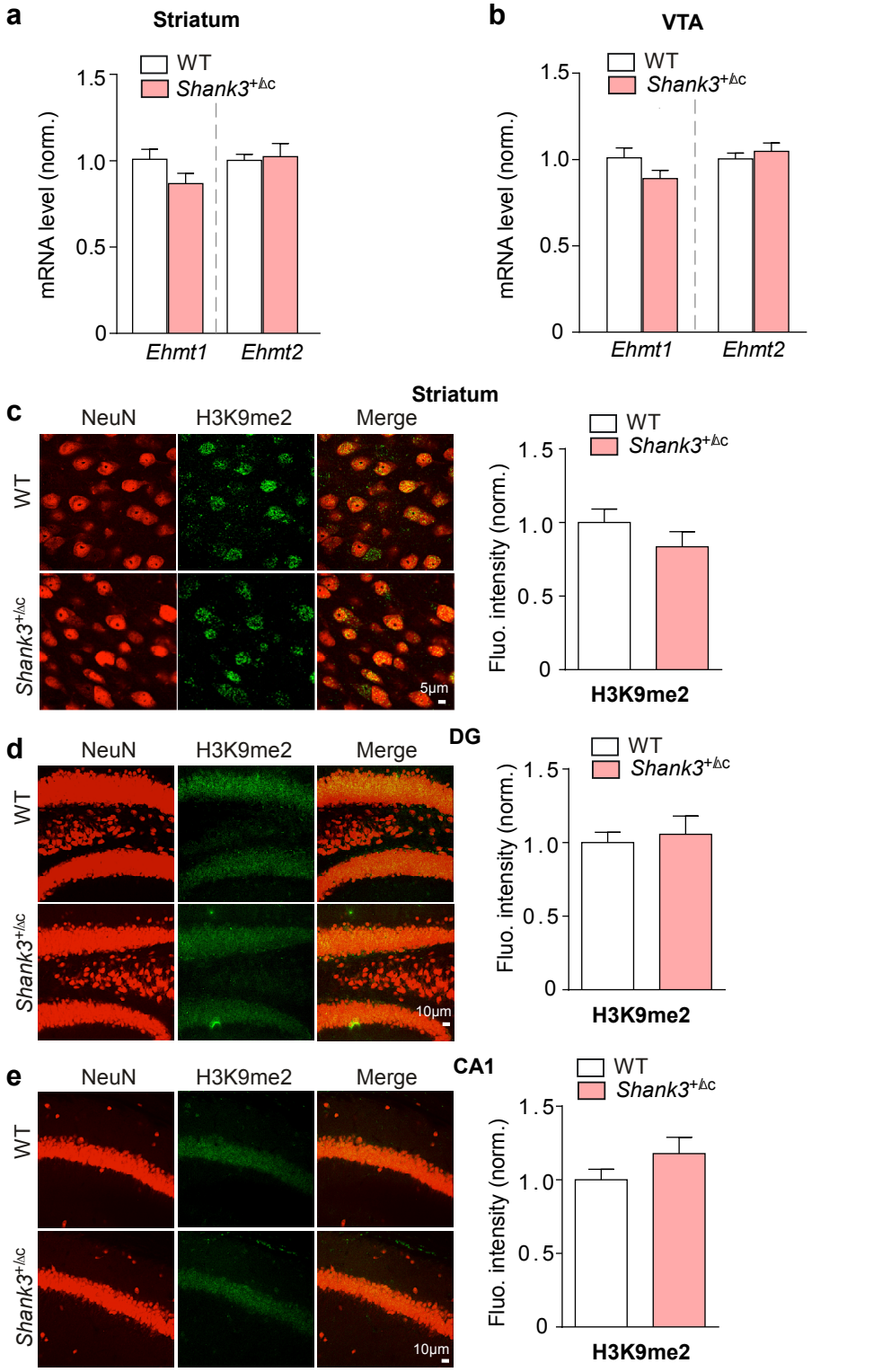
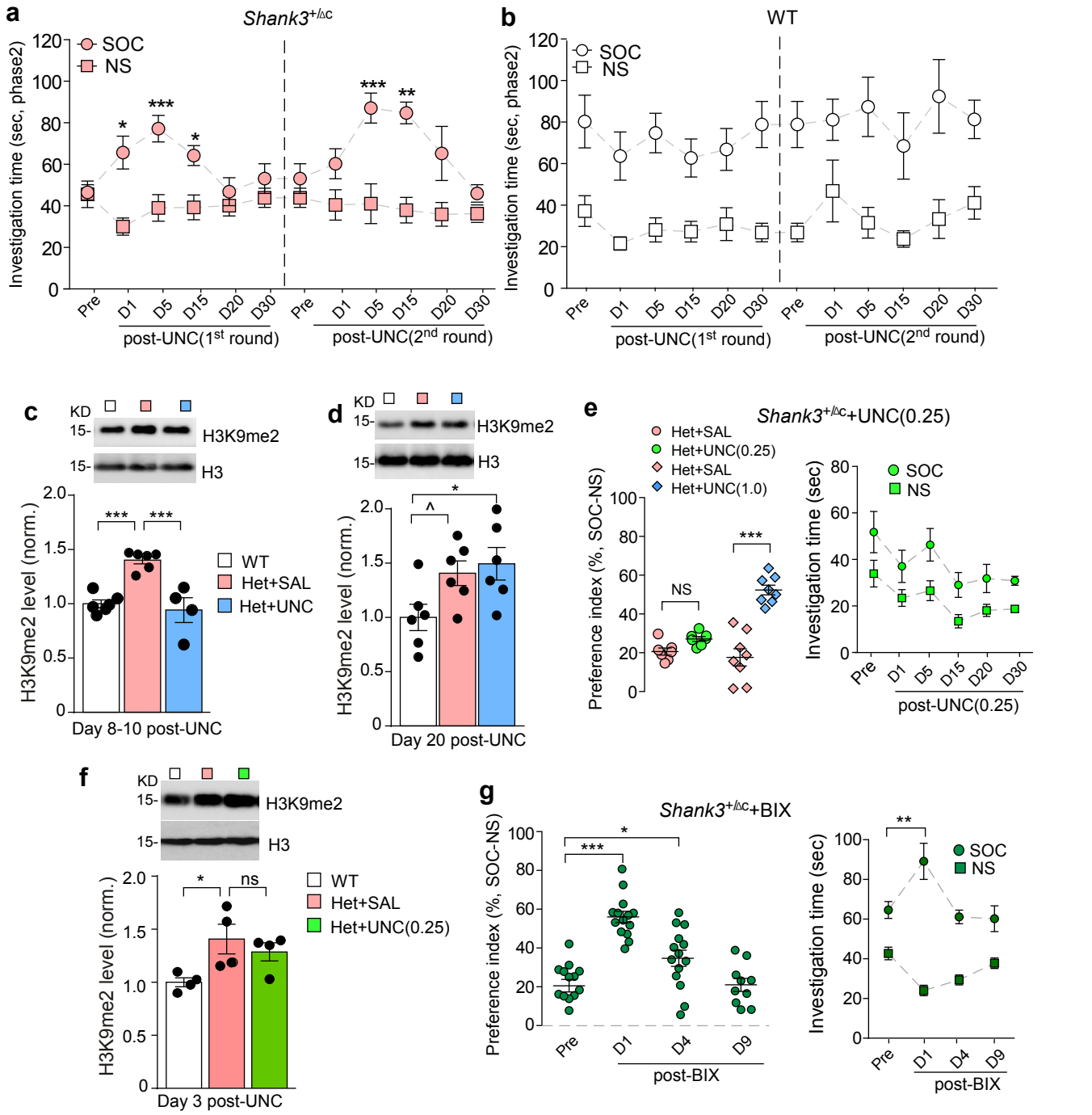
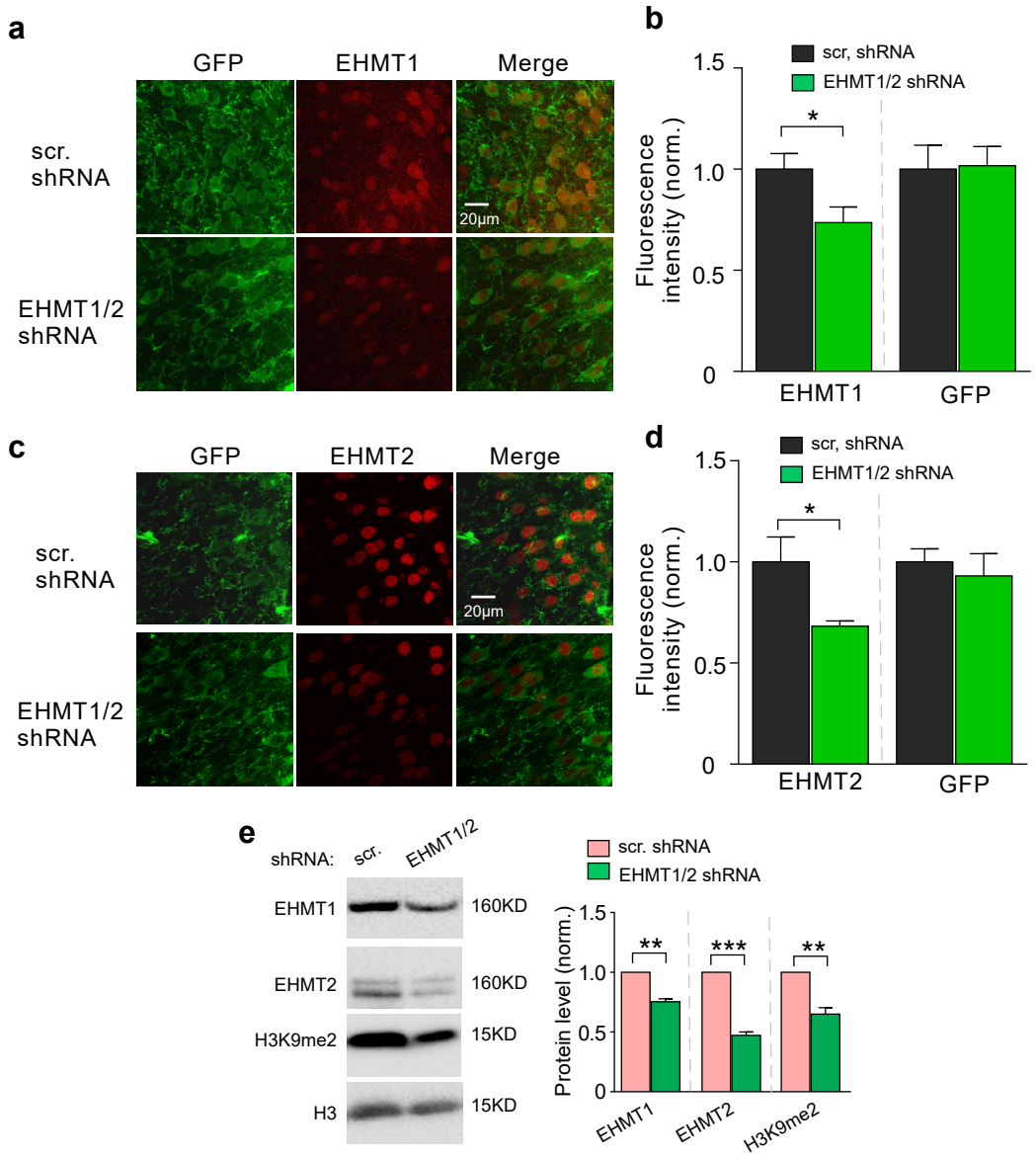


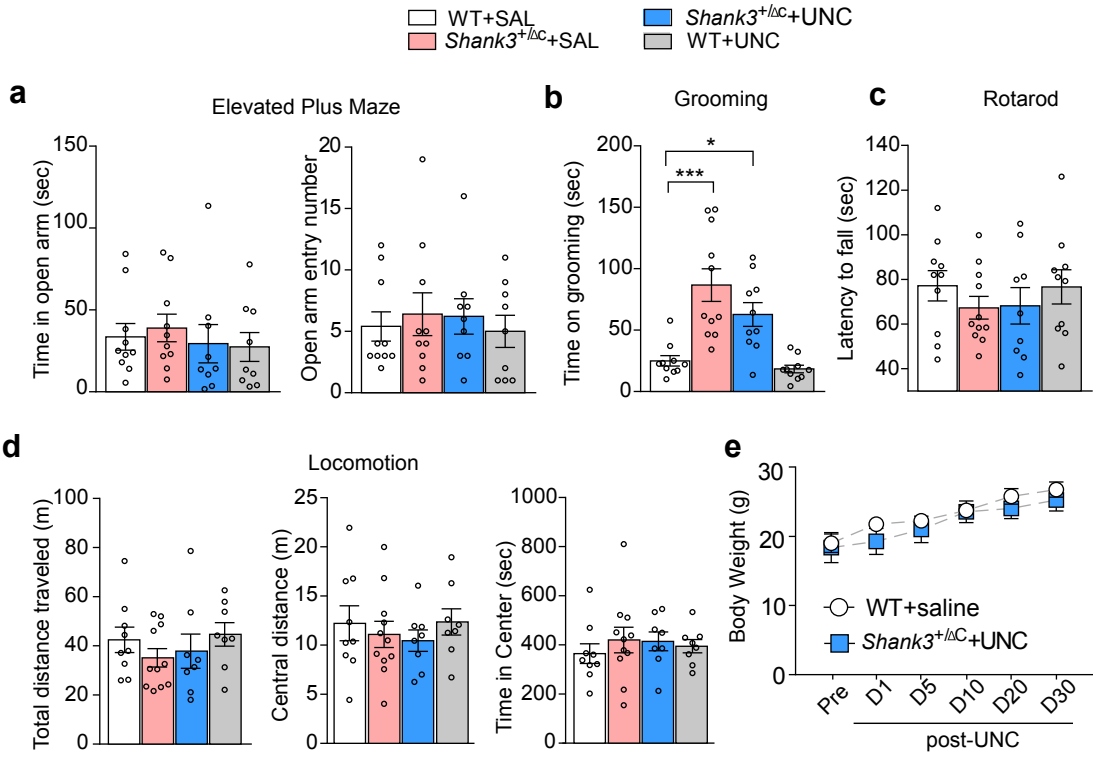
Supplementary Fig. 1. *Ehmt1* and *Ehmt2* transcirption as well as H3K9me2 levels are not changed in other brain regions of *Shank3*-deficient mice. (a, b) Quantitative real-time RT-PCR data on the mRNA level of *Ehmt1* and *Ehmt2* in the striatum (a) and VTA (b) of wild-type and *Shank3*$^{+/\Delta c}$ mice. n=5-6/group, unpaired t-test. (**c, d, e**) Representative images and quantitation of H3K9me2 immunofluorescence levels in striatum (c), dentate