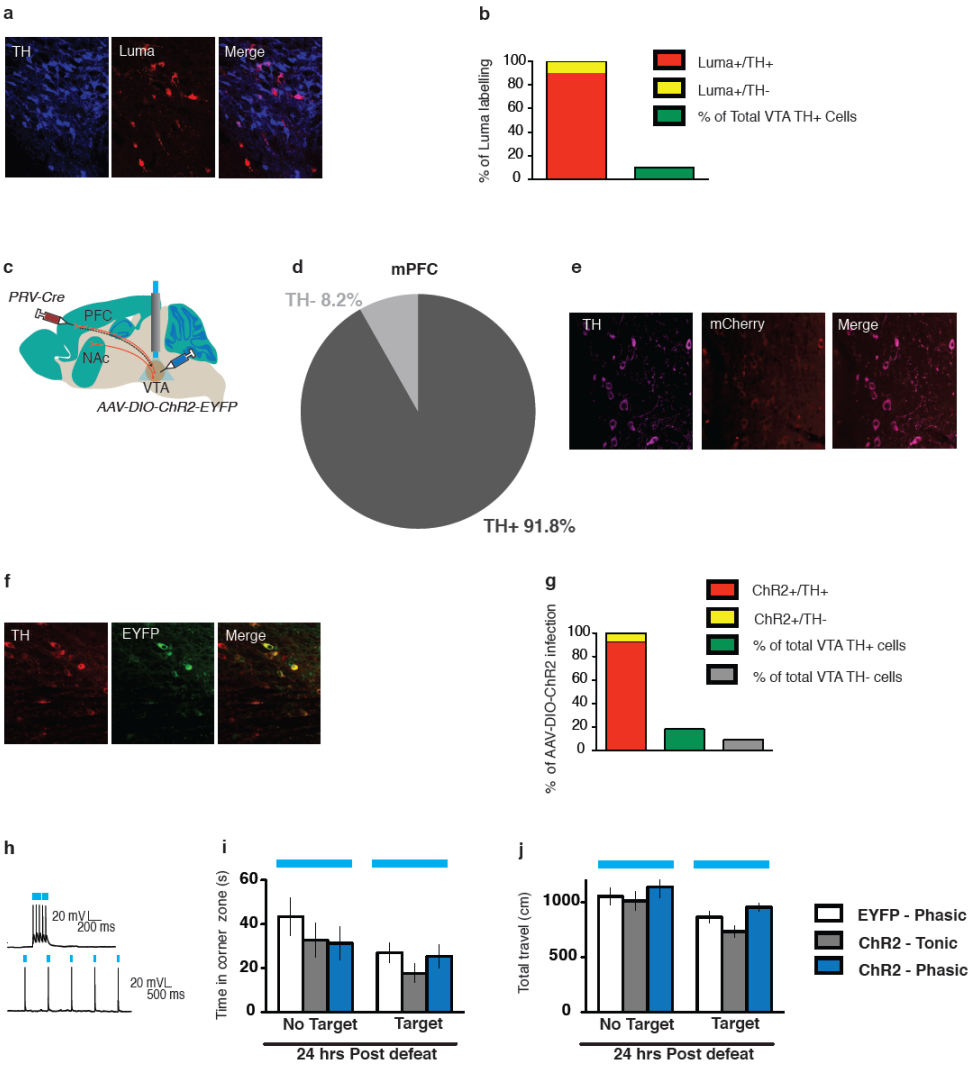


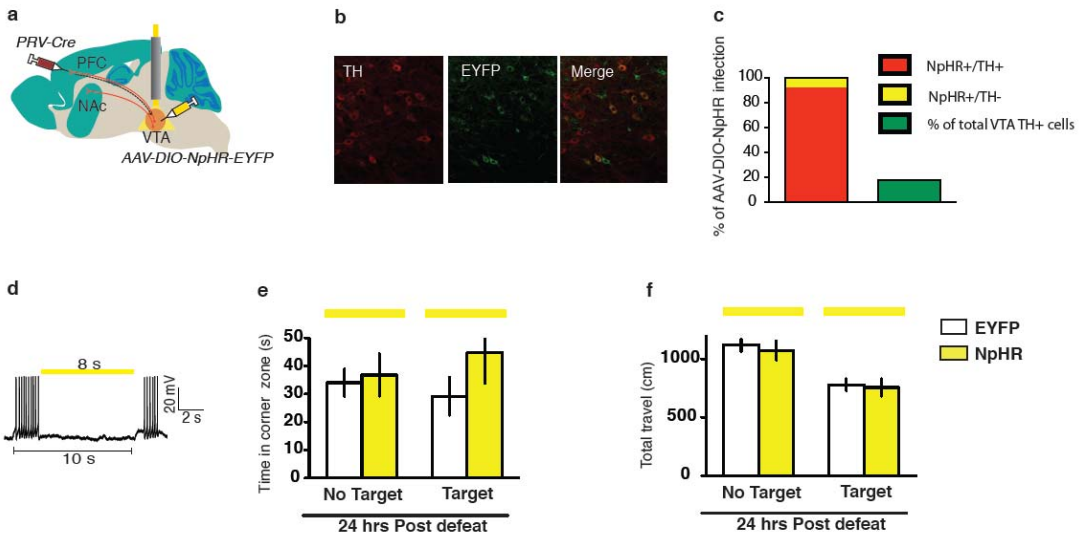
Supplementary figure 7: Validation of AAV-DIO-NpHR and inhibition of the VTA-NAc pathway. a, Time line of 'rescue' experiments. **b**, Schematic showing PRV-Cre injected into NAc and AAV-DIO-NpHR-EYFP injected into VTA so that NpHR expresses specifically in VTA-NAc neurons. **c1**, Whole cell, current clamp recording from AAV-DIO-NpHR-EYFP infected VTA DA neurons in brain slices show that long duration (1 sec) yellow light (560 nm) hyperpolarizes membrane potential. **c2**, Whole cell, voltage clamp recordings show that 1 sec yellow light induces outward photocurrents. **c3**, 1 sec yellow light inhibits current injection-induced spiking *in vitro*. **c4**, 8 sec activation (2 sec off) of NpHR expressed in VTA-NAc neurons inhibits the firing *in vitro*, but does not induce rebound. **c5**, Extracellular *in vivo* response of VTA DA neurons to 50 sec yellow light shows inhibition of spontaneous firing. **d**, Spiking inhibition-response curve of VTA DA neurons to different durations of yellow light. **e**, Yellow light protocol used in behavioural experiments (8 sec on, 2 sec off) did not induce rebound



