Behavioral Characterization and Pharmacological Validation of Chronic Social Defeat Stress Mouse Model

I. Morganstern[1], S. Davis[1], K. Homa[1], E. Sabath[1], R.C. Bagot[2], V. Alexandrov[1], E.J. Nestler[2], T. Hanania[1]

[1]PsychoGenics Inc., Tarrytown, NY, USA

[2]Neuroscience Department, Mount Sinai Icahn School of Medicine, New York, NY, USA

PsychoGenics — Redefining Drug Discovery Through Innovation

INTRODUCTION

The identification of newer and clinically relevant animal models in the field of mood disorders is a key component to efficient drug discovery. The Chronic Social Defeat Stress (CSDS) mouse model as developed and described by Dr. Eric Nestler and colleagues has received much interest recently as it closely mimics the dynamic range of individual responses to stressors such as the development of a major depressive disorder, anxiety, post-traumatic stress disorder or resiliency to these disease states. Such individual responses to social stressors are particularly useful in modeling aspects of depression- and anxiety-like behavior with high construct, face, discriminative and predictive validity. Therefore, our goal here using this multifaceted model was three-fold 1) to confirm and further characterize the distinct behavioral traits in animals most susceptible to social stress after going through the 10-day social defeat paradigm compared to undefeated control animals; 2) to provide pharmacological validation for this model using standard antidepressant medications; and 3) to confirm the utility of this model in combination with our more complex proprietary algorithm-based behavioral platforms such as the SmartCube ® System. The social defeat model was set up in collaboration with Dr. Eric Nestler's laboratory at Mount Sinai Icahn School of Medicine, specifically with the help of Dr. Rosemary Bagot (Postdoctoral Fellow, Neuroscience Department).

METHODS

ANIMALS

➢ Male C57Bl6/J mice (7-8 weeks, Jackson Laboratory) were used as "intruder mice" and male CD-1 mice (Charles River Laboratories, retired breeders 4 months age) were used as "aggressor" mice throughout the study. The aggressor mice were pre-screened over 3 days to fit the following criteria: the CD-1 mouse must attack the screener mouse in at least two consecutive screening sessions (3 min each session), and the latency to initial aggression must be less than 60s.

BEHAVIOR

➢**Chronic Social Defeat Stress (CSDS):** The CSDS was performed as previously described (Berton et al., 2006 and Donahue et al., 2014). Briefly, the C57 intruder mice underwent 10 consecutive days of defeats, during which they were placed in the home cage of a resident CD-1 aggressor mouse for 3-5 minutes. After the defeat session, the mice were separated in the cage with a perforated Plexiglas divider which allowed for sensory exposure. The defeated mice were exposed to a new resident and cage on each of the 10 days. Control C57BL/6J mice are also rotated daily in an identical cage set-up, but not allowed physical contact with their cage mates. After the final defeat session, all C57BL/6J mice are single housed and screened using the Social Interaction Test on day 11.

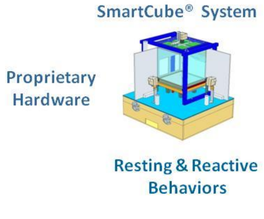
➢**Social Interaction (SI):** SI testing was performed in a custom open field arena (43.2 X 43.2 X 30.5cm) with an automated video tracking program (View Point, Life Sciences) as previously described (Golden et al., 2010; Berton et al., 2006 and Donahue et al., 2014) with minor modifications. Briefly, social approach behavior was evaluated in the presence (phase 2, 2.5 min) and absence (phase 1, 2.5 min) of a novel CD-1 mouse (social target), An SI score of <1 was used as a cutoff for stress-susceptible mice which were used for this study. Control mice were chosen to have a social interaction score>1. ***In our experience, 60-65% of all defeat mice screened are susceptible.*** For the data shown, n=10-15 were used per group.

➢**Fear Conditioning:** During training, mice received 3 presentations of a 0.6mA shock preceded and overlaid by a tone (10s) with a random ITI.. On test day (24 hours later), freezing in response to tone presentation was assessed.

➢**Drug Treatment:** To assess antidepressant effects, control and defeat (susceptible) mice were administered vehicle, Imipramine (20 mg/kg) acute (single) and chronic (2-4 weeks) or Fluoxetine (15 mg/kg) chronic (3 weeks). SI testing occurred 20-22 hours post last dose.

