Effect of chronic social isolation stress after acute traumatic stress on mouse social behavior across the life cycle

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Abstract

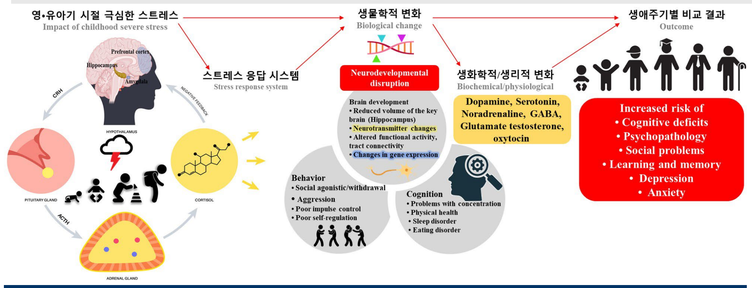
Background: Modern people are exposed to various stressful situations. Stress is a crucial factor for depression and is related to the imbalance of physiological and psychological homeostasis, including brain function and structure. Stress induces the endocrine alterations characterized by the activation of hypothalamic-pituitary-adrenal axis and sympathetic adrenal-medullary system. There are many causes of stress, and each affects us differently. However, it remains unknown how transcriptomic alteration induced by long-term social isolation stress are occurred in the hippocampus regions. Therefore, we used next-generation sequencing (NGS) to profile the transcriptomic changes in mice which could induce behavior changes after long-term social isolation stress. Therefore, we focused on the effect of long-term social isolation stress on mice behavior in each life cycle model by age. Interestingly, we observed different behavioral phenotype in childhood stage compared with early adulthood and adulthood stage.

Methods: We focused on the effect of long-term social isolation stress on mice behavior in each life cycle model by age**.** C57Bl/6J male mice of Childhood, early adulthood, and adulthood were performed a behavioral test after 5 weeks of social isolation after 1 week of severe acute stress. we observed that 5 weeks of social isolation led to different behavioral phenotype in childhood compared with the early adulthood and adulthood stages.

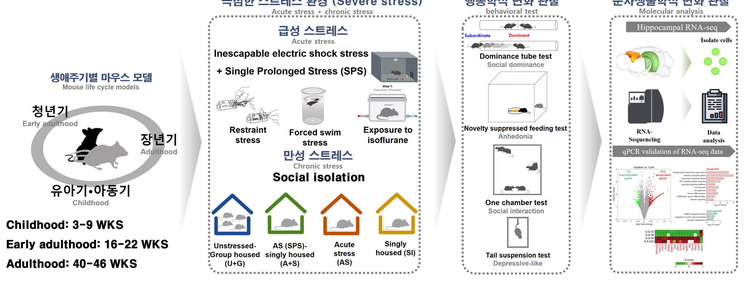
Results: We observed different behavioral phenotype in childhood stage compared with early adulthood and adulthood stage. Long-term social isolation-induced childhood male mice showed high social dominance in the tube dominance test and increased serum CORT levels. The Gene ontology (GO) analysis of Differential Express Genes (DEGs) with significant differences revealed that they are involved in critical biological processes and molecular pathways, such as neuron differentiation, neurogenesis, neuron development, learning or memory, brain development, and neuroactive ligand-receptor interaction. Finally, DEGs analysis revealed that 4 candidate genes were up-regulated, and 4 down-regulated genes. The differential expression of the selected genes were verified using by qRT–PCR and immunohistochemistry (IHC). Therefore, we identified the effect of social isolation on the development of sociability through age by transcriptome analysis that were associated with social behavior, including FABP7 and Cxcl12.

Conclusions: Through this study, we can find the critical factor involved in the sociability that might be utilized to the diagnosis indicators or therapeutic agents for stress-related diseases.