responses in VTA DA neurons expressing DIO-NpHR-EYFP. **f**, Confocal images showing co-expression of DIO-NpHR-EYFP in TH+ DA cells projecting to NAc in C57 mice. **g**, Quantification of DIO-NpHR-EYFP infection in VTA-NAc cells shows that 97±2% of NpHR-expressing neurons were TH+ and 3±2% were TH-. These TH+ NpHR-expressing neurons were 50±4% of total VTA TH+ neurons (n=2-4 sections from n=4 animals). **h,** Both groups of susceptible mice (Pre-EYFP and Pre-NpHR injection) spent significantly less time in the interaction zone in the presence of social target during the 1st social interaction test ($F_{3,70}$ = 3.8, p<0.05; *post-hoc* test: * denotes p<0.05; n=18-19). **i**, In both the absence and presence of a social target, NpHR stimulation-induced inhibition of the VTA-NAc pathway of previously susceptible mice during a 2nd social interaction test resulted in those mice spending less time in the corner zone compared to EYFP control mice (In the absence of target: t_{15} = 2.1, * p<0.05; two tailed t-test, n=0.05). **j,** There was no difference in total travel distance during the social interaction test between the two groups (no target: t_{15} = 0.5; target: t_{15} = 1.3; p>0.05; n=8-9). All error bars indicate ± s.e.m.

