Early life stress: longitudinal monitoring of morphological impact on the hippocampus using in vivo MR-Imaging in mouse model

W. Reichardt[1], C. Gross[2], I. Herpfer[2], C. Haas[3], K. Lieb[2], and D. von Elverfeldt[1]

[1]Dept. of Diagnostic Radiology, Medical Physics, University of Freiburg, Freiburg, Germany, [2]Department of Psychiatry and Psychotherapy, University Hospital Freiburg, Freiburg, Germany, [3]Experimental Epilepsy Group, University Hospital Freiburg, Freiburg, Germany

Introduction:

The hippocampus is a target for adrenal steroids that provides a model for studying neurobiological consequences of stress. The human hippocampus undergoes atrophy in the aftermath of traumatic stress, recurrent depression, and Cushing's syndrome as well as in some aging individuals [1]. Early life stress constitutes a risk factor for the development of psychological disorders, such as depression in human. Early life stress was modelled in C57Bl6 mice using a model of unpredictable maternal separation (daily 3h, between post-natal days 1-14) [2]. Previous results of our group had shown sex-related differences on RNA- and protein level between the experimental groups especially at day 15 post partum. We set out to show, that it is possible to detect morphological changes of the hippocampus in a early life stress model in mice using highresolution in vivo MRI.

Materials and Methods:

Altogether 16 male mice (C57/BL6) were kept with ad libitum access to food and water. Animals were maintained in a temperature-controlled room, with a light/dark cycle of 12/12h (lights on at 0700 h) and were randomly assigned to experimental groups on the first day post partum. Experiments were performed during the light period of the cycle and were conducted in accordance with the principles and procedures of local authorities for the Care and Use of Laboratory Animals. Control animals (n8) were left undisturbed, and stressed animals (n8) were subjected to daily deprivation stress for 3h in a separate room of the same animal facility on the post-natal days 1-14. The animals were imaged using a 9,4 Tesla animal Scanner (BioSpec 94/20, Bruker, Ettlingen, Germany). Mice were anesthetized under spontaneous breathing conditions using isoflurane. Respiration rate was continuously monitored and gating was used to reduce moving artefacts during the scan. The MRI–Protocol consisted of a localizer sequence to plan the brain imaging and a T2-weighted RARE Sequence (TEeff 60ms, Slicethickness: 0,4mm, MTX 196x196, FOV 18x18mm²) was used to depict the Hippocampus in the Mouse brain images Total hippocampus volumes were calculated from sets of contiguous images by summing products of area measurements and slice thickness using MIPAV, a freely available medical image processing software package from the National Institutes of Health (Bethesda, USA)

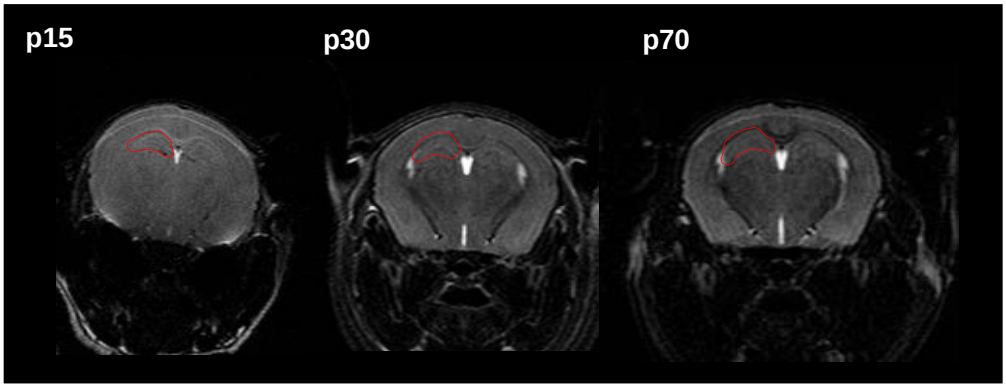
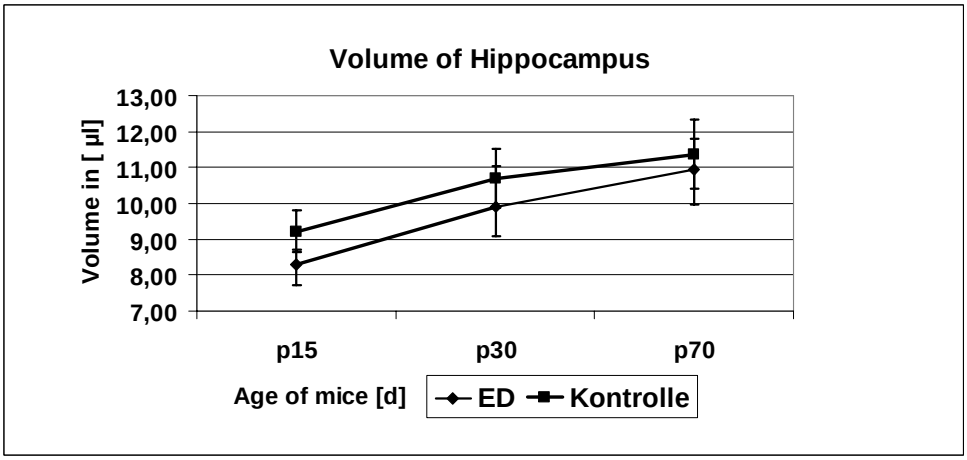


Figure 1: T2-RARE images taken on the day p15, p30 and p70 (postpartum) as indicated to depict the hippocampal area.

Results and Discussion:

We observed a difference in the mean hippocampal volumes (mHV) between the experimental groups on day 15 post partum. The mHV of the stressed mice was 8,31mm³, whereas mHV of the control animals was 9,41mm³. On day 30 postpartum the mHV of the stressed animals was still reduced about 1mm³ versus the control animals (9,92mm³ vs. 10,70mm³). The measurement on day 70 showed a further approximation of the mHV in both groups (10,94mm³ vs.11,42mm³).



Conclusion

In vivo MR imaging allowed us to monitor morphological changes in the brain development in an early life stress mouse model longitudinally.

References: **1**. McEwen BS, Magarinos AM (1997) Stress effects on morphology and function of the hippocampus. Ann N Y Acad Sci 821:271-84
2. Pryce, C.R. and J. Feldon, Long-term neurobehavioural impact of the postnatal environment in rats: manipulations, effects and mediating mechanisms. Neurosci Biobehav Rev, 2003. **27**(1-2): p. 57-71.