gyrus (d) and CA1 (e) regions of wild-type and *Shank3*$^{+/\Delta c}$ mice. n=15-16 slices from 5 animals/group, unpaired t-test. Data are presented as mean ± sem.



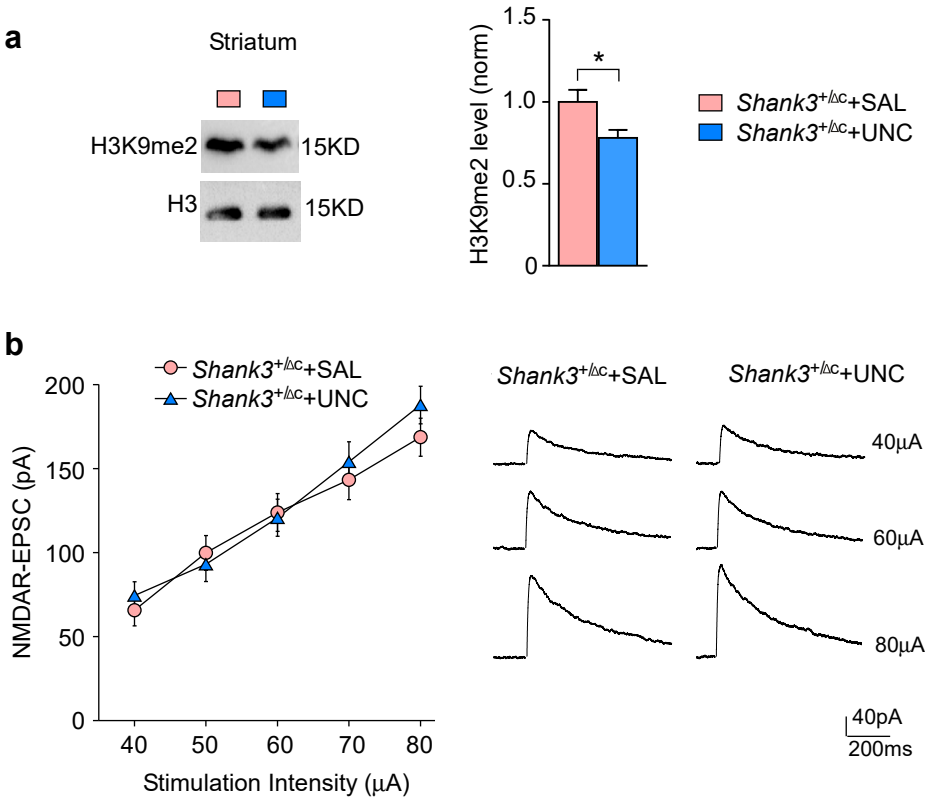
Supplementary Fig. 2. UNC0642 treatment induces the sustained increase of social preference and decrease of H3K9me2 dose-dependently in *Shank3*-deficient mice, while BIX01294 only has a transient effect. (a, b) Plots showing the time spent investigating either the social (SOC) or nonsocial (NS) object during 3-chamber social preference tests of *Shank3*$^{+/\Delta C}$ (a) and WT (b) mice treated with 2 