INTRODUCTION

- Stress is a precipitating factor of many psychiatric disorders including depression. Therefore, chronic exposure to stress is increasingly being employed in rodents to model stress-related psychiatric disorders and to test both the behavioural and neurobiological effects of psychotropic drugs.
- Lithium, the major pharmacotherapy for bipolar disorder, is also an effective add-on agent in antidepressant-refractory depression. While the precise molecular mechanisms underlying the antidepressant effects of lithium remain unresolved, it was recently reported that similarly to antidepressant drugs, chronic lithium treatment prevents stress-induced changes in behaviour and hippocampal neurogenesis in rats. However, comparable studies investigating the effects of chronic stress and lithium treatment in adult mice are lacking. Characterisation of their effects in mice is important because phenotyping of genetically-modified mice in such models could identify novel targets of antidepressant activity.

Neuroimaging studies suggest that altered structure and function of the hippocampus is characteristic of some stress-related disorders such as depression. Moreover, animal studies suggest that the hippocampus is anatomically and functionally divided into dorsal (dhi) and ventral (vhi) regions and that the vhi preferentially regulates emotionality and the stress response while the dhi is primarily involved in cognitive function (Fig. 1). Therefore, in the present study we investigated whether chronic immobilisation stress and/or chronic treatment with lithium would alter cell proliferation and survival along the septo-temporal axis of the hippocampus in a stress-susceptible mouse strain, the BALB/c mouse. Finally, since the neurotrophic factors VEGF and BDNF can regulate antidepressant-like behaviour and hippocampal neurogenesis, the effects of CIS and lithium treatment on hippocampal mRNA levels of these neurotrophic factors was also investigated.

EXPERIMENTAL DESIGN & METHODS

Experimental Design: Mice received either a 2h/day immobilisation stress or control diet for 21 days and were given access to a second sleeping cabin (0.84 m) habituated to the cage for 14 days. Ten days after starting the drug treatment, mice were stressed daily, 2 h per day for 15 days. Chronic immobilization stress (CIS) was employed as the stressor. Non-stressed control mice were not stressed. To assess the effects of lithium and CIS on cell proliferation, mice were killed at the end of the experiment and were perfused 24 h later. To assess the effects of untreated and vehicle control mice and after stress and drug treatment, mice were killed with a lethal dose of sodium pentobarbital (150 mg/kg, i.p.) and the brain was removed and the hippocampi were isolated. These hippocampi were then dissected into three sub-regions (dentate gyrus granular layer, hilus, and sub-granular zone). Sections were cut for immunohistochemistry (BrdU-positive cells) and mRNA analysis (using RT-PCR experiments). Animals were housed in ad-libitum diet (Harlan, Irvine, CA). Mean body weight was not significantly different across the treatment groups. In the hippocampus, mRNA expression was measured using real-time quantitative reverse transcription PCR (RT-qPCR) analysis. Data was analyzed using two-way ANOVA followed by post-hoc analysis with Fisher's LSD. Post-hoc tests were only conducted if one of the main effects of the two-way ANOVA reached a significance level of p < 0.05.

1. LITHIUM SIGNIFICANTLY INCREASES CELL PROLIFERATION IN THE VHI BUT NOT THE DHI OF STRESSED MICE

2. LITHIUM-INDUCED INCREASES IN CELL PROLIFERATION ARE SECONDARY TO REDUCTIONS IN CELL SURVIVAL

3. SUMMARY: EFFECTS OF LITHIUM AND STRESS ON CELL PROLIFERATION AND SURVIVAL

4. EFFECTS OF STRESS AND LITHIUM ON NEUROTROPHIC FACTOR mRNA LEVELS IN THE HIPPOCAMPUS AND ON BODY WEIGHT GAIN

CONCLUSIONS

- Lithium increased cell proliferation in the subgranular zone of the hippocampus but this effect was only significantly statistic in stressed animals. Moreover, these lithium-induced increases in cell proliferation were localized to the ventral region of the hippocampus. Such effects suggest that lithium-induced increases in hippocampal cell proliferation might only occur or become apparent when hippocampal function is confronted with challenges, such as stress.
- In addition to increasing cell proliferation in stressed animals; lithium also reduced the survival of cells that were generated prior to experimental treatment. This lithium-induced reduction in cell survival was observed in both stressed and non-stressed mice. Specifically, lithium decreased cell survival in the dhi of both stressed and non-stressed mice. In the vhi, lithium significantly reduced cell survival in stressed animals only.
- The lithium-induced increase in cell proliferation suggests a compensatory response to decreases in the survival of newly-born cells. However, upon summation of the total number of surviving BrdU-labelled cells with the total number of proliferating BrdU cells, it appears that lithium selectively increases the total number of BrdU-labelled cells in the ventral hippocampus of stressed animals only.
- These effects correlate with the lithium-induced increase in VEGF which was only observed in the hippocampus of stressed mice.
- Finally, lithium treatment also attenuated stress-induced reductions in body weight.
- The localization of lithium-induced cell proliferation to the vhi of stressed mice supports the hypothesis that the Vhi plays a preferential role in response to stress-related disorders. Current studies are investigating the functional role of neurogenesis in the vhi and dhi in various behavioural and physiological responses to chronic stress and antidepressant treatments.

ACKNOWLEDGEMENTS: This work was funded by a career development award (PD/2006/26) from the Health Research Board in Ireland (O.F.O.), Irish Research Council for Science, Engineering & Technology (R.M.D and L.F.C.), Science Foundation Ireland (L.F.C.) and the European Union (L.F.C).