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THE POTENTIAL OF PSYCHEDELICS IN THE TREATMENT OF DRUG ADDICTION IN ANIMAL MODELS

POTENCIÁL PSYCHEDELIK V LÉČBĚ DROGOVÉ ZÁVISLOSTI V ANIMÁLNÍCH MODELECH

Bachelor´s thesis

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V Praze, 1. 5. 2022 Barbara Mallarino

Poděkování

Touto cestou bych chtěla poděkovat své školitelce Kláře Šíchové spolu se skupinou doktora Páleníčka v NUDZ za čas mně věnovaný. Dále svým rodičům a příteli za podporu při studiu. Zároveň děkuji svému kamarádovi Vlastimilovi za uvedení do této problematiky.

Abstrakt

Psychedelika jsou psychotropní látky ovlivňující percepční vnímání a vědomí, které mohou vyvolat významné změny v běžném propojení oblastí mozku. Hlavní pozornost je věnována některým z nejslibnějších sloučenin pro výzkum závislostí a duševního zdraví: LSD, tryptaminům (psilocybin, DMT) a ibogainu. V první části jsou shrnuty základní neurobiologické procesy vzniku závislosti a nejčastěji používané zvířecí modely a metody. Následuje popis změn v konektivitě mozkových oblastí vyvolaných psychedeliky a molekulárních mechanismů účinku s důrazem na potenciální antiadiktivní vlastnosti. Poslední část je pak zaměřena na údaje získané z preklinických studií na zvířatech, které pomáhají hlouběji pochopit mechanismy, jež mohou být základem jejich účinnosti při klinické léčbě drogové závislosti.

Klíčová slova: drogová závislost; psychedelika; LSD; tryptaminy; ibogain; animální modely

Abstract

Psychedelics are mind-altering and perception-changing psychoactive compounds that can produce some significant changes in the ordinary wiring of the brain. Substantial attention is paid to some of the most promising compounds for addiction and mental health research: LSD, tryptamines (psilocybin, DMT) and ibogaine. The underlying neurobiological basis of the development of addiction and relevant animal models and methods are described in the first part. Following, changes in brain regions connectivity induced by psychedelics and molecular mechanisms of action are discussed, emphasising the potential anti-addictive properties. The last part of the thesis focuses on data acquired from preclinical animal studies, which helps to further understand some of the mechanisms underlying their effectiveness in the clinical treatment of drug addiction.

Keywords: drug addiction; psychedelics; LSD; tryptamines; ibogaine; animal models

List of abbreviations

18-MC	18-methoxycoronadirine
5-HT	5-hydroxytryptamine
5-MeO-DMT	5-methoxy-dimehyltryptamine
ADHD	attention deficit and hyperactivity disorder
cAMP	cyclic adenosine monophosphate
CNS	central nervous system
CPA	conditioned place aversion
CPP	conditioned place preference
CRF	corticotropin-releasing factor
DA	dopamine
DD	dopamine deficient
DMN	default mode network
DMT	N,N-Dimethyltryptamine
DOM	2,5-Dimethoxy methylamphetamine
DOI	2,3-dimethoxy-4-iodo amphetamine
DOPAC	dihydroxyphenylacetic acid
DRN	dorsal raphe nucleus
ER	endoplasmic reticulum
fMRI	functional MRI
FR	fixed ratio
GABA	gamma-aminobutyric acid
GPCR	G-protein coupled receptor
HPA	hypothalamic-pituitary adrenal
HVA	homovanillic acid
HVA	homovanillic acid
INMT	indolethylamine-N-methyltransferase
IV	intravenous
LAT	Locomotor activity test
LSA	lysergic acid
LSD	lysergic acid diethylamide
MAO	monoamine oxidase
MCP	mesocortical pathway
MLP	mesolimbic pathway
mPFC	medial prefrontal cortex
mRNA	messenger ribonucleic acid
NAC	nucleus accumbens
NAch	nicotinic acetylcoline
NMDA	N-methyl D-aspartatic acid
NMT	N-methyltryptamine
OFC	orbitofrontal cortex
PCC	posterior cingulate cortex
RNA	ribonucleid acid
SA	self-administration
TAAR	trace amine- associated receptor
THC	tetrahydrocannabinol
VMAT	vesicle monoamine transporter
VTA	ventral tegmental area
mCPP	1-(3-chlorophenyl) piperazine

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1 Introduction

Psychedelics have been used for thousands of years as sacred plant medicines in religious contexts across different world regions, in Africa with iboga, north America with ayahuasca, and Europe with psilocybin and other hallucinogenic mushrooms. Two discoveries in recent history mark the beginning of research into these compounds: the isolation of mescaline from the *Peyote* cactus in 1898 by Arthur Heffter and the isolation and discovery of psychedelic effects of LSD in 1943 by Albert Hoffmann (described in his book "LSD: my problem child"). Almost all research on psychedelics comes either from the early 1960s and 70s or from recent years, as the period in between is characterized by the hippie movement, which popularized these compounds and led to a 40year prohibition period. Although starting in the late 1990s and through recent years, a large interest in the therapeutic application of these psychedelic compounds began to emerge.

Classic psychedelics (DMT, psilocybin, LSD, ibogaine and mescaline) in humans produce some of the same subjective effects: changes in consciousness and perspective, or as Rick Strassmann puts it:

"Psychedelics show you what's in and on your mind, those subconscious thoughts and feelings that are hidden, covered up, forgotten, out of sight, maybe even completely unexpected, but nevertheless imminently present." (Rick Strassmann- *DMT, the spirit molecule*).

With the surge in anecdotal evidence from people with addictive disorders who self-medicate with plant psychedelics in rituals like ayahuasca and clinical evidence from controlled settings, a question started to emerge whether these substances do in fact change brain chemistry or they work strictly by causing a shift in perspective. This hypothesis has been and is currently still being tested using animal models of addiction. Preclinical data from controlled laboratory environments done on rats and mice lay the grounds for ongoing human double-blind clinical trials, resulting in new approaches for the treatment of mental disorders and substance dependence.

This thesis aims to elucidate some of the potential mechanisms through which psychedelics could alter substance craving, compulsive habits, and withdrawal symptoms while describing the animal models currently used and available preclinical data.

2 Substance addiction

Addiction has long been classified as a brain disease in which substances irreversibly change brain chemistry (Leshner, 1997). Later models refer to addiction as not just a chemical imbalance but rather a multi-aspect problem, as the Bio-Psycho-Social-Spiritual model describes (Engel, 1977; further reviewed in Skewes and Gonzalez, 2013). These factors include genetic predispositions, brain changes on a molecular level, social influences, the individual's personality, spirituality, and relationship to oneself. In recent years, new research and literature have come out explaining addiction as "a result of natural neuronal plasticity reinforcing learned behaviours"[1]. Research on this topic is complex because of the multifactorial nature of this behavioural abnormality and commonly used terms such as "dependence" "abuse" or "withdrawal" can be easily misinterpreted and need to be defined.

2.1 Definitions

"Substance abuse" is defined by the world health organization (WHO) as harmful (causing damage to both physical and mental health) or hazardous use of psychoactive substances (both legal and illegal). According to the ICD-10 (international classification of diseases, icd.who.int/browse10/2019/en), substance "dependence" or "addiction", is a combination of behavioural, cognitive, and physiological attributors to repeated substance use and these terms are used interchangeably in the text. "Withdrawal" (also by the ICD-10) is a short-term unpleasant state induced by persistent substance use, which in some cases is followed by a residual change in cognition, affect, personality and behaviour classified as a residual psychotic disorder.

2.2 Neurophysiology

Substance addiction is caused by a dysregulation in motivational circuits, processes of desire, habit formation and reward, leading the individual to favour drug-induced rewards at the expense of natural rewards such as food, sex, and social interaction. Behavioural changes are characterised by strong cravings, a compulsion to use the substance, and a loss of control over the quantity and frequency of use. Compulsive habits are formed, and the individual's impulsivity increases, with changes in the CNS leading to behavioural alteration for repeated administration of the substance. Therefore, new synaptic connections are therefore formed as a result of habit-learning, which predicts future experiences. These connections are being

[1] Prof. Marc Lewis and Dr. Gabor Maté are strong proponents of the BPSS and learning model, arguing that addiction stems from early childhood trauma as a coping mechanism, as a self-medication strategy, which can then become a compulsive, irrational self-destructive habit.

reinforced by every experience of relief in this case by substances. Studies of animal models, as well as brain imaging studies, have helped to further understand the neurocircuitry responsible for the transition from repeated occasional use to drug dependence. There are various ways in which the addiction cycle can be divided, which mainly differ in the way they consider the role of social environment. Classification into three stages including acute intoxication, withdrawal, and craving is prevalent in animal studies (Koob and Volkow, 2010)

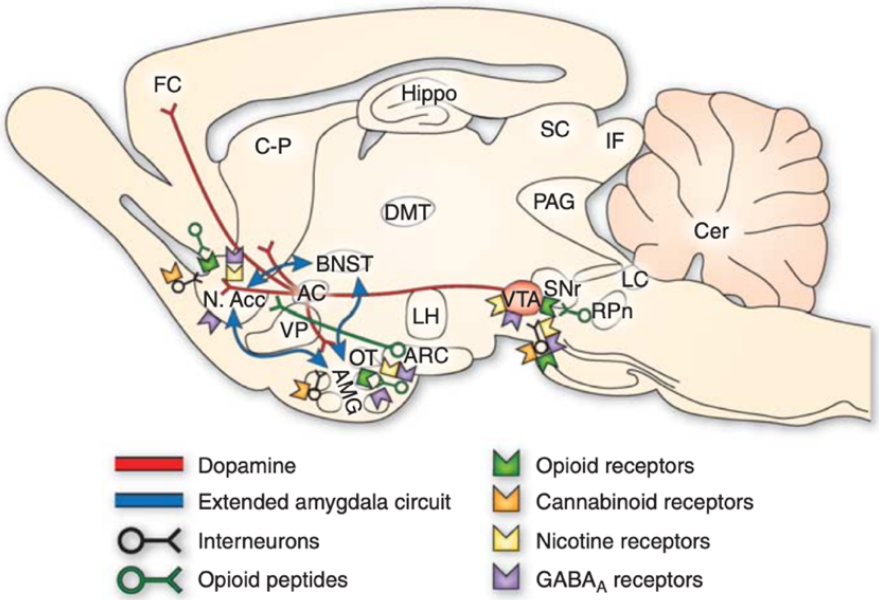
Binge/ acute intoxication

This first phase of the addictive process is mostly regulated by areas in the brain called the basal ganglia. Situated at the base of the forebrain, interconnected with the thalamus and the brainstem. They consist of some key parts: the striatum, the *nucleus accumbens* (NAcc) and the *substantia nigra*. Another key area in the brain is the ventral tegmental area (VTA) located in the midbrain, where dopaminergic cell bodies originate. Through the mesocortical pathway (MCP), the VTA projects to the prefrontal cortex as well as sensory and motor cortices which are involved in motivation, emotion, and executive functions (Fig.1). The prefrontal cortex contains dopamine D_1 and D_2 receptors. These have a higher affinity to DA and are therefore activated at lower constant concentrations, which happens under normal conditions. After acute exposure to a substance, dopamine neurons transmit the signal much faster (phasic firing), leading to higher dopamine levels, thus activating D_1 receptors, which have a lower affinity to dopamine and are key to a reward response. D_1 receptors stimulate reward mechanisms via modulatory pathways in the corpus striatum and memory via the amygdala, medial orbitofrontal cortex (OFC) and hippocampus (Uhl, Koob and Cable, 2019), forming new associations and habits that persist in the face of adverse consequences. A second dopaminergic pathway is the mesolimbic pathway (MLP), where the VTA projects to the *nucleus accumbens* and amygdala (Fig. 1). The NAcc then converts information to motivational action through the connections with motor systems in other brain areas.

Original studies on the pleasure and reward system were conducted by Olds and Milner in 1954, who identified the abovementioned neurocircuitry (the medial forebrain bundle that connects the ventral tegmental area to the basal forebrain). They observed repeated and frequent lever-pressing activity when it was followed by injections of small jolts of current stimulating these areas in the rodent's brains (Olds and Milner, 1954).

In animal models of addiction, the quantification of a described phase can be done using drug self-administration tests, locomotion activity tests and/or conditioned place preference (see Chapter 3 for more details).

Figure 1. Neurochemical circuits involved in drug reward, the red arrow indicates the mesocortical pathway, signalling from the VTA to the prefrontal, cingular, sensory and motor cortexes, the midbrain, the amygdala, and the prefrontal cortex. The blue arrows indicate the mesolimbic pathway, releasing dopamine from the VTA to the NAcc. Source: Neurocircuitry of addiction (Koob and Volkow, 2010)



Involvment of specific receptors and interactions

The most widely studied drugs in preclinical research of addiction are opiates (heroin and morphine), stimulants (cocaine, amphetamine), cannabinoids, and legal drugs such as ethanol and nicotine, which also happen to be the most abused in society (according to 2022 data from globaldrugsurvey.com). The common mechanism of these addictive substances involves the activation and reinforcement of the cortico-limbic dopaminergic circuit (Fig.1) after substances first bind to their specific receptors (Table 1).

