THE RELATIONSHIP BETWEEN SEROTONIN, DECISION-MAKING AND MOOD

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SUBMITTED FOR THE AWARD OF Ph.D

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OCTOBER 2013

DECLARATION

I, Paul Faulkner, confirm that the work presented in this thesis is my own. Where information has been derived from other sources, I confirm that this has been indicated in the thesis.

ACKNOWLEDGEMENTS

I would like to thank the following for their support and expertise throughout the completion of this thesis:

Dr Jonathan P. Roiser

Professor Peter Dayan

Professor Stephen Pilling

Dr Oliver Howes

Professor H. Valerie Curran

Professor David Nutt

Professor Ray Dolan

Dr Quentin Huys

Dr Sudhakar Selvaraj

Dr Elias Mouchlianitis

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ABSTRACT

The serotonin (5-HT) system has been implicated in both depression and reward and punishment processing. This thesis presents data from four studies designed to better understand the role of serotonin in decision-making and mood. Following the general introduction and description of the main experimental methods, the first experimental chapter presents a study that examined the relationship between naturally-varying 5-HT_{1A} receptor availability, measured using positron emission tomography, and decision-making in healthy volunteers. This study identified correlations between 5-HT_{1A} receptor availability in the hippocampal complex and both impulsivity and sensitivity to the probability of an outcome during decision-making. The second experimental chapter examined decision-making in healthy volunteers 3 days following MDMA (3,4-methylenedioxymethamphetamine) administration, when serotonin transmission is thought to be reduced. A specific type of decision-making process, "pruning" (the reflexive avoidance of aversive outcomes when searching through a tree of potential decisions), was significantly attenuated 3 days following MDMA administration. However, the expected positive relationship between the attenuation in this decision-making process and low mood was not observed. The third experimental chapter attempted to extend this finding using the acute tryptophan depletion method, which removes tryptophan (the precursor to serotonin) from the diet and is thought to reduce serotonin synthesis. Performance on three decision making tasks (pruning, gambling and impulsivity) was examined in healthy volunteers following tryptophan depletion. Results revealed that treatment decreased participants' choosing of high probability gambles. The final chapter examined pruning in unmedicated depressed patients, and found that they behaved very similarly to healthy volunteers when evaluating aversive outcomes in the context of a tree of potential decisions, despite the hypothesised disruption to the serotonin system in this disorder. These experiments provide a more complete understanding of the relationship between serotonin, decision-making and mood, and are discussed in relation to theories of depression that pose a central role for disrupted decision-making.

LIST OF ABBREVIATIONS

^{123}I-β-CIT	^{123}I-2 beta carbomethoxy-3 beta-(4-iodophenyl) tropane
5, 7 DHT	5, 7-dihydroxytryptamine
5-CSRTT`	5 Choice Serial Reaction Time Task
5-HIAA	5-hydroxyindoleacetic acid
5-HT	Serotonin
5-HTP	5-hydroxy-l-tryptophan
5-HTT	Serotonin Transporter
8-OH-DPAT	8-hydroxy-2-(di-*n*-propylamino)tetralin
ACC	Anterior Cingulate Cortex
AIDS	Acquired Immunodeficiency Syndrome
ATD	Acute Tryptophan Depletion
ATL	Acute Tryptophan Loading
BBB	Blood-Brain Barrier
BDI	Beck Depression Inventory
BIC	Bayesian Information Criterion
BOLD	Blood Oxygen-Level Dependent
CANTAB	Cambridge Automated Neuropsychological Test Assessment Battery
CBT	Cognitive Behavioural Therapy
CGT	Cambridge Gamble Task
CPP	Conditioned Place Preference
CSF	Cerebrospinal Fluid
CxR	Choice x Risk Gambling Task
DA	Dopamine
DAT	Dopamine Transporter
DDC	Aromatic-l-Amino Acid Decarboxylase