Background



Material and methods



Results

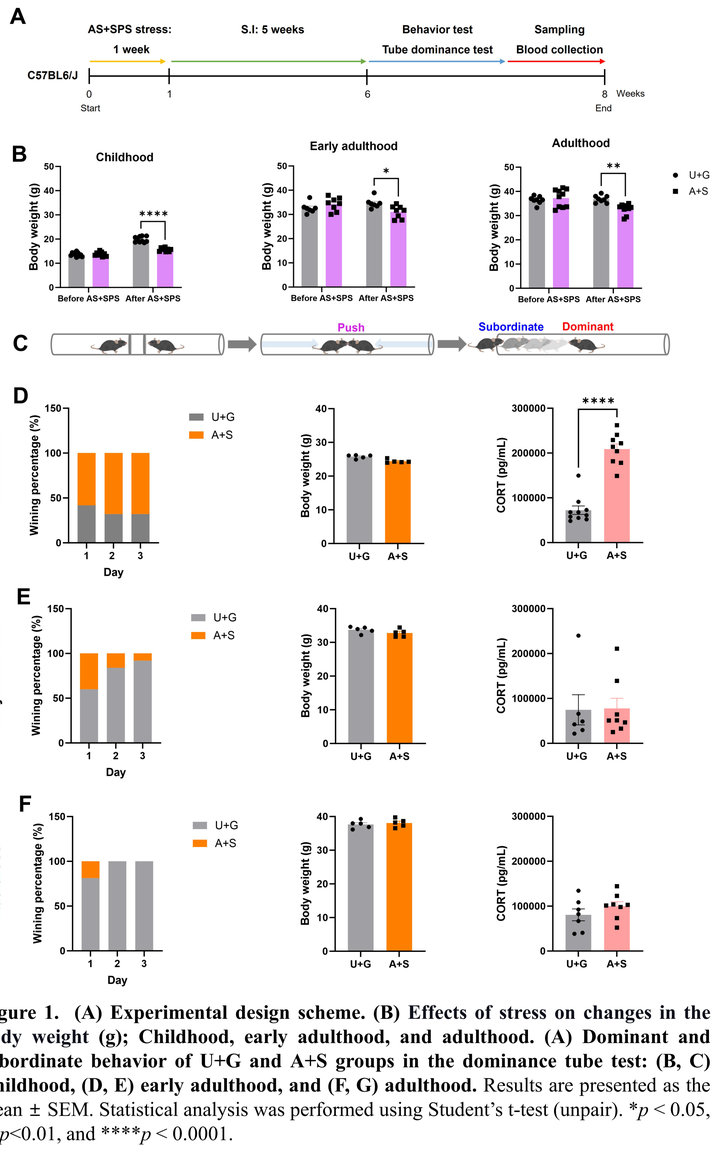


Figure 1. (A) Experimental design scheme. (B) Effects of stress on changes in the body weight (g); Childhood, early adulthood, and adulthood. (A) Dominant and subordinate behavior of U+G and A+S groups in the dominance tube test: (B, C) Childhood, (D, E) early adulthood, and (F, G) adulthood. Results are presented as the mean ± SEM. Statistical analysis was performed using Student's t-test (unpair). *$p < 0.05$, **$p<0.01$, and ****$p < 0.0001$.

Results

Figure 3. Screening of differentially expressed genes (DEGs) and analyzing for their function. (A, B) Transcriptomic analysis, principal component analysis (PCA). (C-F) Volcano plot of differentially expressed genes. (G, I) Pattern analysis of DEGs. (H) KEGG pathway of enrichment analysis. (J) Signaling pathways represented by the DEGs.