Opiates act on mu (μ), delta (δ), and kappa (κ) opioid peptide receptors in the VTA, NAcc and amygdala. These opioid receptors influence the levels of adenosine cyclic monophosphate (cAMP) by inhibiting the activity of adenylyl cyclase. To compensate, the activity of the enzyme increases with a delay, which hinders the effect of morphine and is thought to be the mechanism behind opioid dependence and tolerance (Sharma, Klee and Nirenberg, 1975). Another consequence of opioid receptor signalling is the inhibition of endogenous opioid synthesis by indirectly interacting with proenkephalin messenger RNA (mRNA), which also leads to dependence (Lord *et al.*, 1977) (reviewed in Gupta and Kulhara, 2007). Opioid receptors were shown to be involved in the rewarding properties of not just

opioids but also alcohol (Acquas, Meloni and di Chiara, 1993), nicotine (Berrendero, Kieffer and Maldonado, 2002), and cocaine (Carey *et al.*, 2007).

Stimulants act on dopamine terminals either by inhibiting the DA transporter or by stimulating the release of DA in the NAcc. They also affect serotonin (SERT) and norepinephrine transporters. This was shown using DA deficient (DD) mice for measuring conditioned place preference (CPP) to cocaine (Hnasko, Sotak and Palmiter, 2007).

Cannabinoids bind to G-protein coupled (GPCRs) cannabinoid CB1 receptors, as well as opioid receptors (Tanda *et al.*, 2004) after acute exposure. Although the effects of long-term cannabis exposure leading to dependance are yet to be fully described.

Alcohol acts primarily on gamma-aminobutyric acid ($GABA_A$) receptors in the NAcc and the amygdala, although other systems are also involved. Chronic use of alcohol results in several changes in neurotransmitters, such as decrease in the amount of $GABA_A$ receptors as well as a downregulation of CB_1 receptors (Basavarajappa, Cooper and Hungund, 1998). DA levels were also lower in chronic users and a higher density of D_2 receptors was observed (Rommelspacher *et al.*, 1992).

Nicotine activates the nicotinic acetylcholine (NAch) receptors in the NAcc, VTA and amygdala but in contrast to other substances, the stimulation of dopaminergic mesolimbic neurons lasts only a short period, before the nicotinic receptor shuts down. An interaction with opioid systems was observed as higher levels of adenylyl cyclase and enkephalin mRNA were observed after withdrawal (Houdi, Dasgupta and Kindy, 1998).

Table 1. Substances paired to their targeted receptors, through which their effects are initiated.

Substance	Receptor
Opiates	mu, delta, and kappa opiate receptors
Stimulants	indirect agonists of dopamine
Cannabinoids	CB_1 and CB_2 receptors
Alcohol	GABA agonist and NMDA antagonist
Nicotine	Nicotinic Acetylcholine receptors

Withdrawal/ Negative affect stage

What is considered the second stage of the cycle of dependence, withdrawal, is characterized by feelings of anxiety, dysphoria, stress, and loss of motivation for natural rewards caused by a decrease in dopaminergic and serotonergic transmissions. These are a

result of between-system neuroadaptations as an attempt to neutralize the drug's effects (Koob and Bloom, 1988). Negative reinforcement mechanisms generated by stress and dysphoria are hypothesised to be a result of processes involving the extended amygdala, a brain structure receiving input from limbic structures such as the basolateral amygdala and the hypothalamus. During episodes of withdrawal and stress, the hypothalamic-pituitary-adrenal (HPA) axis mediated by corticotropin-releasing factor (CRF) is activated resulting in an elevation of adrenocorticotropic hormone (Funk *et al.*, 2006).

Conditioned place aversion (CPA) and anxiety response tests are done on rats to determine withdrawal symptoms (see Chapter 3 for more details).

Craving/Anticipation

Craving could be defined as enhanced sensitivity to conditioned cues (stimuli paired with drug-taking) or to stress triggers. During chronic use, a disconnection seems to be created between the prefrontal cortex, responsible for rational decisions, and impulsive behaviour dictated by the ventral striatum, and the *nucleus accumbens*, where the feeling of desire is formed. The feeling of intense desire is mostly mediated by dopamine being released from the midbrain to the NAcc. This pathway is usually called the "reward pathway" although dopamine is not only released after the ingestion of drugs, but rather before, fuelling the feeling of desire and wanting. Two other major circuits are involved in craving, one connects the prefrontal cortex to the NAcc core through glutamatergic signalling, and the other mediates the stress-induced response through activation of corticotropin releasing factor (CRF) and norepinephrine in the basolateral amygdala (Haass-Koffler and Bartlett, 2012). However, precise mechanisms are yet to be explored, given that craving and relapse are not easy to measure clinically. Long-term exposure to drugs of abuse was associated with a lowered level of dopamine D_2 receptors. This decrease in D_2 receptors can lead to a dysregulation of glucose metabolism in the prefrontal cortex (Volkow *et al.*, 1993), leading to learning, memory, and attention impairment as well as impulsive behaviour and compulsive drug administration in addicted rodents (Briand, Gross and Robinson, 2008). Drugs of abuse also increase the threshold for reward during chronic use (Kornetsky, 1979) meaning that nothing can fulfil the craving for drugs other than the substances themselves.

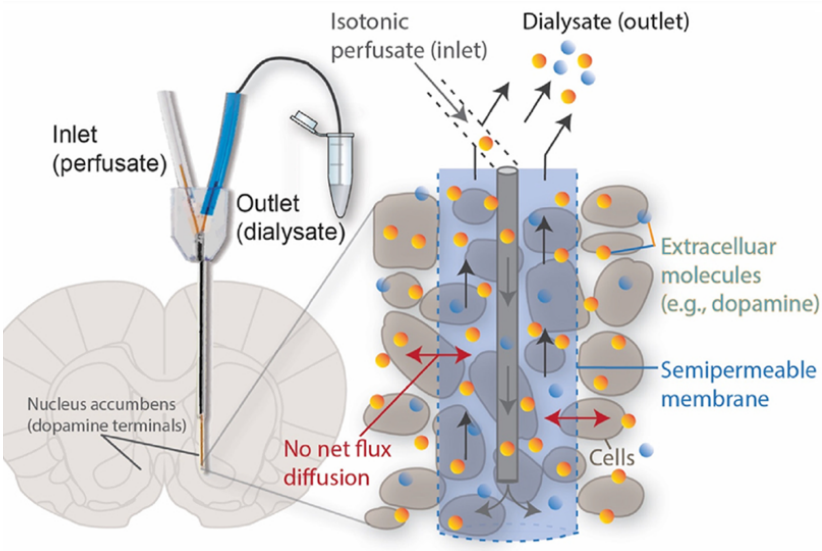
Another factor which cannot be neglected is the increased expression of ΔFosB in neural tissue, as a result of compulsive behaviour. The Fos family of transcription factors are shown to accumulate in neurons in areas associated with addiction, like the NAcc and the dorsal striatum (Nestler, Barrot and Self, 2001). Not just drug addiction but any kind of compulsive

behaviour (compulsive physical activity for example) exhibit these neural changes. Regulation of this genetic abnormality could also lead to a decrease in drug-seeking behaviour. Neurotransmitter concentrations can be examined by electrochemical methods, fMRI imaging or microdialysis (Rusheen *et al.*, 2020). Animal models allow us to measure the changes in various areas of the brain by microdialysis, as the method is quite invasive to be used in human subjects.

Microdialysis

In vivo cerebral microdialysis is an invasive sample collecting method to study the release of neurotransmitters, as it causes some damage to the studied tissue. Despite this, it remains to be highly effective since it allows to analyse more than electroactive substances and the acquisition process is quite simple. Sampling the extracellular fluid from the synaptic cleft, allows measuring the concentration of the monoamines being released (Ungerstedt and Pycock, 1974). The technique consists of inserting a small dialysis catheter (0,2-0,5mm in diameter and 1-2mm long) across the blood-brain barrier into the area of interest. The probe is composed of an outer tube (Fig. 2, in blue) connected to an inner tube. An isotonic perfusate (also called Ringer solution, consisting of NaCl, KCl, $CaCl_2$ and $NaHCO_3$) flows through the inner tube into the tip of the outer tube (Fig. 2).

Figure 2. Microdialysis catheter being inserted in the *nucleus accumbens*. The isotonic perfusate flows through the inner tube towards the outer tube, where neurochemicals cross the semipermeable membrane thanks to a concentration gradient. The dialysate (outlet) is then collected into a vial for further analysis. Source: Evaluation of electrochemical methods for tonic dopamine detection in vivo (Rusheen *et al.*, 2020)



The last 10-30mm of the outer tube consist of a semipermeable membrane which is directly exposed to the extracellular fluid. As the fluid flows through the tube, chemicals enter through the membrane from an area of high concentration to an area of lower concentration (following a concentration gradient). The dialysate (outlet) is then collected into a vial for further analysis. Once the sample is obtained, analytical techniques such as fluorescence detection, enzyme-linked immunosorbent assays (ELISA) or high-performance liquid chromatography can be used to measure the concentration of collected chemicals (Helmy, H. Carpenter and Hutchinson, 2007).

3 The use of animal models in addiction research

Animal models of substance abuse and addiction can vary in their translational validity to clinical situations. Three different categories of model validity can be distinguished including construct, predictive, and face validity. *Construct validity* ensures that the method used matches the theoretical basis of the concept. For example, neurochemical, neuroanatomical, and behavioural mechanisms underlying the voluntary administration of addictive substances in laboratory animals resemble those in humans, which makes these models key tools for understanding the construct of addiction. *Predictive validity* is defined as the measure of how well a model predicts currently unknown aspects of the disease. This is useful in the development of new approaches for human treatment. The *face validity* of a model evaluates how well a model replicates the disease phenotype in humans. Commonly used preclinical models displaying the described types of validity are based on locomotor activity tests (LAT), conditioned place preference (CPP) and drug self-administration (SA).

3.1 Locomotor activity tests

Locomotor activity tests (LAT) are a method used to evaluate the effects of a drug on the animal. By measuring the amount and type of movement under the influence of a substance, it can be evaluated if the effects were stimulating, inhibiting, anxiolytic, or whether the animal was affected at all (Kuhn, Kalivas and Bobadilla, 2019). In an open field test, a special chamber equipped with a light-sensitive detector and light beams can be used, data is then gathered about how much and how often the animal moved. Another way is to gather data from video recordings. Intoxication by psychostimulants like amphetamines or cocaine, for example, leads to heightened locomotor activity and with each dose after first exposure, this locomotor activity increases. This phenomenon is called locomotor sensitization (Valjent *et al.*, 2010).

3.2 Conditioned place preference

Known as CPP, place preference conditioning is a method used to determine whether a stimulus is pleasant or unpleasant. During the experiment, an association is created between a conditioned stimulus, in this case drugs, and an unconditioned stimulus, a context. The experimental box consists of two clearly distinguishable rooms, connected by a central compartment. In the initial phase, the animal has no preference for either room. In the acquisition phase, the animal is exposed to a drug in one of the rooms, creating an association with this context. At the same time, the control group is exposed to a neutral stimulus. During the final, experimental phase, where the animal is no longer exposed to the drug, it is determined whether the experience has been paired with the context by monitoring the length of time the animal has been in the respective chambers. If the time spent in the paired context is longer, then the stimulus was probably pleasurable (CPP), if the time is shorter, then it indicates aversion to the stimulus (conditioned place aversion, CPA) (Cunningham, Gremel and Groblewski, 2006). A similar pairing of drug use to specific contexts is observable in humans, where the unconditioned stimulus is that of a party environment or a specific location/ social group and the conditioned stimulus is that of a drug-induced high. The pairing of these stimuli leads to a conditioned response: craving a drug in that specific environment.

3.3 Self-administration

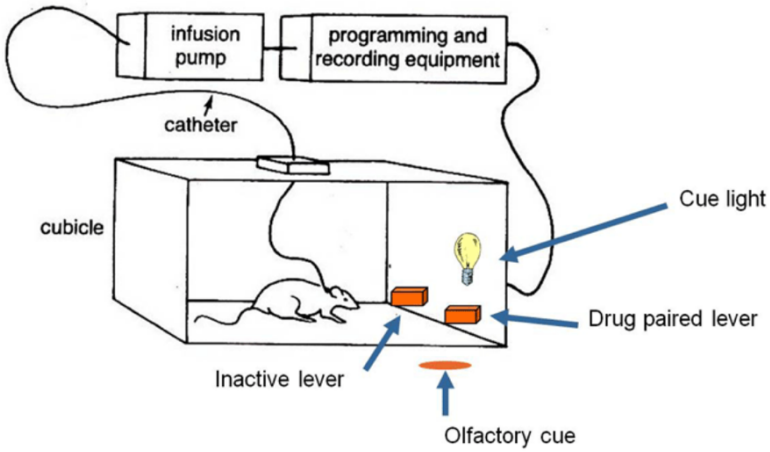
The self-administration (SA) method is used to determine whether the animal wants to continue using the drug after the first intoxication as it has full control over how often the substance is delivered, thus providing high face validity. To self-administrate a drug, the animal must exert some effort, either by pressing a lever ("lever-press") or by inserting their nose into an active opening ("nose-poke"), the substance is then delivered intravenously (IV) by a catheter (Fig. 3) or orally, depending on the type of substance (Panlilio and Goldberg, 2007). SA-based models display an excellent level of face, construct validity and also provide predictive validity (Venniro and Shaham, 2020).