DOI	1-(2, 5-dimethoxy-4-iodophenyl)-2-aminopropan
DRN	Dorsal Raphe Nucleus
DSM-V	Diagnostic and Statistical Manual of Mental Disorders
fMRI	Functional Magnetic Resonance Imaging
HAM-D	Hamilton Rating Scale for Depression
HIV	Human Immunodeficiency Virus
HPA	Hypothalamic/Pituitary/Adrenal axis
ID/ED	Intra-dimensional/Extra-dimensional Attentional Set-Shifting Test
ICC	Intracranial Self Stimulation
IGT	Iowa Gambling Task
LH	Learned Helplessness
LNAA	Large Neutral Amino Acid
LLO	Large Loss Optimal
MDD	Major Depressive Disorder
MDMA	3,4 methylenedioxymethamphetamine
MFFT	Matching Familiar Figures Test
MINI	Mini-International Neuropsychiatric Interview
MRN	Median Raphe Nucleus
NA	Noradrenaline
NEO	Neuroticism/Openness/Extraversion scale
NLLO	Non-Large Loss Optimal
OFC	Orbitofrontal Cortex
pCPA	Para-Chlorophenylalanine
PET	Positron Emission Tomography
PFC	Prefrontal Cortex
PKA	Protein Kinase A
POMS	Profile of Mood States

ROI	Region of Interest
RPM	Receptor Parametric Mapping
SAD	Seasonal Affective Disorder
SIS	Suicide Intent Scale
SPECT	Single-Photon Emission Computed Tomography
SPM8	Statistical Parametric Mapping 8
SPSS	Statistical Package for Social Science
SRTM	Simplified Reference Tissue Model
SSRI	Selective Serotonin Reuptake Inhibitor
SSRT	Stop Signal Reaction Time Task
STAI	State/Trait Anxiety Inventory
TPH	Tryptophan Hydroxylase
TRP	Tryptophan
VTA	Ventral Tegmental Area
WTAR	Wechsler Test Adult Reading

1) GENERAL INTRODUCTION

Major depressive disorder (MDD; also known as unipolar depression) is the most common psychiatric disorder, globally affecting more than 350 million people. It is a leading cause of disability worldwide, and in Western societies affects roughly 10% of men and 20% of women at some point during their lifetime. Further, Greenberg and Birnbaum (2005) reported that in the year 2000 MDD was the second leading cause (behind HIV/AIDS) of lost years of healthy life in adults aged 15-44, with a risk of suicide between 2% and 9% depending on the severity of the illness (Bostwick and Pankratz, 2000). As such, the economic burden of this disorder is huge: Chisholm et al (2001) reported that in the UK alone, depression is associated with direct costs of over £400 million per year for the diagnosis, treatment and rehabilitation of patients. In the US, this figure rises to $2 billion.

A major focus of research has aimed to understand the mechanisms involved in MDD, particularly to better understand dysfunction in monoamine systems, such as dopamine (DA), noradrenaline (NA) and serotonin (5-HT). These transmitter systems have been suggested to be involved in some of the most important symptoms of the disorder such as dysphoria and anhedonia. However, depression has also been associated with dysfunctions in cognitive domains such as memory, attention and executive function (in particular decision-making, which together with attention is one of the core symptoms in standard diagnostic criteria; Elliot et al, 1996). Further, pharmacological treatments that act upon the monoamine systems can have selective effects upon both mood and cognition that in some cases mimic the profile observed in MDD (Harmer and Cowen, 2013). This latter point provides indirect support for the notion of monoamine involvement in the aetiology of the disorder (McLean et al, 2004b).

The focus of this thesis is the link between 5-HT and decision-making, and whether dysfunctions in 5-HT may lead to aberrant decision-making behaviours that are often observed in MDD. This introduction will provide a review of the clinical, cognitive and biological features of MDD. We will review evidence for how 5-HT dysfunction has been implicated in abnormalities in decision-making. In particular, we will evaluate a theoretical account (Dayan and Huys, 2008) of how this relationship might lead to low mood and risk for MDD, in which they posit that low levels of 5-HT result in a decrease in the 'pruning' of aversive options from a tree of potential decisions, which in turn leads to low mood. Finally,

this introduction will consider the major research questions of this thesis before giving a brief summary of each experimental chapter, including the hypotheses examined in each.

1.1 Clinical characteristics of depression

According to the Diagnostic and Statistical Manual of Mental Disorders (DSM-V) guidelines, in order to be diagnosed with a major depressive episode, an individual must display not only low mood or anhedonia (loss of interest in, or the inability to experience pleasure from, activities that are usually found to be enjoyable, such as social interaction, work or hobbies) for a two week period or longer, but must also experience 4 or more symptoms (out of 7) in the same period of time. These symptoms include feelings of worthlessness and/or guilt, significantly altered sleeping patterns, talking or moving more slowly/quickly, fatigue or loss of energy, increased/decreased weight and/or appetite, difficulty in concentrating and making decisions, and suicidal ideation or suicide attempts (American Psychiatric Association, 2013). Furthermore, in order to gain a diagnosis of unipolar depression an individual must not have any symptoms of feeling 'up' or 'high' or hyper' (bipolar) nor psychosis. Several other non-diagnostic characteristics of unipolar depression exist, such as pessimism, rumination (particularly on negative events) decreased sociability, expectation of punishment and helplessness and a decrease in libido.