rounds of UNC0642 (UNC, 1 mg/kg, i.p., 3x). n=8-11/group. * P<0.05, *** P<0.001, post- vs. pre treatment, two-way ANOVA. **(c, d)** Quantitation and representative immunoblots of H3K9me2 protein levels in the nuclear fraction of PFC from WT and *Shank3*$^{+/\Delta C}$ mice sacrificed 8-10 days (c) or 20 days (d) after saline or UNC0642 (UNC, 1 mg/kg, I.P., 3x) treatment. n=4-6/group. *** P<0.001, * P<0.05, ^ P<0.1, one-way ANOVA. **(e)** Plots of social preference index and invesigation time on SOC and NS objects in *Shank3*$^{+/\Delta C}$ mice treated with different doses of UNC0642 (1 or 0.25 mg/kg, i.p., 3x). n= 7-8/ group. *** P<0.001, t-test. **(f)** Quantitation and representative immunoblots of H3K9me2 protein levels in the nuclear fraction of PFC from WT and *Shank3*$^{+/\Delta C}$ mice treated with saline or a low dose UNC0642 (UNC, 0.25 mg/kg, i.p., 3x). n= 4/group. *P<0.05, one-way ANOVA. **(g)** Plots of social preference index and invesigation time on SOC and NS objects in *Shank3*$^{+/\Delta C}$ mice treated with BIX01294 (BIX, 1 mg/kg, i.p., 3x). n=10-14, * P<0.05, ** P<0.01,*** P<0.001, post- vs. pre treatment, two-way ANOVA. Data are presented as mean ± sem.



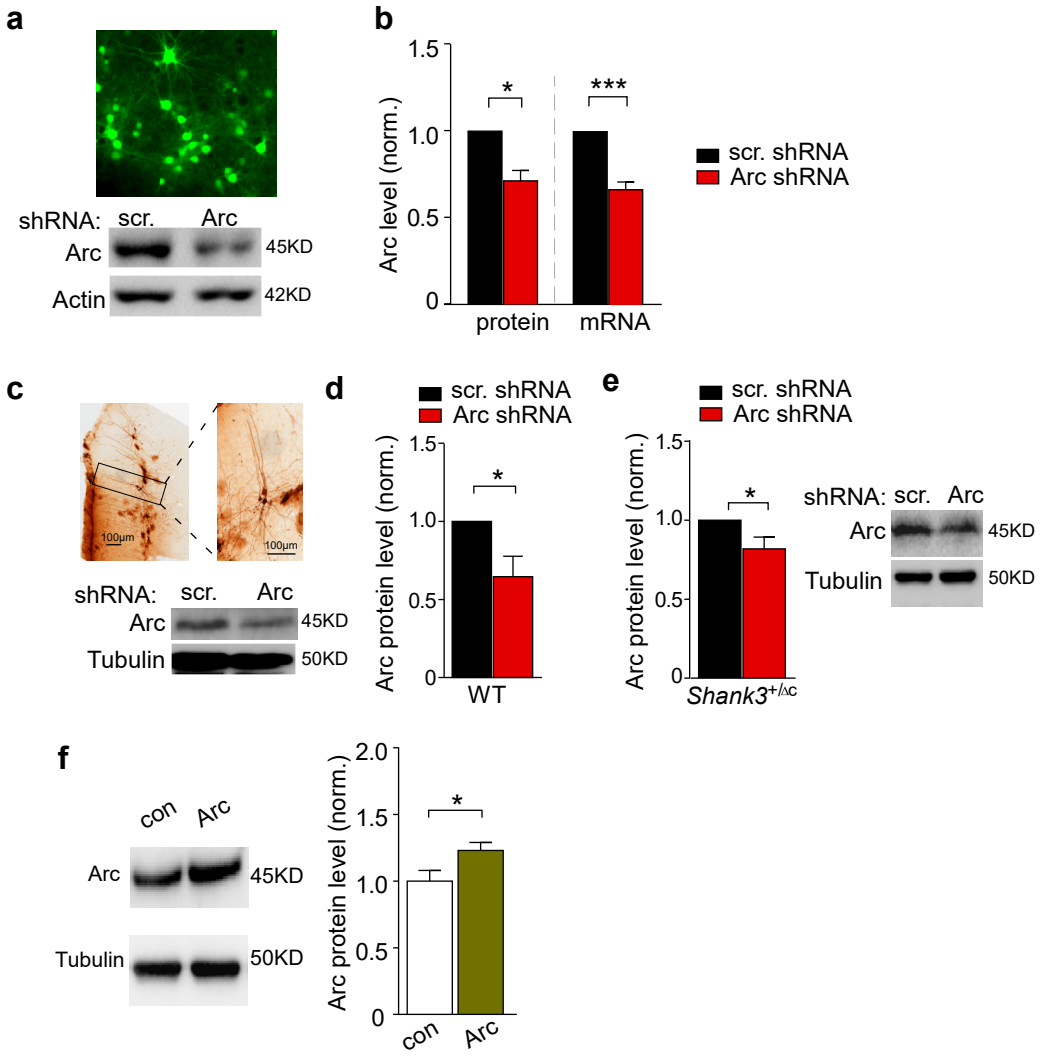
Supplementary Fig. 3. EHMT1 and EHMT2 *in vivo* knockdown. (a-d) Representative images and quantitation of EHMT1(a, b) or EHMT2 (c, d) immunofluorescent signals in PFC from WT mice injected with a scrambled shRNA or EHMT1/2 shRNA lentivirus (mix of EHMT1 and EHMT2 viruses) into the PFC region. n=8-10 slices from 3 animals/group. **P*<0.05, unpaired t-test.(**e**) Immunoblots and quantitation of EHMT1, EHMT2 and H3K9me2 in PFC from *Shank3*$^{+/\Delta C}$ mice injected with a scrambled or EHMT1/2 shRNA lentivirus into the PFC region. n=4/group. ***P*<0.01, ****P*<0.001, unpaired t-test. Data are presented as mean ± sem.