Intravenous self-administration

The chamber used in this experiment, also called the Skinner box after its creator B. F. Skinner, is used to conduct operant and classical conditioning experiments. It contains two levers (Fig. 3), an active lever and an inactive lever. The active lever serves to deliver a fixed dose of a drug directly into the animal's vein through a catheter surgically implanted into their jugular vein (David *et al.*, 2001). The drug is usually a positive reinforcer of behaviour, so every active lever press leads to repeated delivery which increases every day leading to

behavioral sensitization (Venniro and Shaham, 2020). Skinner boxes then contain a system for recording the animal's response (Fig. 3). The box can also be programmed to make it progressively more difficult for the subject to obtain the substance (progressive ratio SA). The impairment is achieved by setting a so-called fixed ratio (FR: the number of correct responses to the release of one unit of reinforcer). First, each correct response by the animal leads to an infusion of the substance, then after each trial/ number of sessions, the ratio is increased. Substance use is thus made more difficult: the animal must press the lever twice, three times or more to obtain a single dose of the drug, which is usually done to measure how hard the animal is determined work to receive a dose. Overdose is avoided by having a so-called time-out period after the dose is injected, where the injection of the substance is followed by a time frame, when the animal can respond but no reward occurs. Another way to prevent overdose is to set a limit on infusions for the experimental day (Venniro and Shaham, 2020).

Figure 3. The operant conditioning chamber, developed by B.F. Skinner, also known as the Skinner box. The infusion pump delivers a fixed amount of a drug through the catheter directly into the animal's vein after pressing the active lever. The rewarding properties of a drug can also be paired with a light or olfactory cue (conditioned stimuli). The programming and recording equipment then serve to record data about the frequency of lever pressing or nose-poking activity. Source: drsimonsaysscience.org



Behavioural sensitisation

Behavioural sensitisation means the reinforcement of specific drug-induced behaviours after repeated exposure to a constant dose of a given substance. In rats, the memory that is formed after drug administration appears to be very stable and sensitization can persist for months (reviewed in Robinson and Becker, 1986). Behavioural sensitisation may not only be caused by a substance, but also by increased stressful situations, olfactory or light cues (Fig.3) or by changes in the social environment of animals, leading to substance

seeking (Kuhn, Kalivas and Bobadilla, 2019). Improvements in living conditions and social environment probably lead to a reduction in drug-seeking related behaviour as shown in the popular experiment from the 1970s also called "Rat Park"[2] led by professor B.K.Alexander (Alexander, Coambs and Hadaway, 1978).

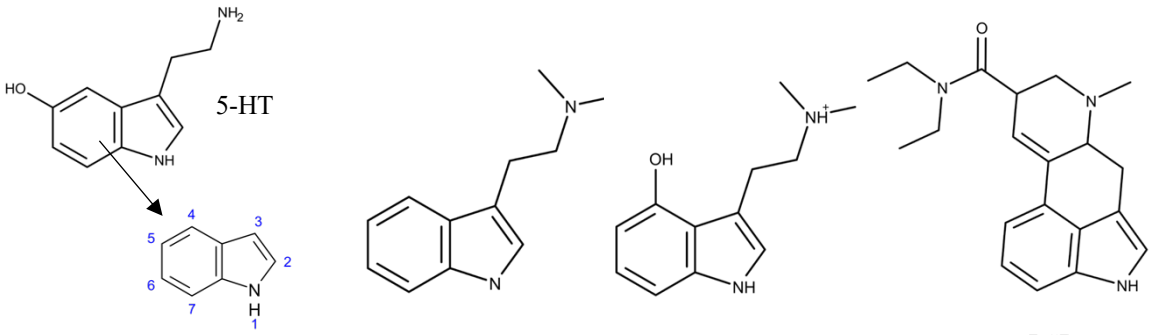
4 Pharmacology and mechanism of action of chosen psychedelics

Among those most studied in the context of drug addiction treatment are serotonergic psychedelics, which include tryptamine derivatives such as N,N-Dimethyltryptamine (DMT), psilocybin and the ergoline derivative lysergic acid diethylamide (LSD). These have a similar mechanism of action and have been used for both clinical and animal studies for longer. The atypical psychedelic ibogaine is also among the most promising in the treatment of mental disorders. Other hallucinogenic substances include the phenylethylamine mescaline, the dissociative anesthetic ketamine and the empathogen MDMA. The last two have a shorter history and the data on their potential use in clinical research is just emerging. The effects of classic psychedelics are mediated mainly by 5-HT receptors, and other receptors like the D_2 in the case of LSD and mGlu2 in the case of DMT.

4.1 The role of 5-HT receptors

Serotonergic psychedelics share an indole nucleus with 5-hydroxytryptamin (5-HT, also known as serotonin), which is the basis of their binding similarity.

Figure 4. Structure comparison of serotonin with its indole nucleus, DMT, psilocin and LSD. Source: psychedelicreview.com

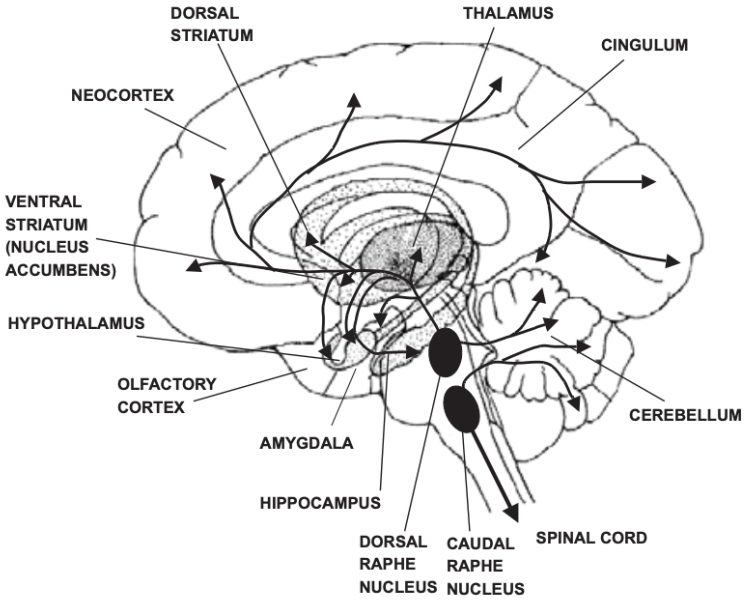


Indole nucleus DMT Psilocin LSD

[2] „Rat Park" was used in to prove the hypothesis that drug consumption could be related to the setting the rats were in during self-administration experiments. Rats had constant access to morphine and cocaine but instead of being isolated in a cage, they were in a community of rats where they were free to interact, socialize, play, and have sex. Rats placed in "Rat Park" voluntarily stopped taking morphine, showed lower rates of compulsive use of drugs, and had no cases of overdose.

Most 5-HT receptors are metabotropic G-protein coupled (GPCR) serotonin binding receptors (apart from the 5-HT_3 receptor, which is a ligand gated ion channel, and is not involved in the action mechanism of psychedelics). Serotonin is produced in neuronal bodies located in a region of the midbrain called the dorsal raphe nucleus (DRN) (Fig.5).

Figure 5. Pathways of 5-HT in the brain. serotonergic neuronal cell bodies are mainly located in the dorsal raphe nuclei (DRN) of the brainstem and from there project serotonin to other regions of the brain, indicated by the black arrows. A high density of 5-HT_2 can be found in the cerebral cortex area and in several other regions of the brain including the olfactory cortex, *nucleus accumbens* and the amygdala, Source: (Bombardi, 2014).



The main types of 5-HT receptors involved in signalling pathways affected by psychedelics are 5-HT_{1A}, 5-HT_{2A} and 5-HT_{2C}. Early experiments measuring extracellular fluid monoamines in the DRN found an increase in serotonin concentration after injection of LSD, thus revealing an inhibitory effect of LSD on 5-HT neurons in the DRN (Aghajanian, Foote and Sheard, 1968). Later the same effect was observed with tryptamines inhibiting presynaptic 5-HT neurons (Aghajanian and Hailgler, 1975). This effect was credited to the high concentration of 5-HT_{1A} in the DRN, proving that LSD has an agonistic effect on the receptor. This receptor is correlated with some aspects of the hallucinogenic effects of psychedelics, which was proved by 1) Stimulus control (tests, in which the behavior of animals is observed under the influence of a substance) by LSD was increased when 5-HT_{1A} agonists were introduced and 2) 5-HT_{1A} antagonist WAY-100,635 blocked the drug-

appropriate responding (Reissig *et al.*, 2005). However, the main candidates for the hallucinogenic effects of psychedelics appear to be 5-HT_{2A} and 5-HT_{2C} receptors.

5-HT_{2A} receptor

The dominant role of the 5-HT2A receptor was proven by several experiments using potent 5-HT_2 agonists, such as 2,5-dimethoxy-4-methylamphetamine (DOM), and several 5-HT_2 antagonists: 1) When micromolar doses of 5-HT_2 antagonists ketanserin and pirenperone were administered prior, an attenuation of stimulating hallucinogenic effects of both DOM and LSD has been observed[3] (Colpaert, Niemegeers and Janssen, 1982; Glennon, Titeler and McKenney, 1984). 2) Pretreatment of neurons with MDL 100907 (a strong 5-HT_{2A} antagonist) before application of LSD and DOI, resulted in the decreased firing of neurons by 19% compared to controls without the antagonist (Marek and Aghajanian, 1996). 3) A similar effect was noticed after administration of the 5-HT_{2A} antagonist Risperidone[1] (Meert, de Haes and Janssen, 1989). 5-HT_2 binding was later confirmed after constructing and analysing crystal structures of 5-HT_{2B}R/LSD complexes (homologous to both 5-$HT_{2A/2C}$) (Wacker *et al.*, 2017).

5-HT_{2C} receptor

Some evidence shows the 5-HT_{2C} to be a key receptor in modulating the anti-addictive properties of psychedelics (reviewed in Canal and Murnane, 2017). Recent behavioral studies exhibited a decrease in self-administration and addictive behavior after injection of 5-HT_{2C} agonists (Lorcaserin and Ro 60-0175), while the opposite was shown for antagonist SB242,084. Lorcaserin (2.3mg/kg) was administered to rhesus monkeys 90 minutes before a self-administration session with both cocaine and methamphetamine, and in both cases resulted in reduced reinforcing effects of the drugs and fewer infusions (Gerak, Collins and France, 2016). In another study, the 5-HT_{2C} agonist Ro 60-0175 (1mg/kg) reduced drug-seeking behavior after both acute and chronic administration in monkeys (Rüedi-Bettschen, Spealman and Platt, 2015). The 5-HT_{2C} specific antagonist SB242084 (0,5mg/kg) was shown to enhance cocaine-induced behavior in rats, suggesting that whether the 5-HT_{2C} is blocked (by an antagonist) or activated (by an agonist), the addictive effects of drugs change.

[3] Both were done using rat drug discrimination methods, where the rodent is trained to recognize the effects of specific substances and doses by pressing a lever in response to the effects of the administered drug.

The activation of the 5-HT_{2C} receptor thus makes classic hallucinogens non-addictive and in addition to their psychological effects, it may therefore contribute to their potential in drug addiction treatment.

Changes in brain connectivity

The increased signalling through serotonergic synapses by LSD causes increased connectivity of the thalamus to cortical areas (the cortico-striatal-thalamic loop circuit) as was shown in recent human fMRI studies (Preller *et al.*, 2019). These are the same brain areas that control habit formation, reward, and compulsive behaviour and are also involved in the regulation of consciousness. Another area affected by LSD is the default mode network (DMN), which is the connection of the posterior cingulate cortex (PCC), medial prefrontal cortex (mPFC) and angular gyrus. The DMN, a neural network active when the brain is at rest, not engaged in any task, is also involved in daydreaming, and was shown to be involved in certain psychiatric conditions such as depression or ADHD (Whitfield-Gabrieli and Ford, 2012). Lower activity of the DMN is observed when the brain is actively engaged in thinking and paying attention. Acute administration of LSD was found to decrease the functional connectivity of this network, leading to a state associated with the feeling of "ego-dissolution[4]" (Müller *et al.*, 2018). A study using fMRI done by R. Carhart-Harris showed a decrease in functional connectivity and activity of the mPFC and the PCC (which form the so-called default mode network, DMN) also after psilocybin intoxication, leading to a state of rampant consciousness and ego-dissolution, similar to LSD (Carhart-Harris *et al.*, 2012).

4.2 LSD

Lysergic acid diethylamide belongs to the Ergoline family of substances, alkaloids derived from the ergot fungus *Claviceps purpurea*. The compound, labelled LSD-25, was first isolated in 1938 as the twenty-fifth derivative of lysergic acid (LSA) by Albert Hoffmann as one of a series of compounds which for a long time were deemed uninteresting. In 1945 the psychedelic effects of LSD were discovered by Hoffman himself and since then it has been listed as the most potent psychedelic. Some of the physiological effects of hallucinogens including LSD are increased blood pressure and energy, dilated pupils, sleeplessness, loss of

[4] Ego dissolution can be described a distortion in the sense of "self" and is typical for higher doses of psychedelics. Self-identity is replaced by a sense of union and connection to the outside leading the individual to view their own thoughts and behaviours from a different perspective. These are also described in books about altered states of consciousness by respected authors within psychedelic literature like Timothy Leary (The Psychedelic Experience: A Manual Based on the Tibetan Book of the Dead, 1964), Stanislav Grof (Realms of the Human Consciousness: Observations from LSD, 1976) and Aldous Huxley (The Doors of Perception, 1954).

appetite, distortion of time and space perception and impaired psychomotor functions (Abramson *et al.*, 1955). The subjective experience also depends on dosage, the subject's anatomy, physiology, mental state (set), environment (setting) and route of administration. The routes of administration of Lysergic acid include intravenous injection, transdermal application, inhalation, and the most common, sublingual. The effects are highly dose-dependent, lasting up to 12 hours (8,2 hours after a dose of 100µg, while after double the dose 11-12hours) (Dolder *et al.*, 2017). Maximal plasma concentrations of LSD were observed in healthy individuals after 1,5 hours (median), and the half-life of the compound was found to be around 3.6 hours while being detectable up to 12 hours following oral administration (Dolder *et al.*, 2016). Absorption of LSD through the stomach and intestines happens almost immediately, it is then metabolised by liver Cytochrome P450 enzymes to its most common metabolites 2-oxo-3-hydroxy LSD (OH-LSD) and 6-norlysergic acid diethylamide (nor-LSD) (Luethi *et al.*, 2019).

