Effects of fluoxetine and environmental enrichment on chronic defeat stress-related behaviors & gene expression in the amygdala.

Zachary A. Cordner, Isaiah Thomas, Gretha J. Boersma,
Richard S. Lee, James B. Potash & Kellie L. Tamashiro

Department of Psychiatry & Behavioral Sciences,
The Johns Hopkins University School of Medicine

JOHNS HOPKINS MEDICINE
SCHOOL OF MEDICINE

Background

The relationship between stress and affective disorders is well known and, collectively, stress-related disorders are among the most burdensome of all diseases. So, understanding mechanisms and developing treatment approaches are urgent priorities.

Historically, our understanding of affective disorders has been reduced to two prevailing hypothesis about pathogenesis. Namely, disruption of monoamine balance and disruption of neurogenesis. Explaining the action of existing therapies and the development of new treatments has remained mostly anchored to these hypothesis, and the monoamine hypothesis has dominated.

However, epidemiology, brain imaging, genetics, and the recent success of novel therapies point to more complex explanations for disease and treatment response.

Objectives

In this study, we sought to determine:

1. The long-term effects of chronic defeat stress on mood-like and anxiety-like behavior in a wild type mouse model,
2. The effect of two different treatments- environmental enrichment and fluoxetine- on the chronic stress induced phenotype, and
3. Changes in gene expression in the amygdala in response to chronic stress and treatment with enrichment or fluoxetine.

Methods

Adult male C57BL/6J mice were exposed to 14 days social defeat stress (SD) or control conditions (Ctrl) followed by 28 days of treatment or control conditions. Treatment consisted of either environmental enrichment (EE) or subcutaneous implantation of a sustained release fluoxetine pellet (fluox). Behaviors in the open field, elevated plus maze, sucrose preference, and forced swim tests were assessed between treatment day 23-28. Brain and blood tissues were collected after 28 days of treatment. Basal corticosterone (cort) level was measured in plasma. Whole amygdala was dissected for gene expression. Expression levels of 168 genes related to synaptic function/plasticity or oxidative stress were initially assessed. The top 10 differentially expressed genes (DEGs), ranked by p-value, were validated by routine qRT-PCR.

All protocols were approved by the Animal Care and Use Committee at the Johns Hopkins University School of Medicine.

Results

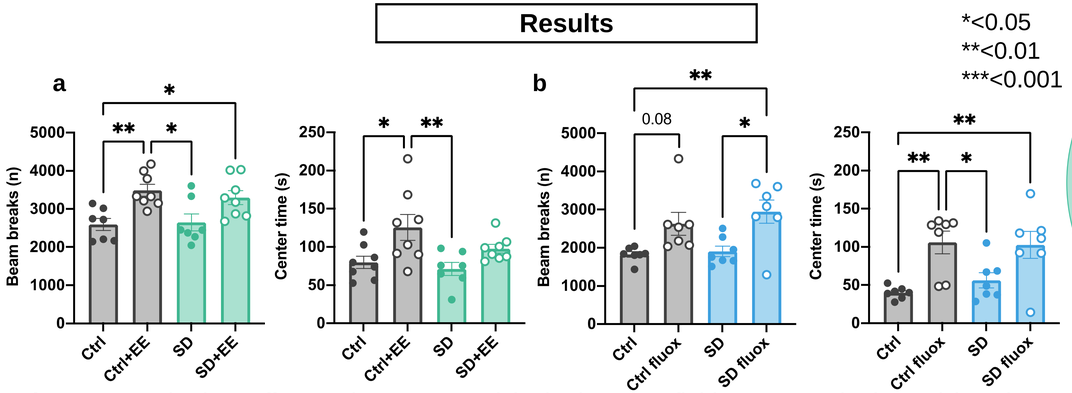

Figure 1. SD had no effect on locomotor activity in the open field. However, both EE (a) and fluox (b) increased locomotor activity.

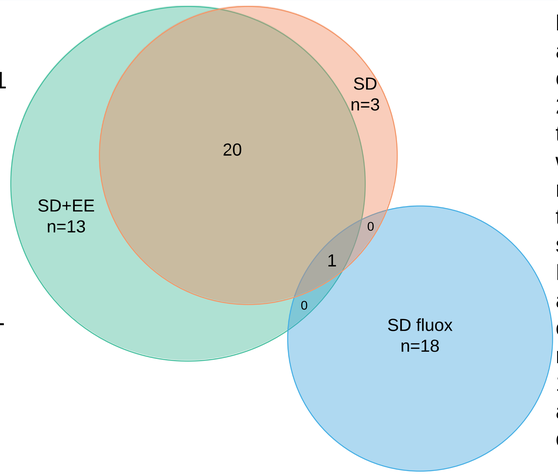

Figure 3. SD was associated with a set of 24 DEGs. Of those, 23 were normalized by treatment with fluox, while only 3 were normalized by treatment with EE. A separate set of 13 DEGs was found among SD+EE mice compared to Ctrl. A non-overlapping set of 18 DEGs was found among SD fluox mice compared to Ctrl.