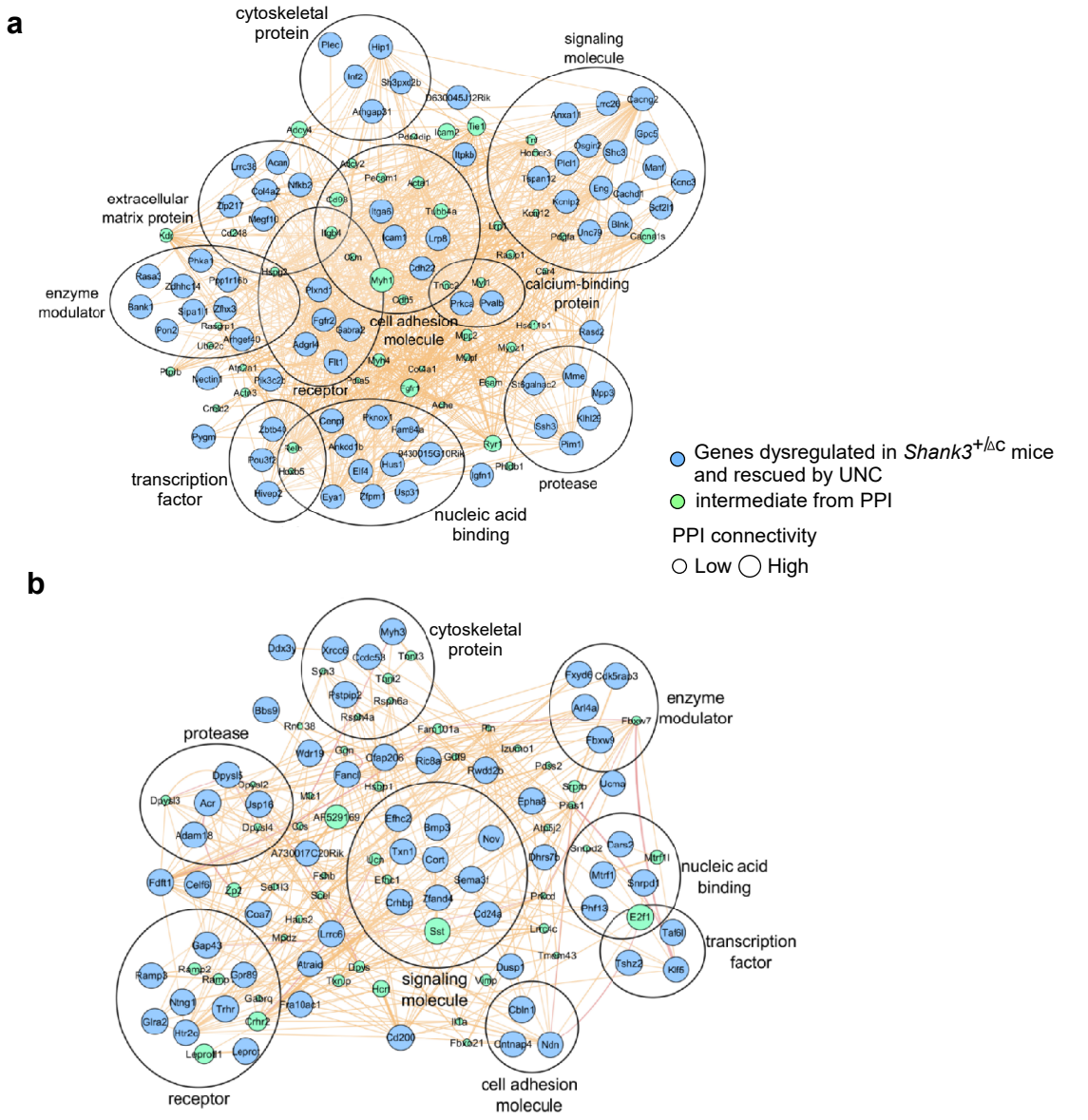
Supplementary Fig. 4. UNC0642 treatment does not alter a variety of behaviors or affect general health in *Shank3*-deficient mice. (a-d) Bar graphs showing a variety of behaviors in WT or *Shank3*$^{+/\Delta C}$ mice treated with saline or UNC0642 (1 mg/kg, i.p., 3x), including elevated plus maze tests (**a**, *n* = 9-10/group), self-grooming (b, *n* = 10-11/group), rotarod tests (**c**, *n* = 9-11/group), locomotion tests (**d**, *n* = 8-11/group). * *P*<0.05, *** *P*<0.001, two-way ANOVA. **(e)** Plot of body weight at different days in UNC0642-treated *Shank3*$^{+/\Delta C}$ and saline-treated WT mice. *n* = 4/group.Data are presented as mean ± sem.