Results

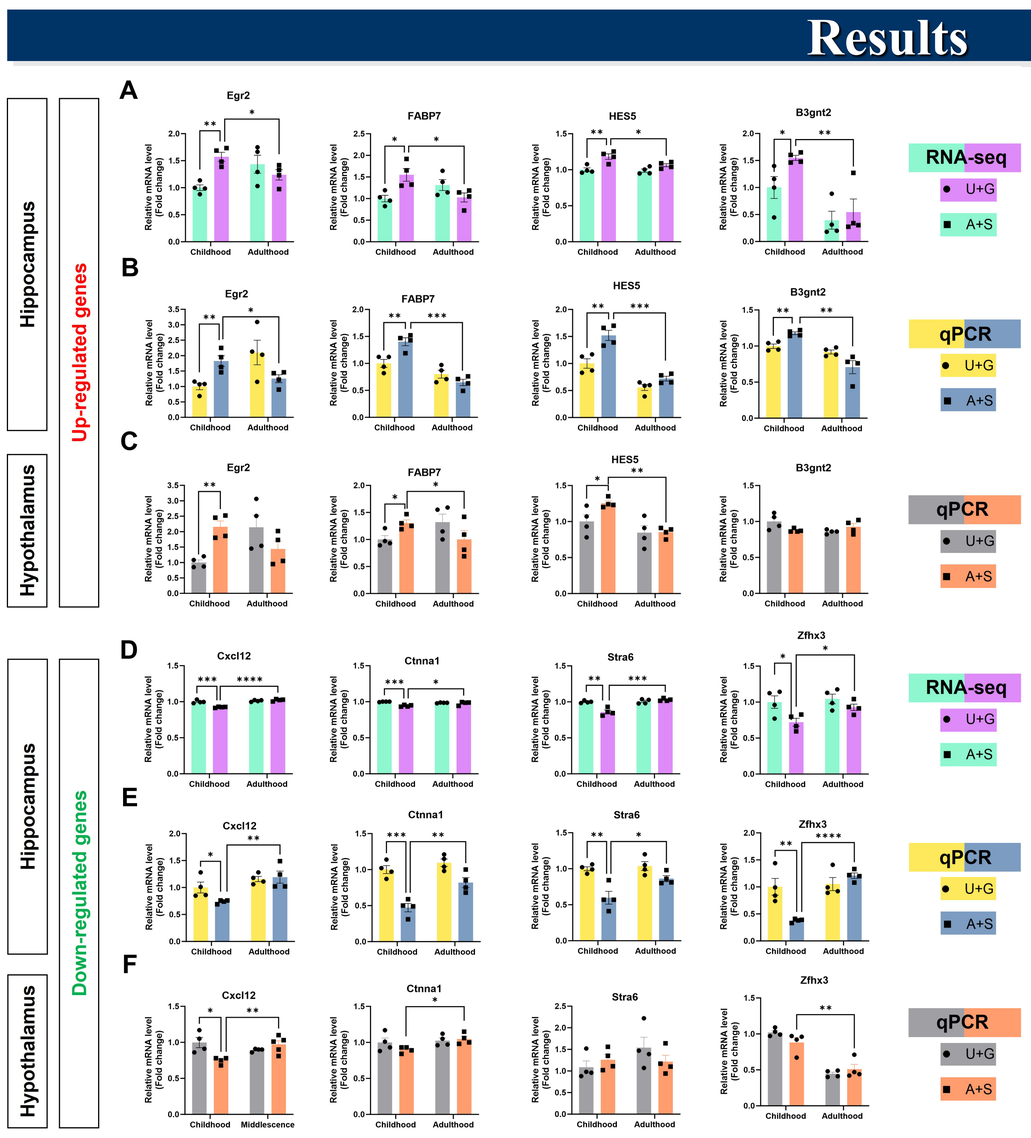


Figure 4. Validation of differentially expressed eight candidate genes using qPCR. (A-C) Up-regulated genes and (D-F) down-regulated genes in the hippocampus and hypothalamus. Results are presented as the mean ± SEM. Statistical analysis was performed using Two-way ANOVA, *p<0.05, **p<0.01, ***p<0.001, ****p<0.0001.