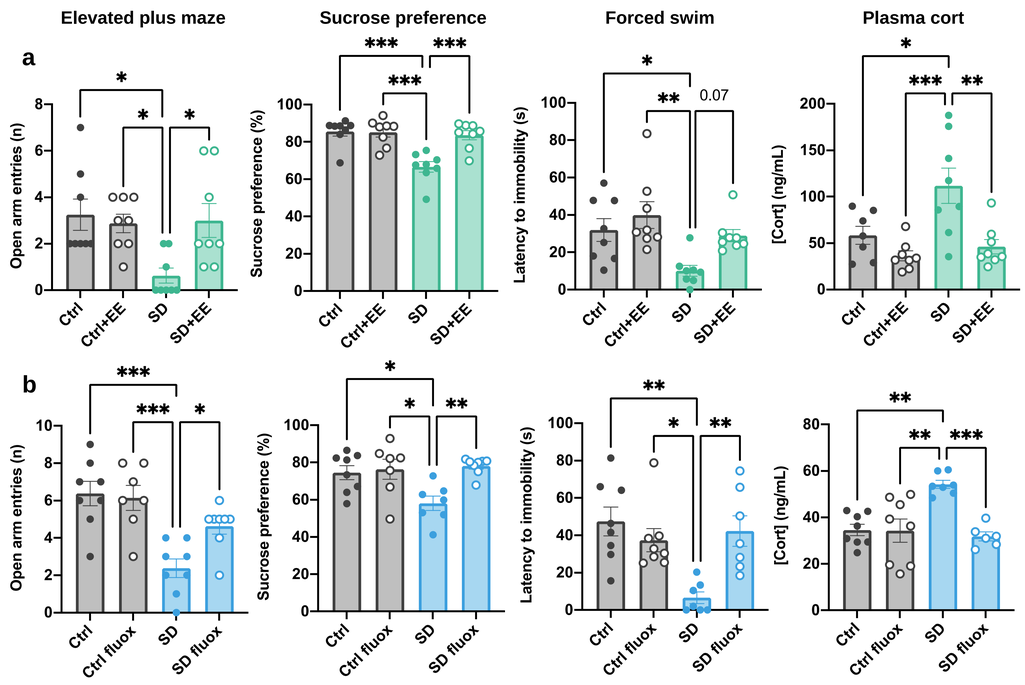

Figure 2. SD resulted in long-lasting behavioral deficits characterized by reduced exploration of the elevated plus maze, reduced sucrose preference, and reduced latency to immobility in the forced swim test. This was associated with long-lasting elevation in basal plasma cort. Treatment with EE (a) or fluox (b) led to similar normalization of the phenotype.

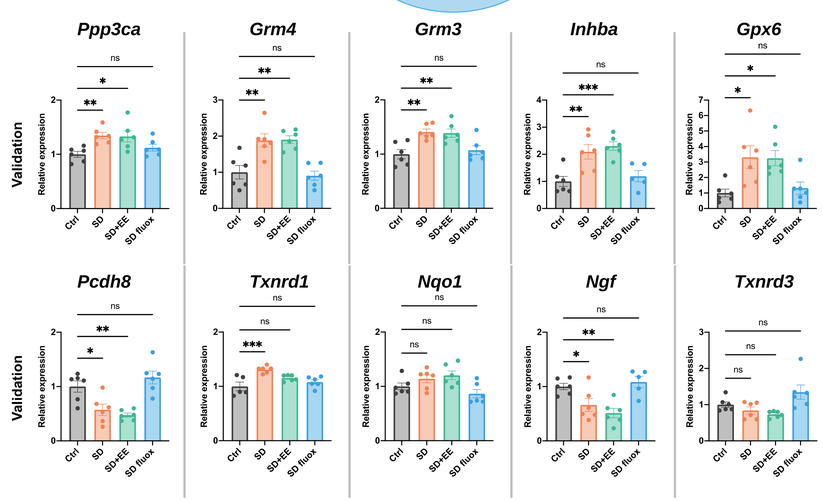

Figure 4. Of the top 10 differentially expressed genes ranked by p-value, 8 were validated - *Ppp3ca*, *Grm4*, *Grm3*, *Inhba*, *Gpx6*, *Pcdh8*, *Txnrd1,* and *Ngf*. Only *Nqo1* and *Txnrd3* failed to replicate. Broadly, validation re-demonstrated the pattern of fluox, but not EE, normalizing stress-related gene expression changes in the amygdala.

Conclusions

SD leads to a long-lasting depression- and anxiety-like phenotype. EE and fluox result in similar recovery from the stress-induced phenotype, but lead to markedly different patterns of gene expression in the amygdala. This may provide novel insights into mechanisms of both disease and treatment.