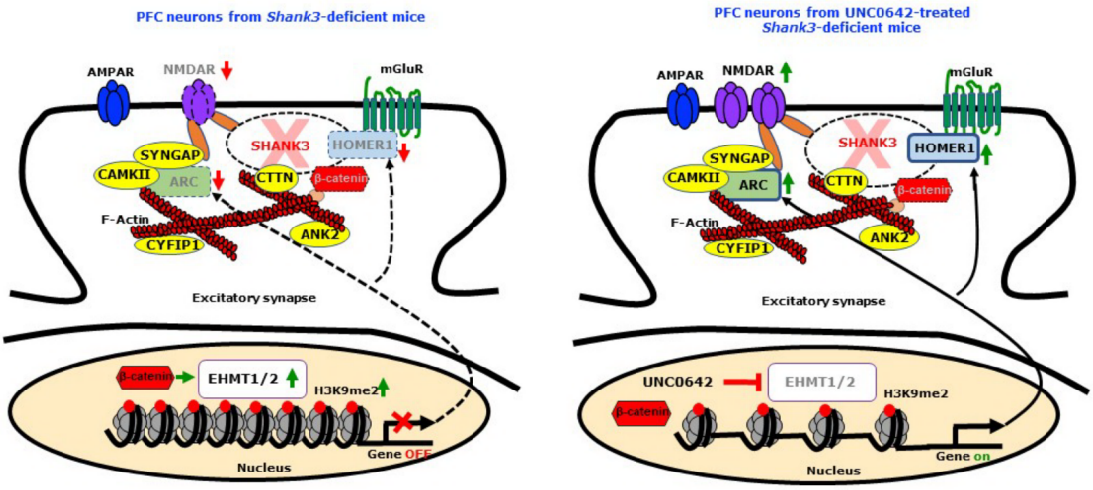
Supplementary Fig. 5. UNC0642 treatment does not affect NMDAR function in the striatum of *Shank3*$^{+/\Delta C}$ mice. (a) Representative images of immunoblots and bar graph showing H3K9me2 protein level in dorsal striatum of Shank3$^{+/\Delta C}$ mice treated with saline or UNC0642 (1 mg/kg, i.p., 3x) (n = 4/group, * $P < 0.05$, unpaired *t*-test). **(b)** Input-output curves of NMDAR-EPSC in dorsal striatal medium spiny neurons from Shank3$^{+/\Delta C}$ mice treated with saline or UNC0642 (1 mg/kg, i.p., 3x) (n = 8-9 cells/2 mice each group, $P > 0.05$, two-way rmANOVA). Data are presented as mean ± sem.