The underlying neurobiological mechanism involved in substance (specifically alcohol) dependence treatment is likely the modulation of the mesolimbic dopamine pathway (MLP) through the 5-HT receptor agonism of LSD. LSD acts as an agonist at 5-HT_{2A} and 5-HT_{2C} receptors, as well as D_1 and D_2 dopaminergic receptors (Watts *et al.*, 1995). Specifically, the action on 5-HT_{2C} appears to inhibit tonic dopaminergic transmission in the frontal cortex (Millan, Dekeyne and Gobert, 1998). As another paper using DOI (a preferential 5-HT_{2A}R agonist) and mCPP (a 5-$HT_{2C/1B}$R agonist) also shows, specific activation of 5-HT_{2A} and 5-HT_{2C} (with 5-HT_{1B}) suppresses operant ethanol self-administration in rats (Maurel, de Vry and Schreiber, 1999).

4.3 Tryptamines

Also referred to as indolamines, tryptamines are substances with a similar structure to serotonin (5-hydroxytryptamine). The most common are N,N-dimethyltryptamine (DMT) (Fig. 4), psilocin (5-hydroxy DMT) (Fig. 4) and 5-MeO-DMT (5-Methoxy-DMT).

Psilocin

Psilocin is the active metabolite of psilocybin (3-[2-(Dimethylamino) ethyl]-1*H*-indol-4-yl dihydrogen phosphate), a compound found in hallucinogenic ("magic") mushrooms mainly of the genus *Psilocybe* and *Conocybe*. After oral ingestion (the most common route of administration), psilocybin is dephosphorylated in the blood, kidney, and intestine by alkaline phosphatase into psilocin, which can easily cross the blood-brain barrier (Horita and Weber,

1961). Psilocin is mainly responsible for the hallucinogenic effects, which come up after 20-40 minutes, peak after about 70-90min and last for 4-6 hours after a full dose of 8-25mg of psilocybin (Hasler *et al.*, 2002).

Dimethyltryptamine

N,N-Dimethyltryptamine is a short-acting entheogen[5], a plant indole alkaloid and an endogenous neurotransmitter in animals. Together with its methoxylated form, 5-MeO-DMT, it can be found in various plant genera including *Acacia, Delosperma, Virola,* and *Psychotria*. The use of these hallucinogenic plants can be traced back to ancient native american shamanistic rituals (Miller *et al.*, 2019) in the form of Ayahuasca, a mixture of *Banisteriopsis caapi* and *Psychotria viridis* as DMT produces very intense visual effects, and sensory, and auditory distortions accompanied by feelings of ego dissolution, unity, and interconnectedness. DMT is mainly consumed by inhalation of free-base DMT (50-100mg), intravenously, or orally in the form of a brew called Ayahuasca (containing usually 24mgDMT/100ml). 5-MeO DMT can be found in the venom of the toad *Incilius alvarius* along with bufotenine (5-HO-DMT). DMT was also found to be endogenously synthesized in rat and human brains by a recent study (Dean *et al.*, 2019) that showed: 1) The key DMT synthetizing enzyme transcript, indolethylamine-N-methyltransferase (INMT) mRNA, was detected via *in situ* hybridization in rat and human cerebral cortex, pineal gland and choroid plexus, 2) Rat brain microdialysate was quantified and DMT was detected in similar concentration to other monoamines in rat cerebral cortex, 3) DMT levels increased significantly in rat brains following cardiac arrest.

DMTs metabolism is very similar to that of serotonin: monoamine oxidases, as well as peroxidases, initiate the metabolism, producing a series of compounds including NMT, 6-OH-DMT, DMT-NO, IAA, none of which produce the same hallucinogenic effects as DMT (Szára, 1956). Because of this metabolic pathway, DMT is often consumed along with MAO inhibitors[6], which cause an increase in plasma levels and accumulation of tryptamines in the CNS (Halberstadt, 2016).

[5] Entheogen is a term used for psychedelic or hallucinogenic substances used in religious and spiritual contexts, for their ineffable mystical-like effects like transcendence, awakening and connection to a higher self. Plants used as entheogens vary between regions, among Native Americans these include *Psychotria viridis* in the Ayahuasca brew, Peyote cactus *Lophophora williamsii*, *Psilocybe* mushrooms and Tobacco leaves (Miller *et al.*, 2019).

[6] For example the Ayahuasca brew is prepared with a mixture of leaves from *Psychotria viridis* containing DMT and *Banisteriopsis caapii*, containing MAOIs. This allows the medicine to come up slower (within 60-90 minutes) and lasting longer, while peaking after about 1,5-2 hours (Riba *et al.*, 2003).

Dimethyltryptamine mainly targets 5-HT_{2A} with a less prominent action on 5-HT_{2C} and mGlu2 receptors, shown by rat drug discrimination and radioligand methods: 1) MDL100907 (a $5HT_{2A}$ inverse agonist) and ketanserin, both blocked the effects of DMT (Smith, 1998) and 2) SB242084 (a 5-HT_{2C} antagonist) and LY341495 (an mGluR2 antagonist) only partially modulated the discriminative stimulus effect of DMT and another tested tryptamine compound, DiPT (Carbonaro *et al.*, 2015). In humans, 5-HT_{1A} receptor agonism also plays a role in the effects, as pindolol (a 5-HT_{1A} agonist) pre-treatment increased the psychological response to DMT in tested subjects (Strassman, 1995). Other targeted receptors are SERT, and VMAT-2 (both serotonin transporters) as DMT was shown to inhibit the transport of 5-HT, while acting as a substrate, thus allowing the accumulation of tryptamines within neurons enough to activate sigma-1 receptors[7] (Cozzi *et al.*, 2009). Higher concentrations of DMT seem to cause the translocation of sigma-1 receptors, thus inhibiting membrane ion channels (Su, Hayashi and Vaupel, 2009). Lastly, the psychedelic effects of DMT are thought to be mediated partly also by trace amine- associated receptors (TAARs), which are G-protein coupled receptors activating adenylyl cyclase and cAMP accumulation, on which DMT acts as an agonist (Bunzow *et al.*, 2001).

4.4 Ibogaine

The indole alkaloid ibogaine is isolated from the root of the *Tabernanthe iboga* shrub. In higher amounts, ibogaine, along with other alkaloids, causes hallucinations and a highly introspective altered state of consciousness, which makes it a promising substance for the treatment of mental disorders like depression or addiction.

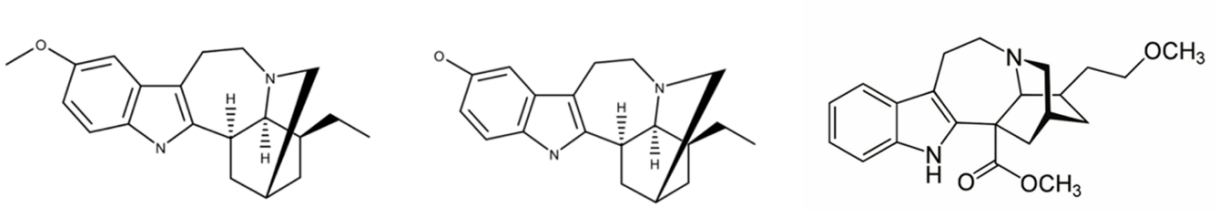
The original use can be traced to west Africa, specifically around Gabon; in small quantities, *iboga* root cuttings were used by the inhabitants as a mild stimulant to fight off hunger, thirst, and fatigue. In higher doses, the crushed root was part of initiation rituals and spiritual practices of the African *Bwiti* religion. In these rituals, *iboga* served to deepen the spiritual experience, allowing visions and contact with the dead, as well as experiencing one's own death and rebirth (Nyongo Ndoua and Vaghar, 2018). In 1901, the alkaloid was first isolated from the root of Iboga, and during the first half of the 20th century it was further researched and used as a mild stimulant, even being sold in tablet form in France under the name *Lambaréne*. However, in the 1960s, after the popularization of its hallucinogenic properties, it was banned, like other psychedelic substances.

[7] Sigma-1 are mitochondrion-associated endoplasmic reticulum membrane receptors, widely studied for their important role in modulation of synaptic plasticity and thus protective role in neurodegenerative diseases (Ryskamp *et al.*, 2019).

After administration, the plasma concentration of ibogaine peaks after two hours and has a half-life of around 4-7 hours in humans and 1 hour in rats (Mash et al., 2000). After ingestion, it metabolizes in the liver by O-demethylation into O-desmethylibogaine, also called noribogaine (Fig. 8.). Noribogaine lasts up to 19 hours after subcutaneous injection and is thought to be responsible for the long-lasting changes in consciousness (Bhargava and Cao, 1997). Additionally, it was also found that ibogaine can last longer than 12 hours stored in adipose tissue, this leads to the hypothesis that a single dose of ibogaine could also cause long-lasting therapeutic effects (Hough, Pearl and Glick, 1996). The derivative of ibogaine, 18-methoxycoronadirine (18-MC) (Fig.6) has also been subject to additional research to determine the metabolism and action on reward pathways. It has comparable effects to ibogaine in terms of reducing self-administration in the rat, but its effects appear to be less hallucinogenic (Glick *et al.,* 2000).

The exact pharmacodynamic action of ibogaine is not yet fully understood, as its action is complex and acts through many different receptors. It is classified as an atypical hallucinogen, for its action directly and indirectly on the dopaminergic, glutamatergic (NMDA), serotonergic, opioid, nicotinic, aminobutyric acid (GABA), cholinergic, and muscarinic receptors (Popik and Skolnick, 1999). The effects of ibogaine are thought to be produced by a combination of interactions with these systems.

Figure 6. Chemical structures of ibogaine, noribogaine and 18-MC. Source: psychedelicreview.com



Ibogaine Noribogaine 18- methoxycoronadirine

Targeted receptors

The chemical structure of ibogaine includes an indole nucleus, a common feature with serotonin. As shown prior, the main 5-HT receptors which interact with psychedelic compounds are the serotonin binding G-protein coupled receptors (GPCRs). In vivo studies done on rats show that after repeated administration (40mg/kg), ibogaine causes an increase of extracellular serotonin in the *nucleus accubens* (NAcc) (Wei *et al.*, 1998). The compound has been shown to inhibit the reuptake of serotonin on the 5-HT transporter (SERT) resulting

in serotonin staying in the synapse longer instead of being recycled into presynaptic neurons (Wells, Lopez and Tanaka, 1999). Data from cryo-electron microscopy show ibogaine binding to both outward-open and inward-open conformations of SERT, stabilizing the state of the transporter from which serotonin is released into the cytoplasm (Coleman *et al.*, 2019). An increase in synaptic 5-HT is shown to lower self-administration and drug-seeking in rats, thus leading to the hypothesis that this may be one of the possible mechanisms by which ibogaine could treat drug dependence. 18-MC on the other hand seems to only increases dopamine levels, so it probably has lower affinity to the 5-HT transporter, explaining its lower hallucinogenic properties (Glick *et al*., 2000). Because studies on the effect of ibogaine on 5-HT receptors haven't determined a specific mechanism of action yet, as with most psychedelics, further scientific research may be needed.

Because dopamine is one of the main neurotransmitters causing behavioural sensitisation and an increase in drug-seeking behaviour, much research is done examining the effect of ibogaine on mesolimbic dopaminergic pathways. Increases in extracellular dopamine in the *nucleus accumbens* (NAcc) in response to drug intoxication led to their positive rewarding effects so when the amount of dopamine is decreased, drug craving is lessened. The role of ibogaine in this process is that of attenuation of the rise of DA in dopaminergic neurons in the mesolimbic pathways (so instead of phasic DA firing we see a return to normal, tonic firing) (Maisonneuve *et al.*, 1997).

To determine the effect of ibogaine and other iboga alkaloids on the release of dopamine in rats, levels of DA metabolites were measured, specifically dihydroxy-phenylacetic acid (DOPAC) and homovanillic acid (HVA) either from dead brain tissue or from extracellular fluid (Glick and Maisonneue, 1998). By using these methods, it was found that both ibogaine and noribogaine cause acute reduction in tissue dopamine and an increase in both its metabolites thus regulating DA metabolism. Extracellular levels of DA in the NAcc are lowered by both ibogaine and its derivative 18-MC (Maisonneuve *et al.*, 1992). Dopamine transporters appear not to be directly affected by ibogaine, so changes in dopamine levels are mostly mediated by different receptors, such as 5-HT, nicotinic, opioid and NMDA (Baumann *et al.*, 2001).

