Introduction:

Exposure to chronic stress has the potential to cause permanent damage to the brain if not remedied properly by our neural system through habituation. It was shown that habituation is mediated by the paraventricular nucleus of the thalamus (PVT), which acts on the hypothalamic-pituitary-adrenal axis. In previous studies, it was found that orexin-1-receptor (OX1R) is important in mediating the habituation response against foreign stressors.

Methods:

In this study, we used a rat model injected with a viral siRNA that inhibits the expression of orexin 1 receptors in the pPVT while also expressing GFP fluorescent protein. We performed immunohistochemistry to quantify injection of the viral siRNA to confirm the validity of the siRNA injection. We also quantified the total struggle duration for each rat in the daily restraint test. By quantifying the daily struggle duration, we were able to investigate the effect of knocking down OX1R in the pPVT on the rat's ability to habituate against a repeated stressor.

Results:

On day 1 of restraint, we did not observe any significant difference between the total struggle time in the first 12 minutes of the restraint test between the scrambled control group and the OX1R knockdown group. However, we did observe a drop in the total struggle duration in the scrambled control group after being subjected to repeated stressors for 5 days. On day 5 of restraint, the average total struggle duration in the first 12 minutes of restraint for the scrambled control group decreased from approximately 200 seconds on day 1 to approximately 100 seconds. The effects of habituation are most significant in the first few minutes of being exposed to a repeated stressor. This difference diminishes during longer intervals following restraint onset.

Conclusion:

Our findings support the hypothesis that OX1Rs in the pPVT are responsible for habituation and knocking down OX1R in the pPVT prevented any behavioral changes due to habituation. Future applications can potentially include genetic therapeutic options for treating chronic stress genetic disorders for lack of OX1R expression using gene-editing techniques including CRISPR-Cas9 or using OX1R as a biomarker to identify patients prone to chronic stress disorders.