1.2 Cognitive impairments in depression

Abnormal performance on a wide range of neuropsychological tests by patients suffering from depression has been described in the literature (Beats et al, 1996, Elliot et al, 1996). Brown et al (1994) report that even depressed patients who display unimpaired normal, everyday functioning still display cognitive deficits such as language function, memory (both recall and recognition) and attention. Furthermore, Pizzagalli (2005) reported that both depressed patients and healthy volunteers with high scores on the Beck Depression Inventory (BDI) display deficits in decision-making. Such deficits are important as they may affect patients' abilities to function efficiently on a daily basis, which in turn may affect their daily experiences and thus mood, and also their ability to respond to various treatments, both behavioural and pharmacological. Research has produced evidence of deficits in memory, executive functioning, emotional processing, and decision-making in depression. This introduction will now consider each of these domains in turn.

1.2.1 Memory deficits

A very consistent finding in the literature is that depressed patients suffer from impairments on tests of episodic memory, both in recognition (e.g. Miller and Lewis, 1977, Beats et al, 1996, Elliot et al, 1996) and recall (e.g. Breslow et al, 1980, Austin et al, 1992, O'Carroll et al, 1997, Fossati et al, 2004). Roediger and McDermott (1992) however report that implicit memory deficits are not normally observed in depression. This has led to the hypothesis that depressed patients are impaired on tasks that require effortful processes, rather than automatic ones. One hypothesis that attempts to support this account is the resource-allocation hypothesis (Ellis and Ashbrook, 1998), which postulates that depressed individuals' general cognitive capacity is reduced, and as such they have deficits in remembering and engaging in other effortful cognitive processes. Other hypotheses have been put forward as to why this may be the case, from reduced motivation (e.g. Miller, 1975), and reduced cognitive initiative, which leaves the patient less able to effectively recruit and control cognitive resources (Hertel and Hardin, 1990). Indeed, studies have indicated that depression is associated with lesser memory impairments in contexts in which attention is constrained by the task: Hertel and Rude (1991) were able to eliminate depression-related memory deficits by providing instructions that focussed participants on the task and decreased the probability that task irrelevant thoughts could occur, meaning participants were more able to control their cognitive resources (see Hartlage et al, 1993 for a review). Gotlib and Joorman (2010) argue that these results suggest that depressed people may have the ability to perform at the same level as non-depressed people in constrained, structured situations, but that the opportunity to ruminate during more unconstrained situations leads to the above deficits in memory performances. As such, it could be argued that depressed patients find it difficult to control their cognitive resources, meaning that their cognitive deficits lie in the domain of executive functioning, rather than memory processes.

1.2.2 Executive function deficits

Executive functions can be considered to be a set of higher-order cognitive processes that optimise behaviour involved in complex tasks. They are often associated with a number of behavioural tasks on which performance is dependent on frontal lobe functioning, such as the Tower of Hanoi (planning: Shallice, 1982), the Wisconsin Card Sorting Test (cognitive flexibility; Nelson, 1976), the Stop Signal Task (response inhibition; Logan et al, 1984) and

the *N*-Back Test (working memory, cognitive flexibility, speeded response; Gevins and Cutillo, 1993).

Depressed patients have been shown to be impaired on a number of tasks that are thought to tap into executive functioning. For example, Rose and Ebmeier (2006) reported that patients were both slower and less accurate than healthy controls on an *N*-back test. Furthermore, Harvey et al (2004) reported that the severity of this deficit in performance on the *N*-back test was correlated with the number of hospitalisations due to depressive episodes. However, the authors here did not report poorer performance by these patients on any other tasks of working memory, including a digit span task. Consistent with this, Engelend et al (2003) argue that reduced performance on working memory tasks is due not to deficits in executive functioning, but to decreased speed and motivation (the latter of which often characterizes depressed patients; Scheurich et al, 2008).