Three types of opioid receptors are involved in the mesolimbic pathway (the brain circuitry modulating drug-seeking behavior and reward): the mu (μ), delta (δ), and kappa (κ) receptors. Activation of the μ- receptor by opioids (agonists) increases the extracellular level of dopamine in the *nucleus accumbens*, leading to euphoria, positive reinforcement and therefore drug addiction (Contet, Kieffer and Befort, 2004). The κ- receptor is associated with

the regulation of reward, mood processing and dysphoria and the activation causes a decrease of dopamine in the NAcc (Mysels, 2009). Ibogaine typically binds to all μ-, δ-, and κ-receptors with low affinity, acting mainly as a μ- opioid receptor agonist (Codd, 1995). However, its metabolite, noribogaine has a significantly higher affinity to κ- receptors making it an effective κ- opioid receptor agonist, while acting as a weak μ- receptor antagonist (Maillet *et al.*, 2015). The derivative 18-MC acts as both a κ- and μ- antagonist, which also leads to changes in the level of extracellular dopamine (Glick et al., 2000). The complex antiaddictive properties could therefore be attributed to the combination of these three compounds, rather than just one of them. Ibogaine acts also on nicotinic acetylcholine and glutamatergic (NMDA) neurons. It is thought that the signalling on these receptors also contributes to a reduction in the frequency of self-administration and bingeing, although the specific mechanisms of action are still not fully elucidated (Popik and Skolnick, 1999).

5 Preclinical research

Studies on the effects of psychedelics on human subjects show relevant results, although it is hard to dissect to what extent the results are just a result of a psychedelic "peak experience"[8] and how much of an underlying biologic determinant there is. Preclinical studies are performed in a laboratory environment which allows to control for as many variables as possible. This is where animal models come into the picture, as they could elucidate the neurological and physiological changes occurring subsequently to substance consumption regardless of the psychological effects.

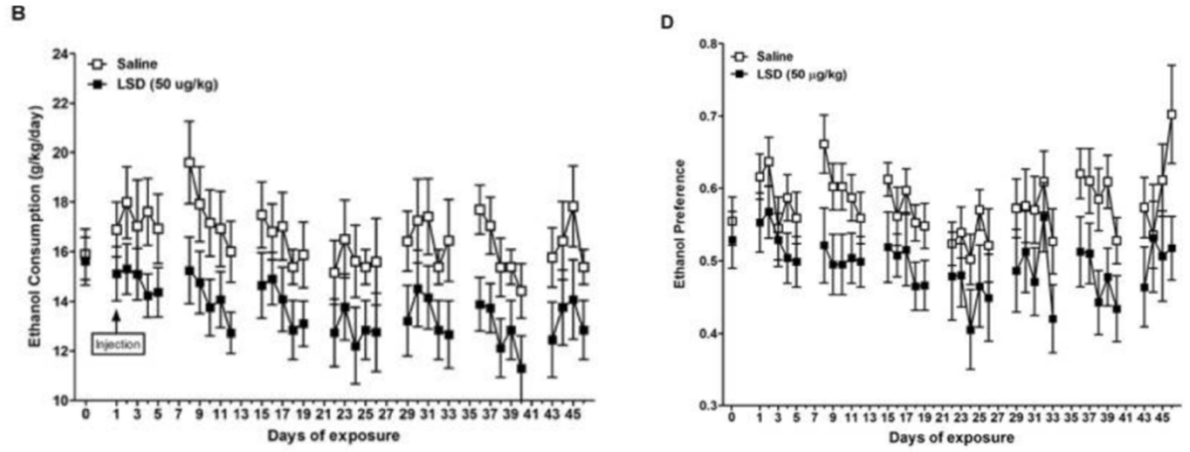
5.1 LSD

The clinical evidence of LSD being effective in alcohol consumption cessation is extensive, although the effect on animals has yet to be further explored. 1) The following recently published paper investigates the effects of a single dose of LSD on an alcohol-preferring strain of mice (Alper *et al.*, 2018). The C57BL/6J inbred strain of mice is known for its high levels of alcohol consumption, which makes it a reliable phenotype for research on alcoholism (Melo *et al.*, 1996). This study used a self-administration model in which mice were caged separately and exposed to two bottles, one containing water, the other one 20%

[8] A Psychedelic peak or mystical experience, often compared to a near-death experience, happens on higher doses of potent psychedelics in the right setting. Such event allows individuals to change their view of themselves and their perspective on life itself. Such a strong insight can cause subjects to improve their lifestyle choices and as a result, overcome their addictions and traumas. As Stanislav Grof describes in his book "LSD psychotherapy".

ethanol. After four weeks of habituation, they were divided into three groups (8-10 rats per group) based on the amount of alcohol consumed and were administered an intraperitoneal injection of either saline, 25µg/kg LSD or 50µg/kg LSD. Water and ethanol consumption was then measured every 24 hours for 46 days following treatment. The group treated with 50µg/kg LSD resulted in a reduction of 18% in ethanol consumption (p=0,0035; Fig.7B) and ethanol preference (ethanol intake/ total fluid intake) (p=0,0024; Fig. 7D) which was sustained for the following 46 days compared to the control group. Alcohol consumption has also never reached equivalence with the consumption of the control group. In contrast, the 25µg/kg LSD has shown equivalence within 23 days of treatment, while alcohol preference was not significantly different. These results, indicating a reduction in alcohol preference in a rodent model, bring hope for LSD as an effective treatment option for alcohol abuse.

Figure 7. Ethanol consumption (B) after 50µg/kg LSD treatment showing a significant decrease over the course of 46 days as well as ethanol preference (D). Source: LSD administered as a single dose reduces alcohol consumption in C57BL/6J Mice. Source: (Alper *et al.*, 2018)



2) A later study from 2020 on an alcohol relapse rat model (alcohol deprivation effect ADE) was conducted. This model uses repeated alcohol deprivation phases followed by renewed access in order to test compulsive relapse-like drinking (Meinhardt *et al.*, 2020). In normal circumstances, after a period of deprivation, the amount of alcohol self-administration temporarily increases (fig. 8) and then returns to baseline. LSD's role in relapse prevention was tested with the introduction of an LSD treatment after a four-week deprivation period and then free access to alcohol. The results showed no difference between the groups tested with the vehicle solution and groups tested with 80µg/kg LSD and 350µg/kg LSD over the course of five days (fig. 8). There is no explanation for the above-mentioned

results so far, (as well as findings by Alper et al., 2018) as preclinical research on animal models is limited to these two papers.

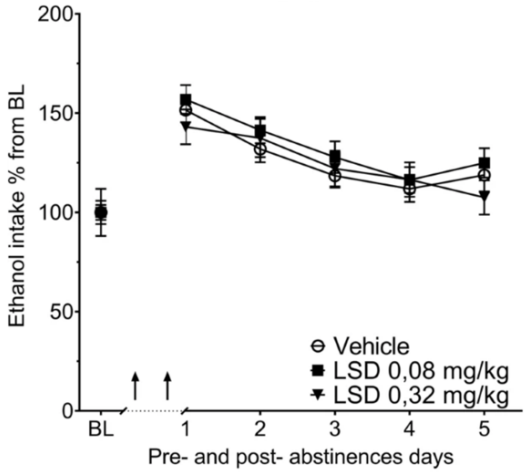


Figure 8. BL: baseline drinking, arrows indicate the administration of either vehicle or LSD. Consumption of ethanol reached a level of 150% after a period of forced abstinence and hasn't changed after treatment with high doses of LSD compared to controls over the course of the following 5 days. The same results were observed with psilocybin treatment.
Source: (Meinhardt *et al.*, 2020)

Most controlled clinical trials come from the 1960s, as reviewed in a 2012 meta-analysis (Krebs and Johansen, 2012), in which a single dose of LSD lead to a significant reduction in alcohol use for several subsequent months (536 subjects were involved). The latest evidence for human alcohol consumption reduction after use of psychedelics comes from a recent meta-analysis using an online survey, where subjects reported a reduction in alcohol use after moderate to high doses of LSD (Garcia-Romeu *et al.*, 2019).

5.2 Tryptamines

Research on DMT is often done through the hallucinogenic brew Ayahuasca, as the effects usually last longer. A few papers on Ayahuasca have been published in the last decade, looking at self-administration and CPP for alcohol and amphetamines in mice and rats. 1) A study that looked at stimulant induced CPP (Reis *et al.*, 2020) found that the group that received an ayahuasca treatment (0,4mgDMT/ 100mg) after the conditioning period with methylphenidate (also known as Ritalin; a similar stimulant to amphetamine) showed significantly reduced methylphenidate induced CPP (Fig. 9a) (Reis *et al.*, 2020). This paper also looked at the expression of Fos (the transcription factor that accumulates in the NAcc and dorsal striatum, usually after repeated exposure to extremely rewarding substances), which is normally higher after methylphenidate ingestion, but when pretreated with Ayahuasca, the expression of Fos was blocked. 2) Another paper (Cata-Preta *et al.*, 2018) showed that co-administration with ayahuasca (100-300mg/kg) reduced the induced preference for the alcohol-paired compartment (Fig. 9b). 3) Ayahuasca was also shown to inhibit the development of alcohol-induced behavioural sensitisation in mice (Oliveira-Lima *et al.*,

2015). 4) Self-administration of methamphetamine was studied by Godinho in 2017. Pretreated mice with 2ml/kg ayahuasca showed a reduced preference for amphetamine-laced water and a reduction in amphetamine-induced anxiety (using an elevated plus-maze and looking at the time spent in the closed arms) (Godinho, 2017).

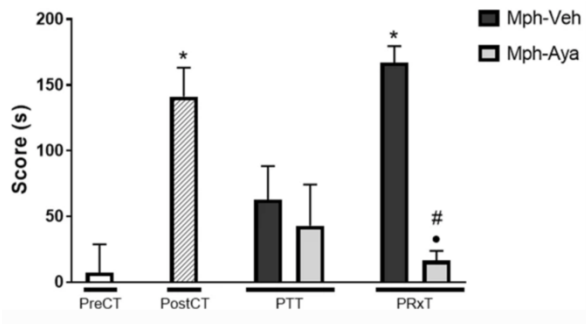


Figure 9a Results of treatment with Ayahuasca on methylphenidate induced CPP. Pre/Post CT- preconditioning/ postconditioning; PTT-posttreatment test after ayahuasca treatment; PRxT- postreexposure test after methylphenydate reexposure. Source: (Reis *et al.*, 2020)

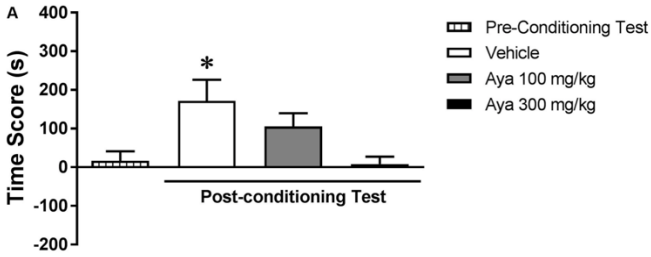


Figure 9b Ayahuasca in moderate and high doses reduced the preference for the compartment previously paired with alcohol consumption.
Source: (Cata-Preta *et al.*, 2018)

The potential of psilocybin for addiction treatment is severely under-studied in animals, in contrast to the promising ongoing clinical trials. There is evidence for the potential of 5-HT_{2A} and 5-HT_{2C} agonists, which include psilocin and other psychedelics, playing a role in nicotine cessation (Higgins and Sellers, 2021), although there is only one preclinical paper looking at the specific effects of psilocybin itself. Although this study looked at the effects of both high doses of psilocybin (1mg/kg) and sub-hallucinogenic microdoses over the course of 4 weeks, it showed no significant differences compared to control groups (same as in fig. 8) (Meinhardt *et al.*, 2020).

5.3 Ibogaine

The idea of using ibogaine in addiction treatment was first popularized by the American Howard Lotsof after he and seven other heroin users ingested a high dose of ibogaine and noticed a decline in their opiate use over the following 6 months. The preclinical research on the effectiveness of *iboga* alkaloids started in the 1990s and has since been extensive (Belgers *et al.*, 2016). The most frequently used models are rat and mice self-

administration of three categories of drugs of abuse: opioids, alcohol and nicotine, and stimulants. Currently, the ibogaine subculture is expanding with addiction treatment centres (mostly in the US) and by ongoing preclinical and clinical trials (Alper, Lotsof and Kaplan, 2008). Ibogaine also shows promising results in the attenuation of symptoms of depression which when untreated can lead to substance abuse. As shown in a very recent study, dose and time-dependent antidepressant effects were observed in rats after noribogaine administration (Rodríguez *et al.*, 2020).

Opioids

Ibogaine has been shown to alter the reinforcing power of opioids, which was studied on the self-administration of morphine, looking at both acute and long-lasting effects. 1) A study done on female Sprague-Dawley rats observed a reduction in morphine intravenous SA; with some rats, the effects lasted several weeks and were significant after just one pre-treatment with 40-80mg/kg ibogaine, some needed three or more doses (in weekly intervals) to show prolonged effects. Most rats though exhibited a decline in morphine intake, regardless of individual differences (Fig. 10a) (Glick *et al.*, 1991). 2) A successive paper, testing several *iboga* alkaloids on morphine self-administration, also showed very similar results: rats pre-treated with 40mg/kg ibogaine showed a prolonged (tested for 27 days) reduction in morphine injections after three weekly sessions and acute reduction (Glick *et al.*, 1994). In the same study, it was observed that *iboga* alkaloids (R-ibogamine and R-coronaridine) significantly reduced phasic firing of DA in the NAcc and striatum. 3) In another study, striatal extracellular DA levels decreased, and morphine-induced DA release was blocked after injection of ibogaine (40mg/kg) (Maisonneuve, Keller and Glick, 1991). Also, the fact that attenuation of addictive behaviour was observed long after the acute effects of ibogaine, suggests that the decrease is due to antagonistic neurobiological mechanisms in morphine's addictive properties. It is thus hypothesized, that normalization of mesolimbic dopamine firing could be responsible for the antiaddictive properties. Another underlying mechanism that is suggested, assigns the antiaddictive properties to kappa opioid and NMDA agonism for opioid and stimulant SA (Glick and Maisonneue, 1998b), although this hypothesis needs further exploration.