Supplementary figure 8: Validation of retrobead labeling, PRV-Cre infection, and DIO-ChR2 expression in the VTA-mPFC pathway followed by ChR2 stimulation of this pathway during the social interaction test. a, Confocal images showing distribution of red lumafluor in VTA from C57 mice. **b,** Quantification of lumafluor retrograde labeling in VTA neurons projecting to mPFC shows that 91±4% of lumafluor labeled cells were TH+ and 9±6% were TH-. These TH+ lumafluor-labeling neurons were 10±2% of total VTA TH+ neurons (n=2-5 sections from n=5 animals). Fewer than 1% of labeled VTA neurons project to both NAc and mPFC (data not shown). **c,** Schematic showing PRV-Cre injected in mPFC and AAV-DIO-ChR2-EYFP injected in VTA so that ChR2 expresses specifically in VTA-mPFC neurons. **d**, Quantification of retrograde PRV-Cre infection following PRV-Cre injection into the mPFC, shows that 92±1% of Cre-expressing cells were TH+ neurons, while the rest of them were TH-. **e**, Confocal images showing colocalization of TH+ DA cells with retrogradely labeled PRV-Cre from mPFC. **f,** Confocal images showing expression of DIO-ChR2-EYFP in VTA cells projecting to mPFC in C57 mice injected with PRV-Cre in the mPFC. **g,** Quantification of DIO-ChR2-EYFP infection in VTA-mPFC neurons shows that 93± 4% of ChR2-expressing cells were TH+ and 7±4% were TH-. These TH+ ChR2-expressing neurons were 18±2% of total VTA TH+ neurons, while 9±3% of total VTA TH- neurons expressed ChR2, which is about 1-2 % of total VTA neurons (n=2-3 sections from n=3-4 animals). **h**, Phasic and tonic light stimulation induced spiking in VTA-mPFC neurons expressing ChR2. **i**, There was no difference in time spent in the corner zone (no target: $F_{2,39} = 0.7$; target: $F_{2,39} = 1.2$, $p>0.05$; n=11-17). **j,** In the presence of a target, mice that received tonic and phasic stimulations had no difference in travel distance compared to EYFP control, although phasic stimulated mice travelled more than tonic stimulated mice (no target: $F_{2,39} = 0.5$, $p>0.05$; target: $F_{2,39} = 4.2$, $p<0.05$; n=11-17). All error bars indicate ± s.e.m.





Supplementary figure 9: Validation of DIO-NpHR expression in VTA neurons projecting to mPFC and inhibition of the VTA-mPFC pathway. a, Schematic showing PRV-Cre injected into mPFC and AAV-DIO-NpHR-EYFP injected into VTA so that NpHR expresses specifically in VTA-mPFC neurons. **b**, Confocal images showing co-expression of DIO-NpHR-EYFP in TH+ DA cells projecting to mPFC in C57 mice. **c**, Quantification of DIO-NpHR-EYFP infection in VTA-mPFC cells shows that 93±7% of NpHR-expressing neurons were TH+ and 7±7% were TH-. These TH+ NpHR-expressing neurons were 18±2% of total VTA TH+ neurons (n=3 sections from n=5 animals). **d**, Behavioural protocol used for NpHR activation-induced inhibition of VTA neurons projecting to mPFC *in vitro*. **e,** In the presence of a social target, NpHR activation-induced inhibition of the VTA-mPFC pathway during the social interaction test resulted in mice spending relatively longer time in the corner zone compared to mice injected with DIO-EYFP (no target: $t_{29} = 0.3$; target: $t_{29} = 1.3$; $p>0.05$; n=12-19). **f**, There was no difference in total travel distance during the social interaction test between the two groups (no target: $t_{29} = 0.4$; target: $t_{29} = 0.3$; $p>0.05$; n=12-19). All error bars indicate ± s.e.m.