Cannabinoid and opioid modulation of impulsive behavior and drug addiction

Occasionally, it happens to all of us: you speak out of turn during a meeting or you make a comment and immediate realize that you shouldn't have said it. Or you buy an expensive pair of shoes even though you actually cannot afford them. Two examples of daily-live impulsive behavior, and more specifically of impulsive action (acting without forethought) and impulsive choice (preferring immediate gratification over a delayed reward). When somebody is hyper-impulsive, this personality trait can negatively affect that person's life. Failure to suppress impulsive behavior (failing impulse control) is a well-known endophenotype in people suffering from Attention-Deficit/Hyperactivity Disorder (ADHD) or drug addiction, but also, for instance, in people with a bipolar disorder (during the manic phases). Hyper-impulsivity results from abnormal brain functioning. However, to date, the exact malfunctions in the 'impulsive brain' remain largely unknown, thereby hampering effective treatment of this maladaptive behavior. An important aim of this Ph.D. thesis was to gain more insight into the neurobiology of impulsivity. In addition, this thesis examines some aspects of the neurobiology of drug addiction, a psychiatric disorder that, as mentioned before, is in most patients characterized by impulse control-related problems, particularly during the abstinence phase when drug addicts have been drug-free for some time and try to prevent relapse. By using rat behavioral models combined with various pharmacological approaches, we have studied the neurobiology of impulsivity and relapse to drug seeking, in search of putative 'targets' for novel medication against hyper-impulsivity and/or drug addiction. Thereby, we focused specifically on the cannabinoid and opioid neurotransmitter systems, as previous studies had already suggested a role for these two neurotransmitter systems in regulating both impulsive behavior and relapse to drug seeking.

Cannabinoid and opioid modulation of impulsive behavior
For the impulsivity project (Chapters 2-4), the main aim was to elucidate under which conditions, and via which neuronal mechanisms, the endogenous cannabinoid and opioid systems can mediate two types of impulsive behavior. To this end, different groups of rats were trained for 2-3 months in either a 5-choice serial reaction time task (5-CSRTT), to measure an aspect of impulsive action (the inability to inhibit inappropriate responses), or a delayed reward task (DRT), to assess impulsive choice (inability to wait for a larger but delayed reward). Subsequently, the role of various receptor subtypes of cannabinoid and opioid receptors in mediating impulsive action and impulsive choice was determined. In a series of test days, different receptor agonists (for receptor activation) and antagonists (for receptor inactivation) were administered to the rats (via systemic or intracranial injections) immediately prior to a behavioral session. On several of the test days, rats would additionally receive a challenge injection with one of three psychostimulant drugs, amphetamine, nicotine, or GBR 12909 (a selective dopamine transporter inhibitor). These compounds each acutely enhance impulsive responding in the 5-CSRTT, while reducing impulsive
decision making in the DRT. Thus, we could additionally determine the role of the cannabinoid and opioid neurotransmitter systems in transient, drug-induced changes in impulsive behavior.

In short, our research shows that the cannabinoid and opioid receptor systems play important, but largely divergent and behavioral-specific roles in regulating impulsive action and impulsive choice. Particularly activity of the cannabinoid CB1 receptor subtype and the µ-opioid receptor subtype was found to affect both types of impulsive behavior. Based on the data and literature, although we cannot rule out involvement of other neurotransmitter systems, it is conceivable that cannabinoid modulation of impulsive action and impulsive choice as well as opioid modulation of impulsive action (but not impulsive choice!) involve interactions with the dopamine neurotransmitter system. However, such interactions are likely to be brain region-specific, occurring in cortical (dopamine-cannabinoid) and subcortical (dopamine-opioid) brain regions. Additionally, the exact properties of dopamine signaling (how much dopamine, between which neurons, and within what time window) may be a crucial determinant of the nature of these interactions. With regard to the localization of cannabinoid and opioid receptors that mediate impulsivity, our data (Chapter 2) demonstrated that µ-opioid receptors in a subcortical brain region, the nucleus accumbens shell, are critically involved in regulating impulsive action. Stimulation of these receptors by administering an agonist induced impulsive responding in the 5-CSRTT, whereas blockade of the receptors with an antagonist prevented amphetamine-induced impulsivity in that task. It is however conceivable that also the activity of µ-opioid receptors in other brain regions may affect impulsive action, and this remains to be established in future studies.

Altogether, our studies demonstrate that the neuronal mechanisms underlying different aspects of impulsive behavior, and the effects of drugs of abuse thereon, are more complex and divergent than previously thought. On a larger scope, our data are consistent with those of previous studies showing that the neuronal mechanisms underlying different aspects of impulsivity (e.g. impulsive action and impulsive choice) are largely dissociable. This calls for a more personalized treatment approach. The two neurotransmitter systems targeted by all current impulse control medication are the dopamine and noradrenaline systems. It is interesting that our data demonstrate that the roles of the cannabinoid and opioid systems in impulsive behavior are at least partly dissociable from those of the dopamine and noradrenaline systems. Thus, future research should investigate whether patients that do not respond to the currently available medication (30-40% of all people suffering from hyper-impulsivity) may be responsive to cannabinoid- and/or opioid-based drugs.

**Release of endocannabinoids during relapse to drug seeking**

Rehabilitation of drug addicts is often hampered by their persistent vulnerability to relapse, particularly when exposed to drug-associated cues, drugs of abuse, or stress. Based on pharmacological intervention studies, the endogenous cannabinoid system, and particularly cannabinoid CB1 receptor activity, is thought to be critically involved in cue- and drug-induced reinstatement of drug and food seeking. However, it was
unknown if there is a change in endocannabinoid release in the brain during a relapse to drug and/or food seeking.

In Chapter 5, we aimed to answer this fundamental question by combining an in vivo microdialysis technique (to measure neurotransmitter release in awake, behaving rats) with an operant rat model of relapse behavior. Using this behavioral model, we first confirmed that CB1 receptor antagonists indeed suppress relapse to heroin and sucrose seeking by preventing endocannabinoid-induced CB1 receptor activation. Intriguingly, our micro-dialysis data subsequently revealed that relapse to heroin seeking is associated with a robust release of specifically the endocannabinoid 2-arachidonyl glycerol (2-AG), whereas a reduction in extracellular levels of the endocannabinoid anandamide was observed following the relapse test. These effects were not observed in rats that were trained to self-administer sucrose rather than heroin. Moreover, the effects were found to be restricted to the dorsal part of the medial prefrontal cortex, which is a brain region known to drive drug seeking in both rats and humans via its glutamatergic projections to the nucleus accumbens core subregion. From this, we hypothesize that aberrant 2-AG signaling in the dorsomedial prefrontal cortex underlies the excessive motivational influence of drug-paired cues, and may be an upstream event in the induction of drug craving and relapse that could be targeted by future anti-relapse medication.

Concluding remarks
This thesis revealed important distinctions between cannabinoid CB1 and µ-opioid receptors in the brain regarding their involvement in regulating two aspects of impulsive behavior, impulsive action and impulsive choice. Furthermore, we demonstrated that the mechanisms underlying drug-induced changes in impulsive behavior may not be identical for all drugs of abuse. Finally, we discovered that relapse to heroin seeking is associated with brain region- and reinforcer-specific patterns of endocannabinoid release in the brain. As such, the present work significantly contributes to our knowledge of the neuronal events that mediate impulsivity and drug addiction. Future studies are clearly warranted to further substantiate our findings. This thesis may guide such experiments as it identified relevant test conditions as well as brain regions for studying the role of cannabinoids and opioids in impulsivity and relapse to drug seeking.