Noribogaine, ibogaine, and 18-methoxycoronaridine were shown to attenuate the withdrawal symptoms of morphine in mice. 1) Treatment with18-MC (40mg/kg) was shown to be effective in attenuating 5 of the 7 signs of morphine withdrawal, which were burying, diarrhoea, weight loss, teeth chattering and wet-dog shaking (Panchal *et al.*, 2005). 2)

Ibogaine was tested in naloxone-induced withdrawal in morphine-dependent mice: this effect was attributed to NMDA receptors as jumping (the major symptom of this type of withdrawal) was significantly inhibited in ibogaine treated mice (Leal *et al.*, 2003). Although a later paper argues that these effects are most probably caused by the metabolite noribogaine (Mash *et al.*, 2016).

Alcohol and nicotine

In the case of alcohol use attenuation, also both ibogaine and 18-MC have proved to be effective. 1) In a study on three alcohol-preferring strains of mice alcohol consumption was measured acutely and over a period of five days. Doses of 60mg/kg (which were shown to be most effective) were administered to all three strains of mice. The amount of alcohol consumed was reduced by 60% acutely and it remained to be consistently lower compared to control groups (Fig.10b) (Rezvani, Overstreet and Leef, 1995). 2) The same effects were demonstrated using 18-MC (Rezvani, 1997). In a recent study, treatments with 18-MC were not just effective in alcohol, but also nicotine cessation (Rezvani *et al.*, 2016). 3) Another paper on nicotine cessation confirmed this finding by treating Sprague-Dawley rats with noribogaine, which showed an acute dose-dependent 64% reduction in IVSA (Chang *et al.*, 2015).

Stimulants

Results with cocaine and amphetamine self-administration are very similar to those using opioids. 1) After one treatment with 40mg/kg ibogaine, a reduction in cocaine consumption was detected for up to 48 hours, although the best results were obtained after three consecutive treatments (one per week) after which a long-lasting gradual decrease was observed (fig. 12) (Cappendijk and Dzoljic, 1993; Sershen, Hashim and Lajtha, 1994). 2) While with methamphetamine, only an acute effect lasting 5 days was observed before the amount consumed returned to baseline (Glick, Maisonneuve and Dickinson, 2000).

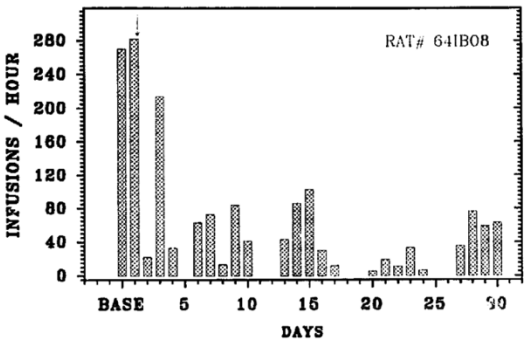


Figure 10a Long-lasting effects of ibogaine on morphine self-administration, over a 30day testing period. Rats were sustaining lower SA either after a single infusion (the arrow) or after weekly infusions of ibogaine. Source: Glick *et al.*, 1991)

Figure 10b Acute effects of different doses of ibogaine on acute self-administration of alcohol in three alcohol-preferring strains of mice. Source: (Rezvani, Overstreet and Leef, 1995)

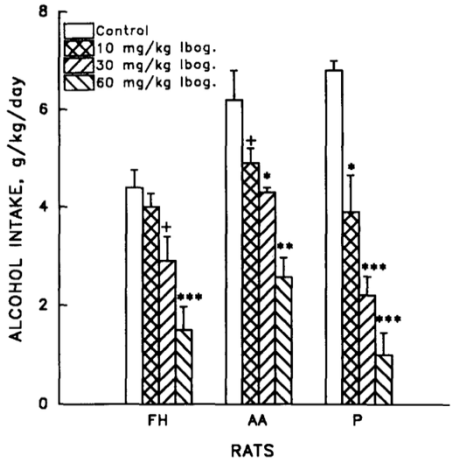
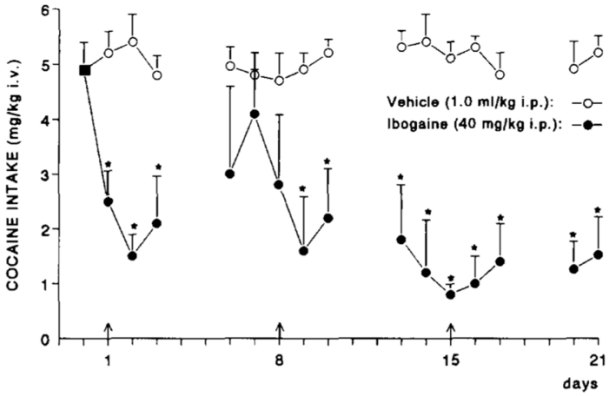


Figure 10c Effects of weekly ibogaine injection on the self-administration of cocaine over several weeks. Arrows indicate the days of ibogaine treatment. Source: (Cappendijk and Dzoljic, 1993)



In summary, as preclinical research on the effectiveness of LSD is limited to only two papers, there is not enough data to show a clear consensus, in contrast to clinical research. In the case of tryptamines, from the few available studies, DMT appears to be effective in the form of ayahuasca. Further psilocin research is highly required for its considerably introspective subjective effects and high selectivity for 5-HT_{2A}. Ibogaine is by far the most researched psychedelic when it comes to this topic, as it appears to be an addiction interrupter, while attenuating withdrawal symptoms and feelings of craving in addicts.

To further measure the effect of psychedelics on the cessation of drug consumption, a self-administration experiment that would consider the social environment of the rat, as well as their preference for alternative rewards, would be needed. High doses of natural psychedelics ayahuasca or *iboga* would be administered in just one or in a series of weekly sessions. And the animals would also have the choice to interact with their social environment and would be presented with a competing reward. In humans, this would correspond to either a psychedelic-assisted psychotherapy approach (where LSD and psilocin are generally administered), or a more naturalistic shamanic ceremony (usually using ayahuasca or iboga).

6 Conclusion

The described model of addiction involves the disruption of neural pathways responsible for motivation and desire, while acknowledging that these are the same pathways being reinforced in learning and habit development. As Marc Lewis writes in *The Biology of desire*:

"Addiction results from the motivated repetition of the same thoughts and behaviours until they become habitual. Thus, addiction develops- it's learned- but it's learned more deeply and often more quickly than other habits, due to a narrowing tunnel of attention and attraction" (Marc Lewis- *The Biology of Desire*)

Addictive substances can thus be viewed as merely a catalyst, an extremely rewarding habit-forming experience. Brain changes that occur subsequently to substance consumption are attributed to the phasic firing of dopamine, as higher levels of dopamine in the mesocortical and mesolimbic pathways create feedback loops that reinforce themselves, neuronal connections then strengthen, and a disruption in normal feelings of desire and motivation is created. The brain is essentially made to become addicted and drugs of abuse just highjack this system that took thousands of years to evolve.

Models of addiction can be studied either clinically on human subjects or on animals, mainly rodents, for whom a variety of methods have been developed including one of the most promising, the self-administration method. Preclinical research was only recently made possible after a surge of interest in the therapeutic application of psychedelics, whereas some controlled clinical studies began to emerge in the late 1950s and 60s, but forcefully being interrupted due to restrictions on the use of psychedelics. Therefore, there is still insufficient data available and further preclinical and controlled double-blind studies will be needed to help us understand the neurobiological and physiological mechanisms regardless of the psychological factors, which can't be disregarded in psychedelic human research. Experimental methods using operant conditioning like intravenous self-administration tests have high face and construct validity, which makes them the most relevant in this context. Nevertheless, considering only the pharmacological aspect of addiction would be an oversimplification, as described by the recent Bio-Psycho-Social-Spiritual model of addiction. Models that consider social environment, like individual housing compared to group housing, and personality traits of experimental animals, reflecting mainly novelty-seeking behaviour, could be therefore promising for further understanding the addiction process.

Now, the reason psychedelics could have potential in the future of mental health and addiction research is that they have the power to disrupt previously established patterns of behaviour. Classic psychedelics interact with 5-HT and other receptors, affecting membrane

potentials and the subsequent release of neurotransmitters, which can result in the formation of new connections, neuroplasticity, or disintegration of already fixed feedback loops (in particular, the thalamus-PCC connection and the default mode network responsible for our sense of “self”). These compounds are therefore interesting because of their characteristic subjective effects, that produce a change in perspective, making individuals able to confront reality without the need for a chemical buffer.

7 Literature

Abramson, H.A. *et al.* (1955) "Lysergic Acid Diethylamide (LSD-25): I. Physiological and Perceptual Responses," *The Journal of Psychology*, 39(1), pp. 3–60. doi:10.1080/00223980.1955.9916156.

Acquas, E., Meloni, M. and di Chiara, G. (1993) "Blockade of δ-opioid receptors in the nucleus accumbens prevents ethanol-induced stimulation of dopamine release," *European Journal of Pharmacology*, 230(2), pp. 239–241. doi:10.1016/0014-2999(93)90809-V.

Aghajanian, G.K., Foote, W.E. and Sheard, M.H. (1968) "Lysergic Acid Diethylamide: Sensitive Neuronal Units in the Midbrain Raphe," *Science*, 161(3842), pp. 706–708. doi:10.1126/science.161.3842.706.

Aghajanian, G.K. and Hailgler, H.J. (1975) "Hallucinogenic indoleamines: Preferential action upon presynaptic serotonin receptors.," *Psychopharmacology communications*, 1(6), pp. 619–29.

Alexander, B.K., Coambs, R.B. and Hadaway, P.F. (1978) "The effect of housing and gender on morphine self-administration in rats," *Psychopharmacology*, 58(2), pp. 175–179. doi:10.1007/BF00426903.

Alper, K. *et al.* (2018) "LSD Administered as a Single Dose Reduces Alcohol Consumption in C57BL/6J Mice," *Frontiers in Pharmacology*, 9. doi:10.3389/fphar.2018.00994.

Alper, K.R., Lotsof, H.S. and Kaplan, C.D. (2008) "The ibogaine medical subculture," *Journal of Ethnopharmacology*, 115(1), pp. 9–24. doi:10.1016/j.jep.2007.08.034.

Basavarajappa, B.S., Cooper, T.B. and Hungund, B.L. (1998) "Chronic ethanol administration down-regulates cannabinoid receptors in mouse brain synaptic plasma membrane," *Brain Research*, 793(1–2), pp. 212–218. doi:10.1016/S0006-8993(98)00175-9.

Baumann, M.H. *et al.* (2001) "In vivo neurobiological effects of ibogaine and its O-desmethyl metabolite, 12-hydroxyibogamine (noribogaine), in rats.," *The Journal of pharmacology and experimental therapeutics*, 297(2), pp. 531–9.

Belgers, M. *et al.* (2016) "Ibogaine and addiction in the animal model, a systematic review and meta-analysis," *Translational Psychiatry*, 6(5), pp. e826–e826. doi:10.1038/tp.2016.71.

Berrendero, F., Kieffer, B.L. and Maldonado, R. (2002) "Attenuation of Nicotine-Induced Antinociception, Rewarding Effects, and Dependence in μ-Opioid Receptor Knock-Out Mice," *The Journal of Neuroscience*, 22(24), pp. 10935–10940. doi:10.1523/JNEUROSCI.22-24-10935.2002.

Bhargava, H.N. and Cao, Y.-J. (1997) "Effects of noribogaine on the development of tolerance to antinociceptive action of morphine in mice," *Brain Research*, 771(2), pp. 343–346. doi:10.1016/S0006-8993(97)00914-1.

Bombardi, C. (2014) “Neuronal localization of the 5-HT2 receptor family in the amygdaloid complex,” *Frontiers in Pharmacology*, 5. doi:10.3389/fphar.2014.00068.

Briand, L.A., Gross, J.P. and Robinson, T.E. (2008) “Impaired object recognition following prolonged withdrawal from extended-access cocaine self-administration,” *Neuroscience*, 155(1), pp. 1–6. doi:10.1016/j.neuroscience.2008.06.004.

Bunzow, J.R. *et al.* (2001) “Amphetamine, 3,4-Methylenedioxymethamphetamine, Lysergic Acid Diethylamide, and Metabolites of the Catecholamine Neurotransmitters Are Agonists of a Rat Trace Amine Receptor,” *Molecular Pharmacology*, 60(6), pp. 1181–1188. doi:10.1124/mol.60.6.1181.

Canal, C.E. and Murnane, K.S. (2017) “The serotonin 5-HT $_{2C}$ receptor and the non-addictive nature of classic hallucinogens,” *Journal of Psychopharmacology*, 31(1), pp. 127–143. doi:10.1177/0269881116677104.