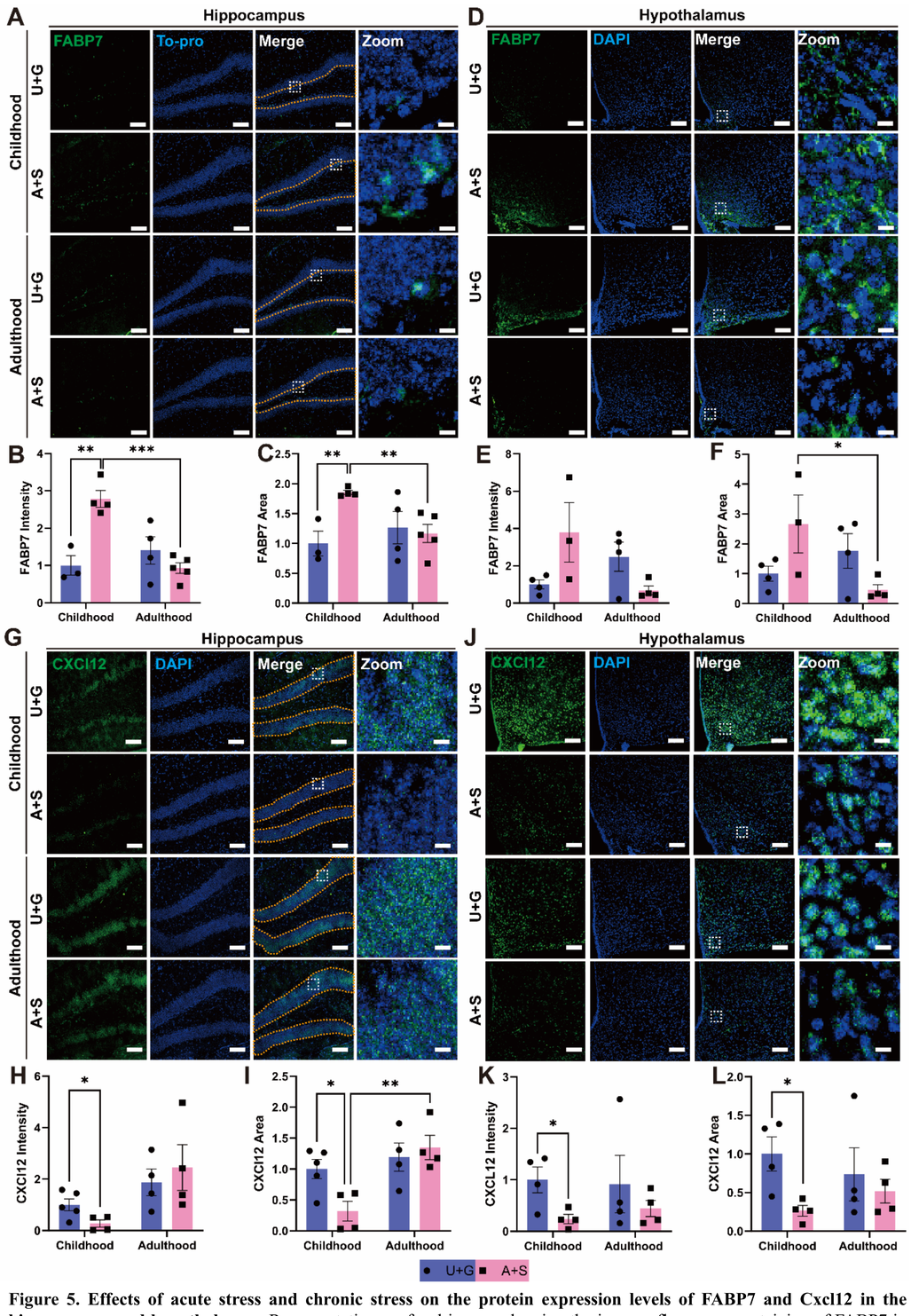


Figure 5. Effects of acute stress and chronic stress on the protein expression levels of FABP7 and Cxcl12 in the hippocampus and hypothalamus. Representative confocal images showing the immunofluorescence staining of FABP7 in the hippocampus (A-C) and hypothalamus (D-F). Representative confocal images showing the immunofluorescence staining of Cxcl12 in the hippocampus (G-I) and hypothalamus (J-L). Results are presented as the mean ± SEM. Statistical analysis was performed using an unpair t-test. $p < 0.05$, **$p < 0.01$, ***$p < 0.001$ versus U+G group or childhood versus adulthood between A+S group.

