This discovery has aptly generated an immense discussion with regards to the possible functional implications of adult neurogenesis in both normal and pathological states. Particularly, sharp focus has been placed on the possible role of adult neurogenesis in depression where changes in adult neurogenesis were initially theorized to play a role in the pathogenesis and treatment of depression. The serotonin reuptake inhibitor (SSRI), Fluoxetine, is one of the most prescribed antidepressant medications and has been widely studied with regards to its effects on neurogenesis. Several studies have demonstrated that chronic fluoxetine treatment increases neurogenesis [1,2]. Furthermore Santarelli et al demonstrated that the ablation of neurogenesis blocks the behavioral effects of fluoxetine, a finding which has since been confirmed via different behavioral tasks in mice as well as rats [2]. These data strongly indicate that anti-depressive effects of fluoxetine are neurogenesis-dependent. The S100 protein family member, p11 (also known as S100A10) has recently been demonstrated to be of significant relevance to depression [3]. Specifically, p11 protein levels have been shown to be decreased in brains of an animal model of depression and moreover have also been shown to be decreased in brain tissue of depressed individuals. Furthermore, this decrease can be rescued in animal models using anti-depressants. Data from p11 knock-out (KO) mice indicate that these mice have a depressive-like phenotype and are less sensitive to the behavioral effects of the tricyclic antidepressant imipramine [3]. The relevance thus of both neurogenesis and p11 to depression makes studies into a possible correlation between p11 and neurogenesis a pressing question. Studies on the effects of the SSRI, fluoxetine, in p11 KO mice are also of interest due to the fact that p11 is known to bind specifically serotonin receptors, namely the 5HT-4 and 5-HT 1B receptors which may play a role in the mediation actions of SSRI's. Experiments were therefore designed to asses the effects of fluoxetine treatment on various aspects of adult neurogenesis in the subgranular zone (SGZ) of p11 KO mice. Cell proliferation measured using Ki-67 revealed that the fluoxetine mediated induction of cell proliferation seen in WT mice is absent in p11 KO mice. Similarly, the fluoxetine mediated induction in survival seen in WT mice is also absent in p11 KO mice. Finally, fluoxetine mediated increases in neurogenesis, measured using the immature neuronal marker DCX, which are seen in WT mice are also absent in p11 KO mice. Subsequent behavioral experiments using the anti-depressive-effects sensitive, novelty suppressed feeding (NFS) test revealed that the behavioral effects of fluoxetine were also absent in p11 KO mice. Together, these data indicate an important role of p11 in mediating anti-depressant effects of fluoxetine.

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P.1.007 Levels of activators of cyclin-dependent protein kinase 5 (Cdk5) p25 and p35 in Alzheimer's disease

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Purpose of the study: Cdk5 is a neuronal kinase involved in synaptic plasticity and memory formation. P35 is the most abundant activator of Cdk5 and can be cleaved to the more stable form, **p25**. In mouse models, low p25 levels can improve learning and memory (L&M) [1]. The underlying molecular mechanisms are still poorly understood. In the project depicted we chose an unbiased proteomics approach to find out protein changes in mice expressing low levels of p25 that could explain improved L&M.

We investigated p25 and p35 levels and Cdk5/p25-regulated proteins in **Alzheimer's Disease** (AD) post-mortem tissue and are currently analysing the behaviour of a potential AD mouse model. Finally, we are testing whether drug treatment can rescue cognitive deficits in the rodent model.

Methods: Quantitative proteomics were performed on hippocampal synaptosomes from wildtype and p25 transgenic mice. Levels of p25, p35 and of our newly identified Cdk/p25-regulated proteins were investigated in hippocampal postmortem tissue of AD patients with postmortem delay <24 h. Tissue from different stages of disease progression was examined by Western Blot. **Heterozygous p35 knockout mice** are currently studied in behavioural paradigms for hippocampal learning to see whether they may be a model for AD. Wildtype mice were treated with **SAHA** (suberoylanilide hydroxamic acid), a non-selective histone acetylase inhibitor to evoke changes in histone acetylation similar to the ones observed in p25 transgenic mice.

Summary of results: Quantitative proteomics indicated an upregulation of Optic atrophy 1 (**OPA1**) in p25 transgenics (P = 0.05(*), D = 27.1%, n = 3,3). OPA1 plays a role in spine formation, mitochondrial fusion and neuroprotection. Changes were confirmed by Western Blot (P = 0.019*, D = 63.7%, n = 10,10).

We found a significant decrease of p25 and p35 early in AD (normalization by actin and NSE; control vs Braak 2: $P_{actin} < 0.001$***, $D_{actin} = -54.6\%$; $P_{NSE} = 0.014$*, D = −53.9%; n = 8,13). Likewise we observed a decrease of OPA1 in a similar time course ($P_{actin} = 0.008$**, $D_{actin} = -32.2\%$; $P_{NSE} = 0.038$*, D = 32.2%; n = 12,18). P35 has never been studied in the context of AD. Therefore we are currently investigating the role of p35 in cognition by employing heterozygous p35 knockout mice.

As changed histone deacetylase activity is implicated in neurodegeneration as well as L&M [2,3], we investigated histone acetylation in the mice expressing p25. We found increased acetylation of H3K18 but not H4K12 in p25 mice. Similar changes in histone acetylation occurred when we treated wildtype mice with **SAHA**. The next step will therefore be to examine whether SAHA treatment can rescue memory deficits in heterozygous p35 knockout mice.

Conclusions: In this project we provide evidence that p35 and p25 are downregulated in AD, an observation that matches the phenotype of improved L&M in p25 transgenic mice. To explore the role of p35 in AD, heterozygous p35 knockout mice are currently being tested in a behavioural battery. SAHA treatment evokes a pattern of histone acetylation similar to the one observed in p25 transgenic mice. We therefore plan to use SAHA to reverse cognitive defects (if any found) in heterozygous p35 knockout mice.

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P.1.008 Evaluation of the endocannabinoid system in post-mortem human prefrontal cortex of alcoholic subjects

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Introduction: Several reports suggest the involvement of the endogenous cannabinoid system in cerebral mechanisms underlying drug addiction, including alcoholism. Behavioural studies in rodents have shown that the administration of CB1 antagonists and FAAH (fatty acid amide hydrolase, the principal endocannabinoid inactivating enzyme) inhibitors reduce alcohol intake, while CB1 agonists increase it. Furthermore, chronic exposure to ethanol also induces alterations in the endocannabinoid system in animal models: increasing endocannabinoid content, while decreasing FAAH activity and CB1 receptor density and functionality. In parallel, human genetic studies have suggested the involvement of particular CB1 and FAAH coding polymorphisms in alcoholism. A recent report described an increase in CB1 receptor expression and density in prefrontal cortex of alcoholic suicide victims when compared with chronic alcoholics dying from causes other than suicide [1].

Aims: The purpose of this study was to evaluate the endocannabinoid system in post-mortem human brain of subjects with a previous history of alcoholism. Thus, the protein expression of the CB1 receptor, its functional coupling at the G-protein level and at the adenylate cyclase (AC) activity, and the activity of FAAH were assessed.

Methods:

1. Expression of the CB1 receptor was determined by immunoblot experiments.
2. Functional coupling of CB1 receptor to G-protein was evaluated by WIN 55,212–2 (10^{-12}-10^{-3} M, 10 concentrations) stimulated [^{35}S]GTPγS binding.