Cappendijk, S.L.T. and Dzoljic, M.R. (1993) “Inhibitory effects of ibogaine on cocaine self-administration in rats,” *European Journal of Pharmacology*, 241(2–3), pp. 261–265. doi:10.1016/0014-2999(93)90212-Z.

Carbonaro, T.M. *et al.* (2015) “The role of 5-HT2A, 5-HT2C and mGlu2 receptors in the behavioral effects of tryptamine hallucinogens N,N-dimethyltryptamine and N,N-diisopropyltryptamine in rats and mice,” *Psychopharmacology*, 232(1), pp. 275–284. doi:10.1007/s00213-014-3658-3.

Carey, A.N. *et al.* (2007) “Reinstatement of cocaine place-conditioning prevented by the peptide kappa-opioid receptor antagonist arodyn,” *European Journal of Pharmacology*, 569(1–2), pp. 84–89. doi:10.1016/j.ejphar.2007.05.007.

Carhart-Harris, R.L. *et al.* (2012) “Neural correlates of the psychedelic state as determined by fMRI studies with psilocybin,” *Proceedings of the National Academy of Sciences*, 109(6), pp. 2138–2143. doi:10.1073/pnas.1119598109.

Cata-Preta, E.G. *et al.* (2018) “Ayahuasca and Its DMT- and β-carbolines – Containing Ingredients Block the Expression of Ethanol-Induced Conditioned Place Preference in Mice: Role of the Treatment Environment,” *Frontiers in Pharmacology*, 9. doi:10.3389/fphar.2018.00561.

Chang, Q. *et al.* (2015) “Noribogaine reduces nicotine self-administration in rats,” *Journal of Psychopharmacology*, 29(6), pp. 704–711. doi:10.1177/0269881115584461.

Codd, E.E. (1995) “High affinity ibogaine binding to a mu opioid agonist site,” *Life Sciences*, 57(20), pp. PL315–PL320. doi:10.1016/0024-3205(95)02171-E.

Coleman, J.A. *et al.* (2019) “Serotonin transporter–ibogaine complexes illuminate mechanisms of inhibition and transport,” *Nature*, 569(7754), pp. 141–145. doi:10.1038/s41586-019-1135-1.

Colpaert, F.C., Niemegeers, C.J. and Janssen, P.A. (1982) “A drug discrimination analysis of lysergic acid diethylamide (LSD): in vivo agonist and antagonist effects of purported

5-hydroxytryptamine antagonists and of pirenperone, a LSD-antagonist.," *The Journal of pharmacology and experimental therapeutics*, 221(1), pp. 206–14.

Contet, C., Kieffer, B.L. and Befort, K. (2004) "Mu opioid receptor: a gateway to drug addiction," *Current Opinion in Neurobiology*, 14(3), pp. 370–378. doi:10.1016/j.conb.2004.05.005.

Cozzi, N. v. *et al.* (2009) "Dimethyltryptamine and other hallucinogenic tryptamines exhibit substrate behavior at the serotonin uptake transporter and the vesicle monoamine transporter," *Journal of Neural Transmission*, 116(12), pp. 1591–1599. doi:10.1007/s00702-009-0308-8.

Cunningham, C.L., Gremel, C.M. and Groblewski, P.A. (2006) "Drug-induced conditioned place preference and aversion in mice," *Nature Protocols*, 1(4), pp. 1662–1670. doi:10.1038/nprot.2006.279.

David, V. *et al.* (2001) "Intravenous self-administration of heroin/cocaine combinations (speedball) using nose-poke or lever-press operant responding in mice," *Behavioral Pharmacology*, 12(1), pp. 25–34. doi:10.1097/00008877-200102000-00003.

Dean, J.G. *et al.* (2019) "Biosynthesis and Extracellular Concentrations of N,N-dimethyltryptamine (DMT) in Mammalian Brain," *Scientific Reports*, 9(1), p. 9333. doi:10.1038/s41598-019-45812-w.

Dolder, P.C. *et al.* (2016) "Pharmacokinetics and Concentration-Effect Relationship of Oral LSD in Humans," *International Journal of Neuropsychopharmacology*, 19(1), p. pyv072. doi:10.1093/ijnp/pyv072.

Dolder, P.C. *et al.* (2017) "Pharmacokinetics and Pharmacodynamics of Lysergic Acid Diethylamide in Healthy Subjects," *Clinical Pharmacokinetics*, 56(10), pp. 1219–1230. doi:10.1007/s40262-017-0513-9.

Engel, G.L. (1977) "The Need for a New Medical Model: A Challenge for Biomedicine," *Science*, 196(4286), pp. 129–136. doi:10.1126/science.847460.

Funk, C.K. *et al.* (2006) "Corticotropin-Releasing Factor within the Central Nucleus of the Amygdala Mediates Enhanced Ethanol Self-Administration in Withdrawn, Ethanol-Dependent Rats," *Journal of Neuroscience*, 26(44), pp. 11324–11332. doi:10.1523/JNEUROSCI.3096-06.2006.

Garcia-Romeu, A. *et al.* (2019) "Cessation and reduction in alcohol consumption and misuse after psychedelic use," *Journal of Psychopharmacology*, 33(9), pp. 1088–1101. doi:10.1177/0269881119845793.

Gerak, L.R., Collins, G.T. and France, C.P. (2016) "Effects of Lorcaserin on Cocaine and Methamphetamine Self-Administration and Reinstatement of Responding Previously Maintained by Cocaine in Rhesus Monkeys," *Journal of Pharmacology and Experimental Therapeutics*, 359(3), pp. 383–391. doi:10.1124/jpet.116.236307.

Glennon, R.A., Titeler, M. and McKenney, J.D. (1984) "Evidence for 5-HT2 involvement in the mechanism of action of hallucinogenic agents," *Life Sciences*, 35(25), pp. 2505–2511. doi:10.1016/0024-3205(84)90436-3.

Glick, S.D. *et al.* (1991) "Effects and aftereffects of ibogaine on morphine self-administration in rats," *European Journal of Pharmacology*, 195(3), pp. 341–345. doi:10.1016/0014-2999(91)90474-5.

Glick, S.D. *et al.* (1994) "Effects of iboga alkaloids on morphine and cocaine self-administration in rats: relationship to tremorigenic effects and to effects on dopamine release in nucleus accumbens and striatum," *Brain Research*, 657(1–2), pp. 14–22. doi:10.1016/0006-8993(94)90948-2.

Glick, S.D. and Maisonneuve, I.M. (1998a) "Mechanisms of Antiaddictive Actions of Ibogaine [a]," *Annals of the New York Academy of Sciences*, 844(1), pp. 214–226. doi:10.1111/j.1749-6632.1998.tb08237.x.

Glick, S.D. and Maisonneuve, I.M. (1998b) "Mechanisms of Antiaddictive Actions of Ibogaine [a]," *Annals of the New York Academy of Sciences*, 844(1), pp. 214–226. doi:10.1111/j.1749-6632.1998.tb08237.x.

Glick, S.D., Maisonneuve, I.M. and Dickinson, H.A. (2000) "18-MC reduces methamphetamine and nicotine self-administration in rats," *NeuroReport*, 11(9), pp. 2013–2015. doi:10.1097/00001756-200006260-00041.

Glick, S.D., Maisonneuve, I.M. and Szumlinski, K.K. (2000) "18-Methoxycoronaridine (18-MC) and Ibogaine: Comparison of Antiaddictive Efficacy, Toxicity, and Mechanisms of Action," *Annals of the New York Academy of Sciences*, 914(1), pp. 369–386. doi:10.1111/j.1749-6632.2000.tb05211.x.

Godinho AF (2017) "Ayahuasca Modifies Amphetamine Self Ingestion and Modifies Anxiety and Locomotor Activity in Adolescent Rats," *Electron J Biol*, 13, p. 2.

Gupta, S. and Kulhara, P. (2007) "Cellular and molecular mechanisms of drug dependence: An overview and update.," *Indian journal of psychiatry*, 49(2), pp. 85–90. doi:10.4103/0019-5545.33253.

Haass-Koffler, C.L. and Bartlett, S.E. (2012) "Stress and addiction: contribution of the corticotropin releasing factor (CRF) system in neuroplasticity," *Frontiers in Molecular Neuroscience*, 5. doi:10.3389/fnmol.2012.00091.

Halberstadt, A.L. (2016) "Behavioral and pharmacokinetic interactions between monoamine oxidase inhibitors and the hallucinogen 5-methoxy-N,N-dimethyltryptamine," *Pharmacology Biochemistry and Behavior*, 143, pp. 1–10. doi:10.1016/j.pbb.2016.01.005.

Hasler, F. *et al.* (2002) "Renal excretion profiles of psilocin following oral administration of psilocybin: a controlled study in man," *Journal of Pharmaceutical and Biomedical Analysis*, 30(2), pp. 331–339. doi:10.1016/S0731-7085(02)00278-9.

Helmy, A., H. Carpenter, K. and Hutchinson, P. (2007) “Microdialysis in the Human Brain and its Potential Role in the Development and Clinical Assessment of Drugs,” *Current Medicinal Chemistry*, 14(14), pp. 1525–1537. doi:10.2174/092986707780831113.

Higgins, G.A. and Sellers, E.M. (2021) “5-HT2A and 5-HT2C receptors as potential targets for the treatment of nicotine use and dependence,” in, pp. 229–263. doi:10.1016/bs.pbr.2021.01.007.

Hnasko, T.S., Sotak, B.N. and Palmiter, R.D. (2007) “Cocaine-Conditioned Place Preference by Dopamine-Deficient Mice Is Mediated by Serotonin,” *Journal of Neuroscience*, 27(46), pp. 12484–12488. doi:10.1523/JNEUROSCI.3133-07.2007.

Horita, A. and Weber, L.J. (1961) “The enzymic dephosphorylation and oxidation of psilocybin and pscilocin by mammalian tissue homogenates,” *Biochemical Pharmacology*, 7(1), pp. 47–54. doi:10.1016/0006-2952(61)90124-1.

Houdi, A.A., Dasgupta, R. and Kindy, M.S. (1998) “Effect of nicotine use and withdrawal on brain preproenkephalin A mRNA,” *Brain Research*, 799(2), pp. 257–263. doi:10.1016/S0006-8993(98)00454-5.

Hough, L.B., Pearl, S.M. and Glick, S.D. (1996) “Tissue distribution of ibogaine after intraperitoneal and subcutaneous administration,” *Life Sciences*, 58(7), pp. PL119–PL122. doi:10.1016/0024-3205(95)02322-4.

Koob, G.F. and Bloom, F.E. (1988) “Cellular and Molecular Mechanisms of Drug Dependence,” *Science*, 242(4879), pp. 715–723. doi:10.1126/science.2903550.

Koob, G.F. and Volkow, N.D. (2010) “Neurocircuitry of Addiction,” *Neuropsychopharmacology*, 35(1), pp. 217–238. doi:10.1038/npp.2009.110.

Kornetsky, C. (1979) “Intracranial Self-stimulation Thresholds,” *Archives of General Psychiatry*, 36(3), p. 289. doi:10.1001/archpsyc.1979.01780030055004.

Krebs, T.S. and Johansen, P.-Ø. (2012) “Lysergic acid diethylamide (LSD) for alcoholism: meta-analysis of randomized controlled trials,” *Journal of Psychopharmacology*, 26(7), pp. 994–1002. doi:10.1177/0269881112439253.

Kuhn, B.N., Kalivas, P.W. and Bobadilla, A.-C. (2019) “Understanding Addiction Using Animal Models,” *Frontiers in Behavioral Neuroscience*, 13. doi:10.3389/fnbeh.2019.00262.

Leal, M.B. *et al.* (2003) “Ibogaine attenuation of morphine withdrawal in mice: role of glutamate N-methyl-d-aspartate receptors,” *Progress in Neuro-Psychopharmacology and Biological Psychiatry*, 27(5), pp. 781–785. doi:10.1016/S0278-5846(03)00109-X.

Leshner, A.I. (1997) “Addiction Is a Brain Disease, and It Matters,” *Science*, 278(5335), pp. 45–47. doi:10.1126/science.278.5335.45.

Lord, J.A.H. *et al.* (1977) “Endogenous opioid peptides: multiple agonists and receptors,” *Nature*, 267(5611), pp. 495–499. doi:10.1038/267495a0.

Luethi, D. *et al.* (2019) "Cytochrome P450 enzymes contribute to the metabolism of LSD to nor-LSD and 2-oxo-3-hydroxy-LSD: Implications for clinical LSD use," *Biochemical Pharmacology*, 164, pp. 129–138. doi:10.1016/j.bcp.2019.04.013.

Maillet, E.L. *et al.* (2015) "Noribogaine is a G-protein biased κ-opioid receptor agonist," *Neuropharmacology*, 99, pp. 675–688. doi:10.1016/j.neuropharm.2015.08.032.

Maisonneuve, I.M. *et al.* (1992) "Acute and prolonged effects of ibogaine on brain dopamine metabolism and morphine-induced locomotor activity in rats," *Brain Research*, 575(1), pp. 69–73. doi:10.1016/0006-8993(92)90424-8.

Maisonneuve, I.M. *et al.* (1997) "Ibogaine and the dopaminergic response to nicotine," *Psychopharmacology*, 129(3), pp. 249–256. doi:10.1007/s002130050187.

Maisonneuve, I.M., Keller, R.W. and Glick, S.D. (1991) "Interactions between ibogaine, a potential anti-addictive agent, and morphine: an in vivo microdialysis study," *European Journal of Pharmacology*, 199(1), pp. 35–42. doi:10.1016/0014-2999(91)90634-3.

