English Summary

Major depression is a chronic mental disorder that affects approximately 1 out of 7 individuals during their lifetime, independently of their cultural, societal, occupational or financial status. Depression onset is traditionally linked to genetic predisposition (inherited vulnerability), exposure to adverse conditions (extreme, prolonged stress) or interaction of the two. The depressive state is characterized by anhedonia, i.e., reduced ability to experience pleasure from otherwise rewarding activities, such as social interaction. Anhedonia encompasses a variety of motivational deficits, including difficulties in reward anticipation, cost-to-benefit evaluation and goal directed decision-making. Furthermore, depression is accompanied by mild cognitive symptoms, such as reduced concentration and impaired memory, which persist even after mood recovery. This phenotype manifests in combination with cognitive biases, i.e., attentional allocation towards negative experiences and difficulty in processing of emotionally charged information. Together, depression incorporates complex endophenotypes with probable multifactorial causes. Due to this, and despite coordinated efforts from researchers and clinicians alike, the neurobiology of depression has remained poorly understood. Consequently, therapeutic options against this disorder are limited, with ample side effects and only a third of patients responding to available treatments.

In the last decades, preclinical models of depression have been developed and extensively used. These mainly include paradigms employing stress exposure (ethological validity), aiming to recapitulate the depressive state concerning the symptoms (face validity) and underlying neurobiological mechanisms (construct validity) and to identify novel treatment targets (predictive validity). During my Ph.D. candidacy, I worked with the social defeat-induced persistent stress (SDPS) paradigm in rats, a credible preclinical model of chronic depression. Using SDPS, a variety of behavioral and neurobiological hallmarks of the human disease can be observed, including decline in social behavior, memory impairment and morphological and functional deterioration of the hippocampus. These depression-associated phenotypes last for months following exposure to social defeat stress and respond positively to currently available antidepressant medication. The first aim of my project was to describe SDPS-induced behavioral endophenotypes of depression, including deficits in the affective domain (e.g. anhedonia). For this I addressed the effect of the depressive state in appetitive and motivational states towards natural (sucrose) and non-natural (alcohol) rewards. The second goal of my Ph.D. was to unravel the molecular and cellular adaptations governing depression-associated cognitive deficits and antidepressant response. Thus,

I used the SDPS model to study the molecular signature of the depressive state with a focus on alterations occurring at the synaptic level.

In **chapter 2**, I developed a behavioral paradigm for primary depression and secondary alcohol use disorder (AUD), establishing the first preclinical model of comorbidity between the two disorders. Using the SDPS model in combination with alcohol self-administration, I investigated the effects of the sustained depressive state on alcohol consumption, motivation to acquire alcohol and reinstatement of alcohol-seeking and -taking following extinction. My data suggested that chronic depression confers susceptibility to the development of AUD, as SDPS animals showed increased motivation for alcohol and an exaggerated relapse response. These SDPS effects were ameliorated by guanfacine, an α2A adrenergic receptor agonist, corroborating a role for this FDA-approved agent as a possible target for the treatment of the comorbid phenotype.

In **chapter 3**, I described the effects of SDPS on sucrose self-administration, aiming to experimentally test a novel concept for depression-induced anhedonia. In particular, I examined instrumental responding for sucrose long after the application of stress, including measures of motivation towards sucrose consumption, extinction and reinstatement of sucrose-seeking behaviors. SDPS had no effect on sucrose consumption, which is reduced following exposure to acute stress. In contrast, chronic depression increased motivation for sucrose, impeded initial extinction of sucrose-seeking and exaggerated the relapse response. According to my data, the anhedonic state incorporates a broader spectrum of motivational deficits that are independent of preference for an enticing natural reward. These deficits are reflected in difficulty in assigning value to a given reward and in subsequent reward-oriented action following the integration of reward-coding information.

In **chapter 4**, I examined individual variability to the effects of stress in subsequent development of the depressive pathology. Particularly, by employing the SPDS paradigm in an outbred population of rats and assessing its effects in different behavioral readouts, I reliably identified depression-prone *vs.* depression-resilient rats, long after stress exposure. This led to a detailed profiling of depressive symptoms in the affective (social behavior) and the cognitive (spatial memory) domain, as they develop over time. Importantly, my findings indicated that affective susceptibility emerges before cognitive deficits in chronic depression, mimicking the temporal progression of depression observed in the clinic. Furthermore, this approach confirmed the usability of the SDPS model in studying resilience to the effects of depression-inducing stress.

In **chapter 5**, I questioned the impact of depression susceptibility in subsequent vulnerability to alcohol abuse. Long after stress exposure, SDPS-prone and -resilient rats were subjected to alcohol self-administration. Based on the diagnostic criteria of addictive pathologies in humans, aspects of addiction-vulnerability were examined in the two subpopulations, such as alcohol craving, compulsive alcohol-seeking and relapse. My work highlighted depression proneness as a risk factor for the emergence of dependence-like phenotypes, as SDPS-prone rats exhibited excessive preoccupation with alcohol and alcohol-associated impulsivity. Likewise, I demonstrated that depression resilience limits the development of addiction-like behaviors, suggesting common (epi)genetic protective mechanisms for depression and alcohol dependence.

In **chapter 6**, I examined the synaptic, cellular and network-level changes that underlie cognitive dysfunction in chronic depression. For this, I combined behavioral, biochemical and electrophysiological techniques to assess the impact of synaptic and cellular remodelling of hippocampal extracellular matrix (ECM) on depression-induced cognitive decline. In particular, I showed that the sustained depressive-like state is characterized by an antidepressant-reversible increase in the expression of hippocampal perisynaptic ECM, as observed in increased synaptic levels of chondroitin sulphate proteoglycans. This was in parallel with an increase in pericellular ECM, as reflected in increased number of perineuronal net-coated interneurons of the CA1 hippocampal subfield. These molecular and cellular adaptations were accompanied by reduced plasticity potential (long-term potentiation) and disrupted inhibitory tone (IPSCs frequency) in the hippocampus after SDPS. Enzymatic removal of excessive ECM using chondroitinase ABC was able to restore ECM organization and hippocampus function. Together, these data suggested an ECM-mediated disruption of excitatory and inhibitory transmission in the hippocampus, resulting in reduced plasticity and consequently, in cognitive impairment in depression. Importantly, my work highlighted a previously unexplored value to ECM organisation in the antidepressant response.

To conclude my thesis, **chapter 7** was dedicated to discussing the usefulness of the SDPS paradigm as a preclinical model of depression. I reviewed the combined output of my work in light of the diagnostic criteria for depression in humans, with a focus on affective and cognitive symptoms that were best described in my work. Collectively, the research described here aimed to contribute to our understanding on the behavioral and molecular underpinnings of depression. Novel opportunities for treating depression, such as the manipulation of hippocampal ECM, were identified that might lead future therapeutic attempts.