In addition to the above deficits, depressed patients have also been shown to display poorer performance on tasks of verbal fluency (indicating difficulties in cognitive flexibility; Fossati et al, 2003), on the Tower of Hanoi (Watts et al 1988) and the computerized version, the Tower of London test (Owen et al, 1990), indicating impairments in planning. Beats et al (1996) reported impaired performance on this task in an elderly depressed sample, whilst Elliot et al (1996) showed the same in a middle-aged sample. However, Purcell et al (1997) failed to show the same in a younger sample. Studies have been able to show that patients display 'perseverative' impairments (responding to a previous rule following a rule change) on the Wisconsin Card Sorting Test (Lockwood et al, 2002, Moritz et al, 2002, Merriam et al, 1999). Performance on this task relies on the ability to form, maintain and shift an attentional set, and in order to assess each of these individually, the Cambridge Automated Neuropsychological Test Assessment Battery (CANTAB) Intra-dimensional/Extra-dimensional Attentional Set-Shifting test (ID/ED): Roberts et al, 1998 and Downes et al, 1989 was designed. Purcell et al (1997) report that a younger sample of patients displayed difficulties only in shifting attentional sets, whereas Beats et al (1996) reported that an elderly sample had difficulty in both forming and shifting attentional sets.

In sum, evidence is conflicting as to the specific cognitive deficits inherent in depression, and it is not clear whether there indeed exists a specific neuropsychological profile in the disorder. One of the reasons for this may be the lack of homogeneity between depressed

samples: the above results highlight the differences between samples of different ages (particularly in older patients, who may have more frontal lobe impairments), and a recent review (Castaneda et al, 2008) concluded that certain deficits in executive control and attentional deficits characterise depression in general, but that the evidence for learning and memory deficits is more mixed. Indeed, the authors argue that studies report deficits to be most prominent in those who are severely depressed.

1.2.3 Biased emotional processing and processing of feedback in depression

Depression is also associated with biased emotional processing and impaired emotional regulation. In line with this, early research characterised depression as a disorder associated with negative self-schemata (Beck, 1967, 1976): studies have shown depressed patients to attribute more negative adjectives to themselves in a self-referential encoding task (e.g. Clifford and Hemsley, 1987). However, there is also strong evidence for biased processing of emotional information that does not pertain to the self in depression. For example, studies have reported preferential recall of negative compared to positive memories (Mathews and MacLeod, 2005, Lloyd and Lishman, 1975, Williams et al, 1997) which in Lloyd and Lishman's (1975) study correlated positively with the severity of depression. Further, Matt et al (1992) in a review report that studies show depressed patients to remember 10% more negative words than positive, whilst 20 out of 25 studies report healthy controls to exhibit a memory bias for positive information. Some studies have also found evidence for a bias in the interpretation of events in depressed patients: Butler and Mathews (1983) report that depressed patients showed a bias towards interpreting information about ambiguous scenarios in a negative manner. Whilst some studies have failed to find such an effect (e.g. Lawson and MacLeod, 1999 and Bisson and Sears, 2007), Lawson et al (2002) argue that this is due to such studies using response latencies as the dependent variable. Further, Dearing and Gotlib (2009) reported a negative interpretation bias in never-depressed daughters of depressed mothers, indicating that these biases may be involved in an increased risk for the onset of depression. Finally, attentional biases in depressed patients have been reported, such as interferences in the Stroop colour-naming task using emotional words (Nunn et al, 1997), biases towards sad faces (Gotlib et al, 2004) and increased response times to happy words in a go/no-go task (Murphy et al, 1999).

Studies have also shown that depressed patients may display abnormal responses to negative feedback, performing poorly on trials immediately following negative feedback (Elliot et al, 1996). Shah et al (1999) did report a negative result however. Despite this, Holmes and Pizzagalli (2007) showed that healthy volunteers with high BDI scores adjusted their responses significantly less after errors on the Simon (Simon, 1969) and Stroop tasks (Stroop, 1935) than those with low BDI scores. Murphy et al (2003) report that a group of depressed patients displayed difficulties in maintaining a response in the face of misleading information on a probabilistic reversal learning paradigm. This is consistent with the results of Teasdale and Barnard (1993), which suggest that tests with feedback contain both informational components (which demand 'cold' processing of information) and emotional components (which demand 'hot' processing of information). The authors of the latter study suggest that depressed patients may have difficulty in thinking about the past failures (on previous trials) without generating a negative state of mood (e.g. without processing 'hot' information), such that if they feel they are performing poorly (potentially even in the absence of feedback) their resultant negative mood may interfere with their future performance.

