Summary

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Proper visual experience during postnatal development is of critical importance for the maturation of the visual system and its functioning later in life. It determines the way in which neurons in the visual cortex respond to visual stimuli, which is exemplified by the consequences of monocular deprivation, the occlusion of input through one of the two eyes.

Monocular deprivation changes the responsivity of neurons in the visual cortex to input from the deprived eye and also reduces visual acuity through this eye. These functional changes are accompanied by structural rearrangements such as dendritic spine turnover and reorganisation of neurites; therefore proteins involved in the regulation of neuronal morphology are likely to play a role in visual plasticity.

One such protein that is known to regulate the morphology of neuronal dendrites and also influences learning is the transmembrane receptor Notch1. In **chapter 2** we describe our study into the role of Notch1 signalling in plasticity of the mouse visual cortex, in which we make use of transgenic mice that conditionally express active Notch1 postnatally in cortical pyramidal neurons. We demonstrate that neuronal Notch1 signalling cell-autonomously reduces dendritic spine- and filopodia densities and reduces long-term potentiation in the visual cortex. Upon monocular deprivation, these effects of Notch1 activity predominantly affect responses to visual stimuli with higher spatial frequencies. This results in an enhanced effect of monocular deprivation on visual acuity.

After having established the effects of Notch expression in pyramidal neurons on cortical plasticity, we aim to characterise the molecular signalling downstream of Notch in cortical pyramidal neurons in order to identify potential mediators of these effects. Since Notch functions as a transcriptional regulator, we performed microarray experiments on the cerebral cortex of mice transgenically overexpressing constitutively active Notch1 in all cortical pyramidal neurons. In **chapter 3** we compare gene expression in these animals with littermate control mice and in this way screen for target genes downstream of Notch signalling. In these animals neuronal Notch1 signalling predominantly affects the expression of genes involved in transcription and in signalling through the Ras/MAPK pathway. In addition we identify a negative feedback mechanism that involves the histone deacetylase HDAC4, restricting Notch1-mediated transcription.

Visual plasticity is enhanced during the critical period, enabling the acquisition of knowledge and skills that we depend on in later life. Essential to the persistent changes in neuronal connectivity and plasticity are the underlying molecular changes tak-

ing place at the level of the synapse. In order to provide an insight into the molecular pathways involved in visual plasticity, genetic screens were adopted during the past decade. Studies using differential display or microarray techniques analysed expression patterns of thousands of genes under experimental conditions of high and low plasticity. Studies in which proteins rather than genes are the subject of investigation are scarce and the same holds true for studies focussing on the neuronal compartment featuring the actual neuronal connection and thus the subject of synaptic plasticity: the synapse. In **chapter 4**, we describe our study into the expression of synaptic proteins under different plasticity-modulating experimental conditions. We compare the synaptic expression of proteins in animals at the peak of the sensitive period (P30), in animals of that age that were monocularly deprived for 4 days from P26 onwards (P30-MD), in animals in which the sensitive period had ended (P46) and in animals of that age that were reared in the dark (P46-DR). We characterise synaptic expression of proteins in these four groups by means of an iTRAQ proteomics approach based on sequential liquid chromatography and tandem mass spectrometry. We show that induction of visual cortex plasticity by means of monocular deprivation during the critical period results in increased levels of kinases and proteins regulating the actin-cytoskeleton and endocytosis. Closure of the critical period with age coincides with increased levels of proteins associated with transmitter vesicle release and the tubulin- and septin-cytoskeletons, and decreased levels of proteins regulating the actin-cytoskeleton, in agreement with increased stability and efficacy of synapses. Dark rearing, a process that keeps the visual cortex plastic, prevents only some of these changes. It also results in increased levels of G-proteins and protein kinase A subunits. We conclude that dark rearing does not simply result in delayed cortical maturation but may also activate specific signalling pathways that provide increased visual cortex plasticity. Interestingly, we also identify many proteins of which synaptic expression is regulated by age and visual input, and that are associated with Wallerian axon degeneration. We subsequently demonstrate that in mutant mice in which Wallerian degeneration is delayed, visual plasticity is indeed reduced.
Finally, **chapter 5** provides a discussion of the findings in this thesis, and discusses them within the framework of current knowledge.