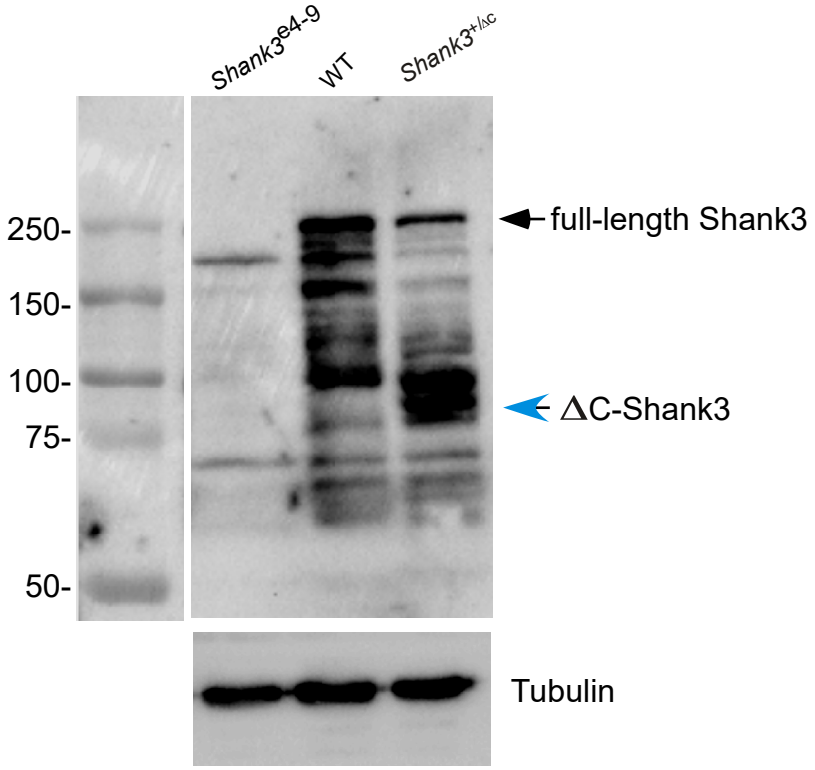
Supplementary Fig. 6. ***In vitro*** **and** ***in vivo*** **Arc knockdown. (a)** Representative images of viral infection (GFP+) and immunoblots of Arc in primary neuronal cultures treated with Arc shRNA or a scrambled shRNA lentivirus. **(b)** Bar graph showing the Arc protein level and *Arc* mRNA level in primary neuronal cultures with different viral infections. n=4 repeats/group. ***P<0.001, *P<0.05, unpaired t-test. **(c)** Representative images of viral infection and immunoblots of Arc in PFC slices from WT mice injected with Arc shRNA or a scrambled shRNA lentivirus into the PFC region. **(d, e)** Bar graphs showing the Arc protein level in PFC slices from WT (d, n=4/group) or *Shank3*$^{+/\Delta C}$ mice (e, n=5/group) with different viral injections. *P<0.05, unpaired t-test. Inset (e): Representative immunoblots. **(f)** Representative immunoblots and bar graphs showing the Arc protein level in PFC slices from *Shank3*$^{+/\Delta C}$ mice injected with Arc lentiviral activation paticles or a control virus (n=7/group). *P<0.05, unpaired t-test. Data are presented as mean ± sem.



Supplementary Fig. 7. Network for genes rescued by UNC0642 in *Shank3*-deficient mice. Protein-protein interaction network created by seeding genes downregulated in *Shank3*-deficient mice and rescued by UNC0642 treatment **(a)**, as well as genes upregulated in *Shank3*-deficient mice and reversed by UNC0642 **(b).** Protein classification clusters are demarcated with circles. Only proteins showing physical and predicted interactions are included. **(c)** qPCR data showing the mRNA level of genes selected from top lists of RNAseq results in PFC from WT or *Shank3*$^{+/\Delta C}$ mice treated with UNC0642 (1 mg/kg, i.p., 3x) or saline (SAL). n= 4-5/group, **P<0.01, *P<0.05, one-way ANOVA. Data are presented as mean ± sem.



Supplementary Fig. 8. Schematic diagram illustrating a potential mechanism underlying the therapeutic effect of UNC0642 on social deficits in autism models. In *Shank3*-deficient mice, the upregulation of EHMT1/2 elevates the repressive H3K9me2, leading to the suppression of genes that are critical to the synaptic NMDAR function, such as synaptic plasticity gene *Arc*, mGluR anchor *Homer1*, and actin regulators. Treatment with UNC0642 restores or elevates many of these target genes, which collectively leads to the normalization of NMDAR synaptic function and consequently the rescue of autism-like social deficits.



Supplementary Fig. 9. Verification of Shank3 mutant mouse lines. Western blots showing Shank3 expression in the cortical lysates from heterozygous mice expressing C term-deleted Shank3, *Shank3*$^{+/\Delta C}$, wild-type (WT), and homozygous mice expressing N term-deleted (exon 4-9) Shank3, Shank3^{e4-9}.