



**Figure 4 - Supplement 1: Long-lasting effects of acute social isolation in adolescence**

(A) Experimental design: WT mice were isolated between P28 and P35 and regrouped until P60 or always kept in group. (B) Three-chamber task experimental paradigm. (C) Time in chamber during social preference phase (One-way RM-ANOVA followed by Tukey's multiple comparisons test. Grouped: chamber main effect  $F(1.684, 18.52)=111.4$   $p<0.001$ . Regrouped: chamber main effect  $F(1.390, 5.30)=79.34$ ,  $p<0.001$ .  $n=12$  mice each group). (D) Interaction time with object or social stimulus 1 ( $###$  Grouped: Paired sample t-test  $t(9)=4.941$ ,  $p<0.001$ ;  $##$  Regrouped: Paired sample t-test  $t(9)=4.082$ ,  $p=0.0027$ . Two-way RM-ANOVA, Target main effect  $F(1, 22)=0.3602$   $p=0.5545$ ; House condition main effect  $F(1, 22)=0.8850$   $p=0.3571$ ; Target x House condition  $F(1, 22)=0.3602$   $p=0.5545$ ,  $n=12$  mice each group). (E) Preference index calculated as object interaction time/(object+stimulus1) or stimulus1 interaction time/(object+stimulus1) (One-sample t-test against chance level 0.5;  $###$  Grouped  $t(11)=6.611$   $p<0.001$ ;  $###$  Regrouped  $t(11)=5.698$   $p<0.001$ ). (F) Time in chamber during social novelty phase (One-way RM-ANOVA followed by Tukey's multiple comparisons test. Grouped: chamber main effect  $F(1.416, 15.57)=16.18$ ,  $p<0.001$ . Regrouped: chamber main effect  $F(1.768, 19.450)=15.61$ ,  $p<0.001$ .  $n=12$  mice each group). (G) Interaction time with stimulus 1 (familiar) and stimulus 2 (unfamiliar) ( $###$  Grouped: Paired sample t-test  $t(9)=4.736$ ,  $p<0.001$ ;  $###$  Regrouped: Paired sample t-test  $t(9)=4.939$ ,  $p<0.001$ . Two-way RM-ANOVA. Target main effect  $F(1, 22)=25.71$   $p<0.001$ ; House condition main effect  $F(1, 22)=2.949$   $p=0.1$ ; Target x House condition  $F(1, 22)=1.927$   $p=0.1790$ ,  $n=12$  mice each group). (H) Preference index calculated as stimulus1 interaction time/(stimulus1+stimulus2) or stimulus2 interaction time/(stimulus1+stimulus2) (One-sample t-test against chance level 0.5;  $###$  Grouped  $t(11)=4.694$   $p<0.001$ ;  $##$  Regrouped  $t(11)=3.480$   $p=0.0051$ ). (I) Distance moved during the 3-chamber (Social preference: Mann-Whitney test  $U=30$ ,  $p=0.0145$ . Social novelty: Unpaired samples t-test  $t(18)=2.260$   $p=0.0364$ ,  $n=12$  mice each group). Data are represented as mean $\pm$ SEM.