Marek, G.J. and Aghajanian, G.K. (1996) "LSD and the phenethylamine hallucinogen DOI are potent partial agonists at 5-HT2A receptors on interneurons in rat piriform cortex.," *The Journal of pharmacology and experimental therapeutics*, 278(3), pp. 1373–82.

Mash, D.C. *et al.* (2000) "Ibogaine: Complex Pharmacokinetics, Concerns for Safety, and Preliminary Efficacy Measures," *Annals of the New York Academy of Sciences*, 914(1), pp. 394–401. doi:10.1111/j.1749-6632.2000.tb05213.x.

Mash, D.C. *et al.* (2016) "Oral noribogaine shows high brain uptake and anti-withdrawal effects not associated with place preference in rodents," *Journal of Psychopharmacology*, 30(7), pp. 688–697. doi:10.1177/0269881116641331.

Maurel, S., de Vry, J. and Schreiber, R. (1999) "5-HT receptor ligands differentially affect operant oral self-administration of ethanol in the rat," *European Journal of Pharmacology*, 370(3), pp. 217–223. doi:10.1016/S0014-2999(99)00125-9.

Meert, T.F., de Haes, P. and Janssen, P.A.J. (1989) "Risperidone (R 64 766), a potent and complete LSD antagonist in drug discrimination by rats," *Psychopharmacology*, 97(2), pp. 206–212. doi:10.1007/BF00442251.

Meinhardt, M.W. *et al.* (2020) "Psilocybin and LSD have no long-lasting effects in an animal model of alcohol relapse," *Neuropsychopharmacology*, 45(8), pp. 1316–1322. doi:10.1038/s41386-020-0694-z.

Melo, J.A. *et al.* (1996) "Identification of sex–specific quantitative trait loci controlling alcohol preference in C57BL/6 mice," *Nature Genetics*, 13(2), pp. 147–153. doi:10.1038/ng0696-147.

Millan, M.J., Dekeyne, A. and Gobert, A. (1998) "Serotonin (5-HT)2C receptors tonically inhibit dopamine (DA) and noradrenaline (NA), but not 5-HT, release in the frontal

cortex in vivo," *Neuropharmacology*, 37(7), pp. 953–955. doi:10.1016/S0028-3908(98)00078-1.

Miller, M.J. *et al.* (2019) "Chemical evidence for the use of multiple psychotropic plants in a 1,000-year-old ritual bundle from South America," *Proceedings of the National Academy of Sciences*, 116(23), pp. 11207–11212. doi:10.1073/pnas.1902174116.

Müller, F. *et al.* (2018) "Altered network hub connectivity after acute LSD administration," *NeuroImage: Clinical*, 18, pp. 694–701. doi:10.1016/j.nicl.2018.03.005.

Mysels, D. (2009) "The Kappa-Opiate Receptor Impacts the Pathophysiology and Behavior of Substance Use," *American Journal on Addictions*, 18(4), pp. 272–276. doi:10.1080/10550490902925862.

Nestler, E.J., Barrot, M. and Self, D.W. (2001) "ΔFosB: A sustained molecular switch for addiction," *Proceedings of the National Academy of Sciences*, 98(20), pp. 11042–11046. doi:10.1073/pnas.191352698.

Nyongo Ndoua, P.D. and Vaghar, K. (2018) "Bwiti, iboga, trance and healing in Gabon," *Mental Health, Religion & Culture*, 21(8), pp. 755–762. doi:10.1080/13674676.2018.1504012.

Olds, J. and Milner, P. (1954) "Positive reinforcement produced by electrical stimulation of septal area and other regions of rat brain.," *Journal of Comparative and Physiological Psychology*, 47(6), pp. 419–427. doi:10.1037/h0058775.

Oliveira-Lima, A.J. *et al.* (2015) "Effects of ayahuasca on the development of ethanol-induced behavioral sensitization and on a post-sensitization treatment in mice," *Physiology & Behavior*, 142, pp. 28–36. doi:10.1016/j.physbeh.2015.01.032.

Panchal, V. *et al.* (2005) "Attenuation of morphine withdrawal signs by intracerebral administration of 18-methoxycoronaridine," *European Journal of Pharmacology*, 525(1–3), pp. 98–104. doi:10.1016/j.ejphar.2005.09.060.

Panlilio, L. v. and Goldberg, S.R. (2007) "Self-administration of drugs in animals and humans as a model and an investigative tool," *Addiction*, 102(12), pp. 1863–1870. doi:10.1111/j.1360-0443.2007.02011.x.

Popik, P. and Skolnick, P. (1999) "Pharmacology of Ibogaine and Ibogaine-Related Alkaloids," in, pp. 197–231. doi:10.1016/S0099-9598(08)60027-9.

Preller, K.H. *et al.* (2019) "Effective connectivity changes in LSD-induced altered states of consciousness in humans," *Proceedings of the National Academy of Sciences*, 116(7), pp. 2743–2748. doi:10.1073/pnas.1815129116.

Reis, H.S. *et al.* (2020) "Ayahuasca blocks the reinstatement of methylphenidate-induced conditioned place preference in mice: behavioral and brain Fos expression evaluations," *Psychopharmacology*, 237(11), pp. 3269–3281. doi:10.1007/s00213-020-05609-6.

Reissig, C.J. *et al.* (2005) "The 5-HT1A receptor and the stimulus effects of LSD in the rat," *Psychopharmacology*, 182(2), pp. 197–204. doi:10.1007/s00213-005-0068-6.

Rezvani, A. (1997) “Attenuation of Alcohol Consumption by a Novel Nontoxic Ibogaine Analogue (18-Methoxycoronaridine) in Alcohol-Preferring Rats,” *Pharmacology Biochemistry and Behavior*, 58(2), pp. 615–619. doi:10.1016/S0091-3057(97)10003-X.

Rezvani, A.H. *et al.* (2016) “Acute oral 18-methoxycoronaridine (18-MC) decreases both alcohol intake and IV nicotine self-administration in rats,” *Pharmacology Biochemistry and Behavior*, 150–151, pp. 153–157. doi:10.1016/j.pbb.2016.10.010.

Rezvani, A.H., Overstreet, D.H. and Leef, Y.-W. (1995) “Attenuation of alcohol intake by Ibogaine in three strains of alcohol-preferring rats,” *Pharmacology Biochemistry and Behavior*, 52(3), pp. 615–620. doi:10.1016/0091-3057(95)00152-M.

Riba, J. *et al.* (2003) “Human Pharmacology of Ayahuasca: Subjective and Cardiovascular Effects, Monoamine Metabolite Excretion, and Pharmacokinetics,” *Journal of Pharmacology and Experimental Therapeutics*, 306(1), pp. 73–83. doi:10.1124/jpet.103.049882.

Robinson, T. and Becker, J. (1986) “Enduring changes in brain and behavior produced by chronic amphetamine administration: A review and evaluation of animal models of amphetamine psychosis,” *Brain Research*, 396(2), pp. 157–198. doi:10.1016/S0006-8993(86)80193-7.

Rodríguez, P. *et al.* (2020) “A Single Administration of the Atypical Psychedelic Ibogaine or Its Metabolite Noribogaine Induces an Antidepressant-Like Effect in Rats,” *ACS Chemical Neuroscience*, 11(11), pp. 1661–1672. doi:10.1021/acschemneuro.0c00152.

Rommelspacher, H. *et al.* (1992) “Adaptive changes of dopamine-D2 receptors in rat brain following ethanol withdrawal: A quantitative autoradiographic investigation,” *Alcohol*, 9(5), pp. 355–362. doi:10.1016/0741-8329(92)90032-6.

Rüedi-Bettschen, D., Spealman, R.D. and Platt, D.M. (2015) “Attenuation of cocaine-induced reinstatement of drug seeking in squirrel monkeys by direct and indirect activation of 5-HT2C receptors,” *Psychopharmacology*, 232(16), pp. 2959–2968. doi:10.1007/s00213-015-3932-z.

Rusheen, A.E. *et al.* (2020) “Evaluation of electrochemical methods for tonic dopamine detection in vivo,” *TrAC Trends in Analytical Chemistry*, 132, p. 116049. doi:10.1016/j.trac.2020.116049.

Ryskamp, D.A. *et al.* (2019) “Neuronal Sigma-1 Receptors: Signaling Functions and Protective Roles in Neurodegenerative Diseases,” *Frontiers in Neuroscience*, 13. doi:10.3389/fnins.2019.00862.

Sershen, H., Hashim, A. and Lajtha, A. (1994) “Ibogaine reduces preference for cocaine consumption in C57BL/6By mice,” *Pharmacology Biochemistry and Behavior*, 47(1), pp. 13–19. doi:10.1016/0091-3057(94)90105-8.

Sharma, S.K., Klee, W.A. and Nirenberg, M. (1975) “Dual regulation of adenylate cyclase accounts for narcotic dependence and tolerance.,” *Proceedings of the National Academy of Sciences*, 72(8), pp. 3092–3096. doi:10.1073/pnas.72.8.3092.

Skewes, M.C. and Gonzalez, V.M. (2013) "The Biopsychosocial Model of Addiction," in *Principles of Addiction*. Elsevier, pp. 61–70. doi:10.1016/B978-0-12-398336-7.00006-1.

Smith, R. (1998) "Agonist Properties of N,N-Dimethyltryptamine at Serotonin 5-HT2A and 5-HT2C Receptors," *Pharmacology Biochemistry and Behavior*, 61(3), pp. 323–330. doi:10.1016/S0091-3057(98)00110-5.

Strassman, R.J. (1995) "Human psychopharmacology of N,N-dimethyltryptamine," *Behavioural Brain Research*, 73(1–2), pp. 121–124. doi:10.1016/0166-4328(96)00081-2.

Su, T.-P., Hayashi, T. and Vaupel, D.B. (2009) "When the Endogenous Hallucinogenic Trace Amine *N,N* -Dimethyltryptamine Meets the Sigma-1 Receptor," *Science Signaling*, 2(61). doi:10.1126/scisignal.261pe12.

Szára, St. (1956) "Dimethyltryptamin: Its metabolism in man; the relation of its psychotic effect to the serotonin metabolism," *Experientia*, 12(11), pp. 441–442. doi:10.1007/BF02157378.

Tanda, G. *et al.* (2004) "The opioid antagonist naltrexone reduces the reinforcing effects of ? 9 -tetrahydrocannabinol (THC) in squirrel monkeys," *Psychopharmacology*, 173(1–2), pp. 186–194. doi:10.1007/s00213-003-1693-6.

Uhl, G.R., Koob, G.F. and Cable, J. (2019) "The neurobiology of addiction," *Annals of the New York Academy of Sciences*, 1451(1), pp. 5–28. doi:10.1111/nyas.13989.

Ungerstedt, U. and Pycock, C. (1974) "Functional correlates of dopamine neurotransmission.," *Bulletin der Schweizerischen Akademie der Medizinischen Wissenschaften*, 30(1–3), pp. 44–55.

Valjent, E. *et al.* (2010) "Mechanisms of Locomotor Sensitization to Drugs of Abuse in a Two-Injection Protocol," *Neuropsychopharmacology*, 35(2), pp. 401–415. doi:10.1038/npp.2009.143.

Venniro, M. and Shaham, Y. (2020) "An operant social self-administration and choice model in rats," *Nature Protocols*, 15(4), pp. 1542–1559. doi:10.1038/s41596-020-0296-6.

Volkow, N.D. *et al.* (1993) "Decreased dopamine D2 receptor availability is associated with reduced frontal metabolism in cocaine abusers," *Synapse*, 14(2), pp. 169–177. doi:10.1002/syn.890140210.

Wacker, D. *et al.* (2017) "Crystal Structure of an LSD-Bound Human Serotonin Receptor," *Cell*, 168(3), pp. 377-389.e12. doi:10.1016/j.cell.2016.12.033.

Watts, V.J. *et al.* (1995) "LSD and structural analogs: pharmacological evaluation at D1 dopamine receptors," *Psychopharmacology*, 118(4), pp. 401–409. doi:10.1007/BF02245940.

Wei, D. *et al.* (1998) "Acute iboga alkaloid effects on extracellular serotonin (5-HT) levels in nucleus accumbens and striatum in rats," *Brain Research*, 800(2), pp. 260–268. doi:10.1016/S0006-8993(98)00527-7.

Wells, G.B., Lopez, M.C. and Tanaka, J.C. (1999) "The effects of ibogaine on dopamine and serotonin transport in rat brain synaptosomes," *Brain Research Bulletin*, 48(6), pp. 641–647. doi:10.1016/S0361-9230(99)00053-2.

Whitfield-Gabrieli, S. and Ford, J.M. (2012) "Default Mode Network Activity and Connectivity in Psychopathology," *Annual Review of Clinical Psychology*, 8(1), pp. 49–76. doi:10.1146/annurev-clinpsy-032511-143049.

7.1 Books

A.Hoffmann, *LSD, my problem child*, Profess 1997, ISBN 0199639418

Marc Lewis, *The Biology of Desire*: why addiction is not a disease, 13th edition, Scribe Publications 2016, page 7, ISBN 9781925228779

R.Strassmann, *DMT, the Spirit Molecule: A Doctor's Revolutionary Research in the Biology of Near-Death and Mystical Experiences*, Simon and Schuster 2000, ISBN 9798200096411,

7.2 Websites

drsimonsaysscience.org
psychedelicreview.com
psychonautwiki.com
globaldrugsurvey.com