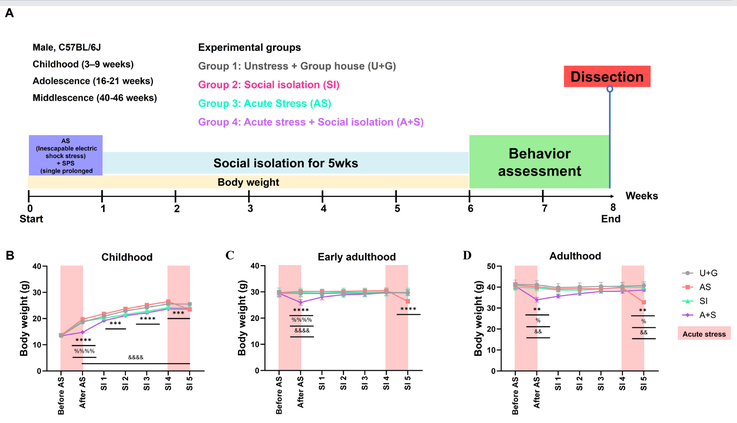


Figure 6. (A) Experimental scheme. (B-D) Changes in body weight (g): Childhood, early adulthood, and adulthood. Results are presented as the mean ± SEM. Statistical analysis was performed using Two-way ANOVA. U+G vs A+S; **p<0.01 ****p<0.0001, AS vs A+S; %p<0.05, %%%%p<0.0001, and SI vs A+S; &&p<0.01, &&&& p<0.0001. U+G or AS or SI vs A+S; **p<0.01, %p<0.05, and &&p<0.01.

