Summary

Voltage-dependent Calcium channels (Ca$_{v}^{2+}$ channel) respond to a depolarization with calcium influx. An action potential (AP) reaching the axon terminal causes opening of Ca$_{v}^{2+}$ channels. Calcium flows into the axon due to electro-chemical driving force. The increased calcium concentration within the axon terminal elicits the release of neurotransmitter. The high-voltage-gated calcium channels of the R- (Ca$_{v}^{2.3}$), P/Q- (Ca$_{v}^{2.1}$) and N- (Ca$_{v}^{2.2}$)-type are mainly expressed in neurons where they initiate neurotransmission of fast synapses. In many synapses P/Q-, N- and R-type channels cooperate in controlling neurotransmitter release, but P/Q-type channels have a dominant role, partly because of a more efficient coupling to the exocytotic machinery. The somatodendritic localization of Ca$_{v}^{2.1}$ channels points to additional postsynaptic roles, e.g. in neural excitability. Ca$_{v}^{2.1}$ channels are coupled to calcium-activated potassium channels (big conductance (BK)- and small conductance (SK)-channel), which contribute to repolarization of APs and to the fast component of the afterhyperpolarization. Mutations in the CACNA1A gene that encodes the pore-forming α$_1$ subunit of Ca$_{v}^{2.1}$ channels cause several autosomal-dominant neurologic disorders, including familial hemiplegic migraine type 1 (FHM1), episodic ataxia type 2, and spinocerebellar ataxia type 6 (SCA6). Knockout of the α$_1$ subunit in the whole brain of mice provokes death within 3-4 weeks of age which demonstrates the important role of Ca$_{v}^{2.1}$ channels.

The neurotransmitter serotonin (5-Hydroxytryptamine, 5-HT) plays an important role in a variety of physiological processes such as regulation of blood pressure, temperature, appetite, sleep cycle, sexual behavior, mood and nociception. Pathological changes of the serotonin level in the central nervous system (CNS) induce neurological disorders such as i. a. depression, anxiety and aggression. Serotonergic neurons are located within the brainstem raphe nuclei consisting of the dorsal (DRN), median (MnR) and caudal (CRN) raphe nuclei. So far, it is not much known about the function of Ca$_{v}^{2.1}$ channels in serotonergic neurons. Therefore, this thesis focused on the electrophysiological characterization of Ca$_{v}^{2.1}$ channels in 5-HT neurons. In addition, a P/Q-type channel knockout (KO) mouse line which lacks P/Q type channels in serotonergic neurons only (ePet-Cre/α$_{1a}$-floxed-citrine$^{(+/-)}$) was created. Subsequently, KO mice were examined in anxiety and aggression-related behavioural tests.

This thesis demonstrates that KO mice exhibit an increased aggressive behavior in the resident intruder and displacement test compared to control mice. The electrophysiological in vitro experiments in acute brains slices showed an increased spontaneous firing frequency of DRN and CRN 5-HT neurons after inhibition of Ca$_{v}^{2.1}$ channels with ω-Agatoxin IVA. Following inhibition of Ca$_{v}^{2.2}$ with ω-Conotoxin GVIA frequency was further increased in DRN neurons but not in CRN 5-HT neurons. There was no difference between the spontaneous activity in DRN and CRN neurons before and after application of ω-Agatoxin IVA. The inhibition of SK and BK channels with Apamin and Iberiotoxin, respectively had no effect on spontaneous activity either. In the KO the frequency was unchanged compared to the WT. However, their firing pattern was irregular compared to WT.
Further, it was examined which role Ca\textsubscript{2.1} channels play in transmitter release. This was studied in autosynapses within the DRN and in heterosynapses of the hippocampus. The amplitudes of 5-HT\textsubscript{1A}-autoreceptor mediated slow inhibitory postsynaptic potential (sIPSP) was reduced about 39.3 % and 40 % after inhibition of Ca\textsubscript{2.1} and Ca\textsubscript{2.2} channels, respectively. Moreover the kinetics of hyper- and repolarization was faster after blocking the two calcium channels. Ca\textsubscript{2.1} channel inhibition caused no effects on paired-pulse-inhibition (PPI). Additional inhibition of Ca\textsubscript{2.2} channels depressed PPI. KO sIPSP amplitudes were not changed compared to WT amplitudes before and after application of calcium blockers. The time constant of repolarization is faster in KO 5-HT neurons.

Neurotransmission in heterosynapses was investigated using optogenetic tools. MnR neurons innervate hippocampal interneurons. Therefore, an adeno-associated virus carrying an eYFP tagged channelrhodopsin2 under control of the neuron specific promotor synapsin was injected into the MnR. The virus was transported anterogradly transported in serotonergic axons. Hence it was possible to visualize the eYFP tagged channelrhodopsin in hippocampal slices. Light induced excitatory postsynaptic currents (EPSC) were reduced after application of ω-Agatoxin IVA (57 % reduction) and ω-Conotoxin GVIA (25 % reduction).

This thesis reveals for the first time that P/Q-type calcium channels play a prominent role in modulating behavior. Further electrophysiological experiments showed that inhibition of Ca\textsubscript{2.1} channels caused a reduction of postsynaptic responses, within the DRN as well as in in hippocampal interneurons. This highlights the involvement of Ca\textsubscript{2.1} channels in transmission of serotonin. In addition, it seems that P/Q-type calcium channels are important to maintain firing frequency and regularity in 5-HT neurons.