➢**SmartCube® System:** This proprietary platform uses computer vision to automatically capture and score changes in activity, spatial patterns, spontaneous behavior, reactive behavior, gait, and other measures in mice ***(see diagram and video below).***

➢**Feature Analysis:** Data are typically presented by three classes (see Figure 6): Control, Disease (defeat), and Treated. The drug treatment effect can be represented as a combination of two components: one along the direction of the "recovery line" (connecting the centers of the Control and Disease clouds) shown as a blue arrow, and the component orthogonal to ("pointing away" from) that direction shown as a yellow arrow. The relative length of the "recovery" (blue) arrow with respect to the Control-Disease distance can then be interpreted as the "recovery due to the drug", whereas the relative length of the "other effect" (yellow) arrow represents feature changes that move the Treated group away from the Control group. The summary of this analysis can be effectively represented as a bar graph (right pane) which we typically refer to as the recovery signature.



Click to play

RESULTS

Figure 1: Social Avoidance Behavior in Defeat Versus Control Mice in the Social Interaction Test

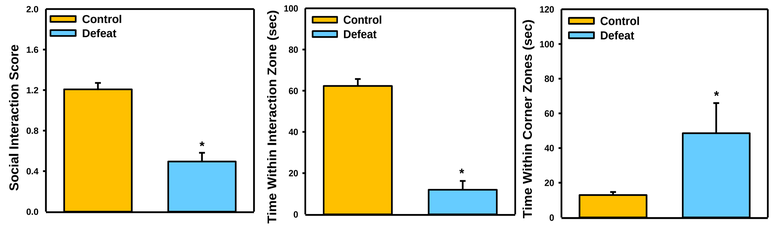


Figure 2: Increased Freezing Behavior During Training and in Response to Sound Cue in Fear Conditioning Test

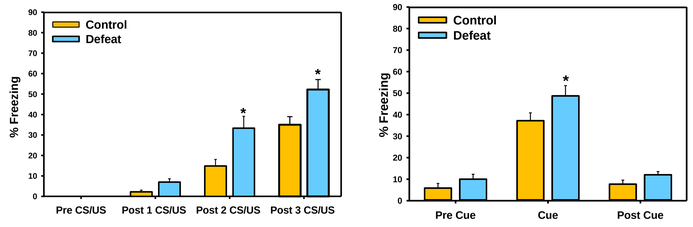


Figure 3: Chronic, but not Acute Treatment with Imipramine (20 mg/kg) Attenuated Social Defeat-Induced Social Avoidance Behavior in the Social Interaction Test

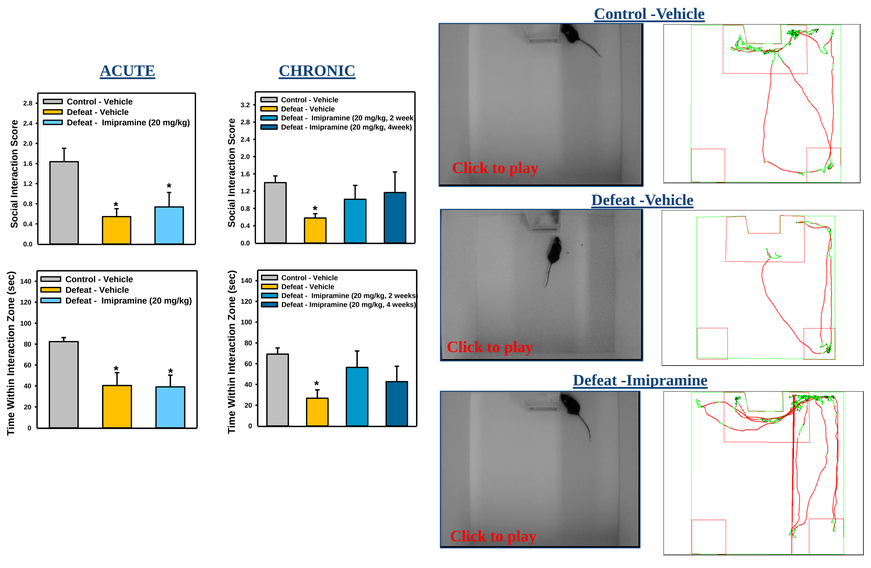


Figure 4: Chronic Fluoxetine (15mg/kg) Attenuated Social Defeat-Induced Social Avoidance in the Social Interaction Test