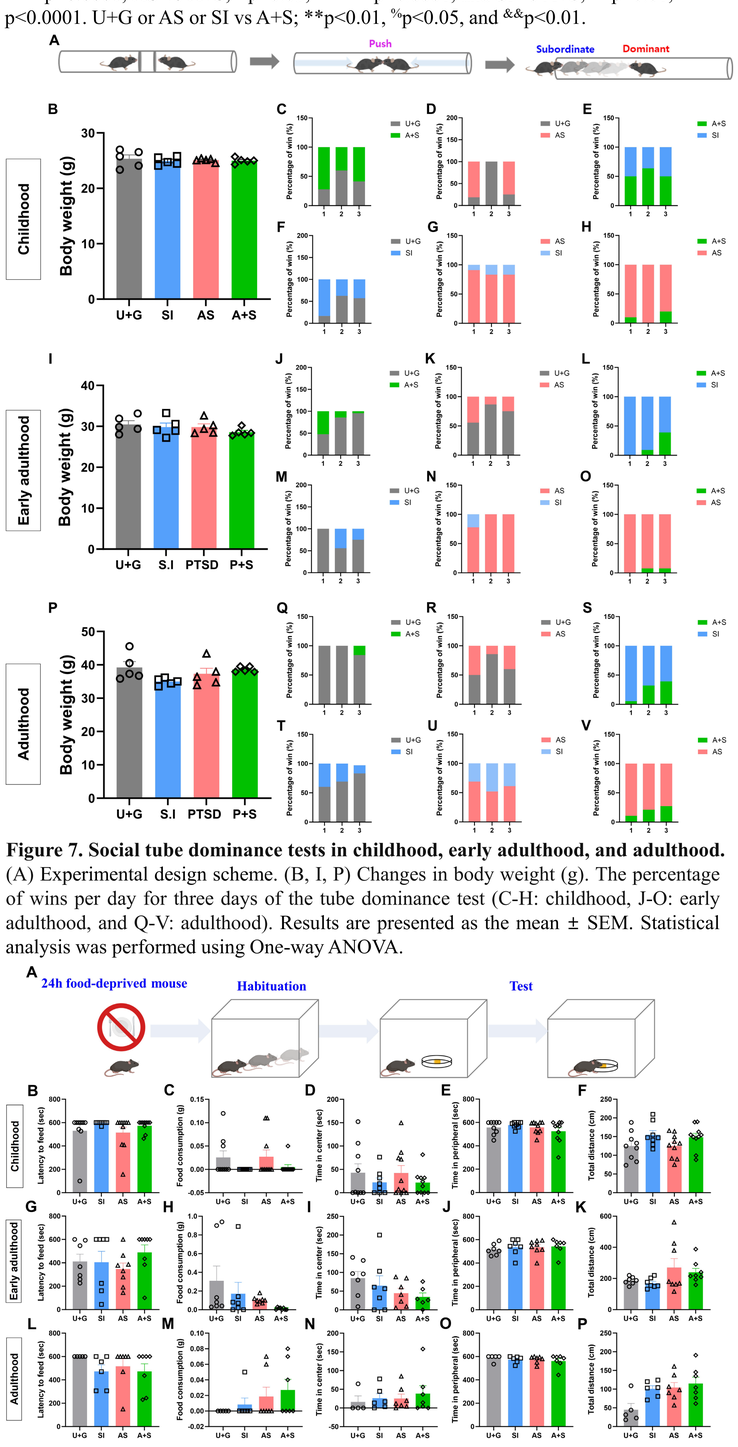


Figure 7. Social tube dominance tests in childhood, early adulthood, and adulthood. (A) Experimental design scheme. (B, I, P) Changes in body weight (g). The percentage of wins per day for three days of the tube dominance test (C-H: childhood, J-O: early adulthood, and Q-V: adulthood). Results are presented as the mean ± SEM. Statistical analysis was performed using One-way ANOVA.

Figure 8. Novelty suppressed feeding test (NSFT). (A) Experimental design scheme. NSFT monitored for 10 min. Latency to feed (B, G, F). Food consumption (C, H, M). Time in chamber (D, I, N). Time in peripheral (E, J, O). Total distance (F, K, P) in the childhood (B-F), early adulthood (G-K), and adulthood (L-P). Results are presented as the mean ± SEM. Statistical analysis was performed using One-way ANOVA, *p<0.05.

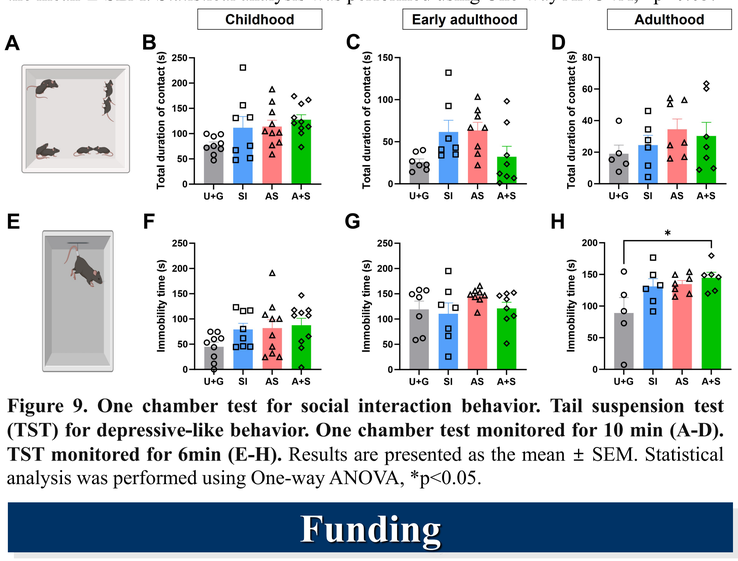


Figure 9. One chamber test for social interaction behavior. Tail suspension test (TST) for depressive-like behavior. One chamber test monitored for 10 min (A-D). TST monitored for 6min (E-H). Results are presented as the mean ± SEM. Statistical analysis was performed using One-way ANOVA, *p<0.05.

Funding

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