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Survivors of critical illness often report traumatic memories of their illness period, and these memories are thought to contribute to development of neuropsychiatric disorders, such as PTSD. Many patients are treated with high doses of glucocorticoids for their vasoactive and anti-inflammatory properties, and glucocorticoids have also been shown to prevent the development of PTSD after trauma. Due to their activity in the hippocampus and amygdala, the putative protective effect of glucocorticoids may occur via memory formation during illness. To examine the effect of glucocorticoids on memory formation during acute infectious illness, male and female C57BL/6 mice (N=80, 40 male/40 female) underwent cecal ligation and puncture and were treated with either corticosterone (16 mg/kg) or vehicle in the early afternoon daily for five days beginning on the day of surgery. All mice were habituated to a neutral object in their home cage for five days and underwent one 30-minute footshock/no shock training session during the illness period. After physiologic recovery (2 weeks), the mice underwent behavioral testing including open field exploration, object recognition testing in which they were presented with both the familiar (habituated) object and a novel object, and testing in the shock context. The results showed that drug treatment had no effect on behavior in the open field, including time spent in the center (VEH: 20.19±10.81 vs CORT: 22.32±12.87 sec; P=0.476). Drug treatment increased overall object exploration (12.28±10.79 vs 19.17±15.88 sec; P=0.049). Corticosterone-treated mice showed a preference for the familiar object (60.9±23.0% of total exploration time with familiar object; P=0.015), while vehicle-treated mice did not (54.1±23.3%; P=0.378). The increase in overall object exploration seen in corticosterone-treated mice could be accounted for by an increase in exploration of the familiar object. History of footshock increased freezing in the training context (3.96±2.54% vs 36.08±15.42%; P<0.0001) and corticosterone treatment had no effect (18.06±17.65% vs 22.16±21.19%; P=0.557). In conclusion, administration of corticosterone during infectious illness facilitated memory of a neutral object from the illness period, and recovered mice exhibited a preference for this object over a novel one. Corticosterone treatment had no impact on fear memory formed during illness. This is consistent with human literature suggesting that hydrocortisone decreases PTSD symptoms without impacting traumatic memories. These findings suggest that glucocorticoids selectively enhance the formation, consolidation, and/or recall of neutral but not fear memories during illness, which may rely on hippocampal circuitry. We further suggest that accurate memories of the illness period may influence patients' perception of this experience and alter their risk for psychiatric sequelae.

Neuroendocrinology and Pituitary

NEUROENDOCRINOLOGY AND PITUITARY BASIC RESEARCH ADVANCES

Development of a Novel Tetracycline-Inducible *Kiss1*-Cre Mouse Line for Temporally Controlled Gene Deletion in Kisspeptin Neurons

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The tetracycline (Tet)-controlled inducible system is the most widely used reversible system for transgenic expression in mice. Previously, we generated a GnRH-CreTeR mouse model, using a first-generation Tet-inducible system to temporally induce expression of Cre recombinase in GnRH neurons. Recently, the Tet-inducible system has undergone several modifications to significantly reduce previous limitations that include leaky background expression and lower sensitivity to tetracycline induction. Therefore, we have developed a novel mouse model bearing a Tet-inducible kisspeptin-Cre allele (iKiss-Cre mouse) that will enable temporal control over the selective deletion of genes from Kiss1 neurons. This temporally controlled gene deletion will eliminate a longstanding technical limitation of conventional steroid receptor knockout models in which steroid regulation of the axis is confounded by steroid developmental and organizational effects in the reproductive axis. Two mouse lines were generated. The first line targets kisspeptin neurons with a third generation Tet-inducible reverse tetracycline transactivator (rtTA, Tet-On 3G) expressed under the control of the Kiss1 allele. Using CRISPR-Cas9 technology, we inserted a cassette containing an internal ribosome entry site (IRES) sequence followed by the rtTA downstream of the Kiss1 coding region as was previously done using Cre recombinase. Transcription of the recombinant Kiss1 allele yields a bicistronic messenger RNA, from which both kisspeptin and rtTA are independently translated. The second mouse line, TRE-Cre mice, was constructed to express Cre recombinase under control of the PTRE3G promoter. The PTRE3G promoter is bidirectional to allow simultaneous monitoring of Cre expression and a second florescent protein reporter (ZsGreen1). The Kiss1-rtTA only binds to and activates the PTRE3G promoter in the presence of doxycycline (an analog of tetracycline). Offspring from breeding of these mouse lines, iKiss-Cre, result in a system capable of generating Tet-induced expression of Cre recombinase in kisspeptin neurons. In order to document specificity and sensitivity of this system, we performed immunofluorescent staining and observed colocalization of kisspeptin with Cre recombinase in brain sections of iKiss-Cre mice only after doxycycline treatment. We will use this model to investigate negative feedback actions of E_2 on the adultpulse generator after E_2 exerts its organizational actions on maturing kisspeptin neurons. We expect that this mouse model will become a major tool used by the neuroendocrine community and serve as proof of principle for development of similar inducible knockout models employing the current generation of inducible methodologies.

Neuroendocrinology and Pituitary

NEUROENDOCRINOLOGY AND PITUITARY BASIC RESEARCH ADVANCES

Differences in Menin Expression in Pituitary Adenomas in Multiple Endocrine Neoplasia Type 1, Its Phenocopies and Sporadic Acromegaly