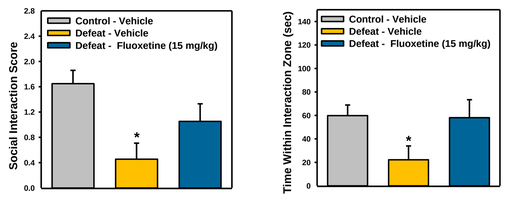


Figure 5: Discrimination Probability of Defeat Versus Control Mice Using SmartCube ® Technology

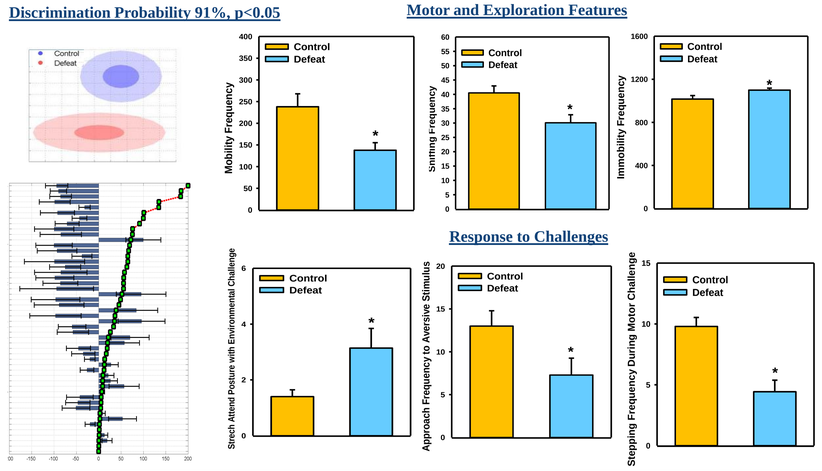
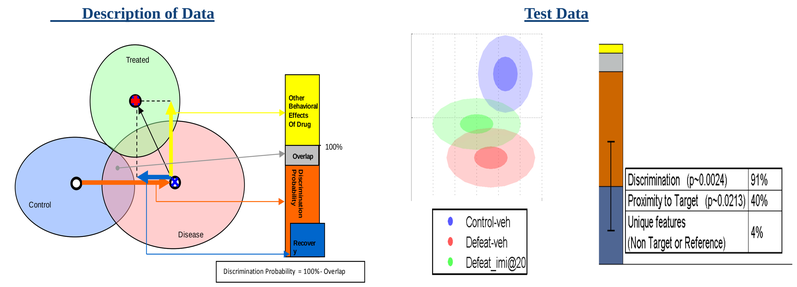


Figure 6: Recovery of Defeat Phenotype with Chronic Imipramine (20 mg/kg) Treatment Using SmartCube ® Technology



SUMMARY

➢ Using the chronic social defeat model, we successfully verified the social avoidance phenotype of stress-susceptible mice and additionally noted enhanced fear-associated learning in these animals compared to control mice.

➢This model was further validated with a pharmacological approach as chronic treatment with Imipramine or Fluoxetine was able to oppose the development of social aversion induced by chronic social defeat. Acute Imipramine was however without effect, confirming previously published studies using this antidepressant (Berton et al., 2006) .

➢ Aside from using these standard behavioral tests, we were able to discriminate the defeat phenotype with a probability of 91% that was evident 5 weeks post-defeat by employing our proprietary algorithm-based behavioral platform, SmartCube ®.

➢This system was also able to demonstrate a 40% recovery of defeat phenotype with chronic Imipramine treatment.

➢This suggests that such computer modeling may be used to predict clinical success for antidepressant drug candidates in this new model of depressive behavior.

ACKNOWLEDGEMENTS

➢We would like to extend a special acknowledgement to Dr. Eric Nester and Rosemary Bagot of Mount Sinai Icahn School of Medicine for all their essential help in developing this model at our facility. We would also like to thank the Behavioral Pharmacology Team for their help in performing the behavioral experiments and the IT department for their continuous support and innovation of SmartCube ® Technologies.

REFERENCES

Berton et al., Science 311, 864 (2006)
Donahue et al., Biol Psychiatry 76, 550 (2014)
Golden et al., Nature Protocols 6 (8), 1183 (2011)