In summary, studies have highlighted biased emotional processing and impaired emotional regulation in depression. Depressed patients have been shown to display a bias towards processing more negative information and a bias towards interpreting events in an overly negative manner. Further, it has been shown that patients display abnormal responses to negative feedback, both in the form of exhibiting difficulties in thinking about previous failures and responding appropriately following a failure, which has been argued to be due to difficulties in regulating 'hot' processing during such performance.

1.2.4 Decision-making deficits in depression

Just as with the link between depression and other forms of cognition, much research has been conducted into the deficient decision-making processes seen in depression (see Eshel and Roiser (2010) for a review). Studies have examined depressed patients' abilities to process information pertaining to rewards and punishments, along with their performance on tests of impulsive responding, and this section will discuss the findings of these experiments.

Much research has been conducted into depressed patients' processing of rewards and punishments, and how such processing affects the decisions that they make. In a recent

review Eshel and Roiser (2010) argue that two major conclusions arise from the literature: that depressed patients display hyposensitive responses to rewards and positive feedback, and that they show maladaptive responses to punishment.

The fact that depressed patients exhibit hyposenstivity to rewards has been identified in many studies. For example McFarland and Klein (2009) reported that depressed patients who were asked to rate their mood prior to performing a task in which correct responses were rewarded and incorrect responses were punished, displayed significantly decreased enjoyment when anticipating reward than controls, despite no difference in anxiety when anticipating punishments. Pizzagalli et al (2005, 2009) administered a task in which correct responses to one target were three times more likely to be rewarded than correct responses to another. Whilst healthy controls gained a preference for the former target, experiencing more reward, both participants with high BDI scores and participants with MDD did not. However, Huys et al (2013) argue that these deficits in depression are not due to an inability to learn from prior rewards or reinforcements (e.g. a reduction in sensitivity to prediction errors for rewards that determine reward-related learning), but are simply due to a specific reduction in reward sensitivity. Further, as discussed in *1.2.3* above, depressed patients also show maladaptive responses to punishments and display abnormal responses to negative feedback (Elliot et al (1996).

Studies using gambling tasks have also shown deficits in the processing of rewards in depressed patients; for example Smoski et al (2008) reported increased risk aversion for rewarding stimuli on the Iowa Gambling Task (IGT; Bechara et al, 1994), and Murphy (2001) reported that depressed patients display slower deliberation times and suboptimal betting strategies on the Cambridge gamble task (CGT; Rogers et al, 1999). In addition Clark et al (2011) administered this CGT to depressed patients who had a history of suicide attempts, a group of depressed patients with active suicidal ideation but without attempts, a group of depressed patients without a history of suicide attempts, and a group of healthy controls. The authors discovered that suicide attempters displayed poorer ability to choose the more likely rewarding outcome compared to non-suicide attempters, with older suicide attempters seeming to neglect outcome probability significantly more than younger suicide attempters. Studies using the CGT have also provided evidence for dysfunctional reward processing influencing early vulnerability for depression. As parental depression is the most robust risk factor for adolescents in developing depression (Rice et al, 2002), Rawal et al

(2013) examined 197 such adolescents in a 1-year longitudinal study. The authors examined participants' reward seeking behaviours on the CGT both at baseline and at 1-year, and discovered that low reward seeking on this task predicted depressive symptoms and new-onset depression at 1-year in those participants who were depression-free at baseline. Furthermore, those participants who currently exhibited depressive symptoms also displayed reduced reward seeking compared to those who were free of depressive symptomatology. The results of this study indicate that diminished reward processing may be a risk factor for those who are at risk for developing depression.

Finally, few studies have been conducted into impulsive responding in depression. However, Takahashi et al (2008) administered a temporal discounting paradigm that contained both rewards and losses to both depressed and control participants. This temporal discounting paradigm posed scenarios to participants in which they had to choose between a smaller, sooner reward, or a larger, later reward (or the opposite in the loss condition), enabling the authors to examine participants' discount factor (which describes the notion that a reward loses intrinsic value based upon its temporal delay) and the consistency with which either the sooner or later options were chosen. The results revealed that depressed patients were both more impulsive (as shown by them displaying increased discounting) and more inconsistent in their choices.

In summary, studies have found it difficult to fully characterise the cognitive profile of depression, potentially due to the heterogeneous nature of the disorder. It appears that depressed individuals may have more difficulty with controlling their cognitive resources and executive functioning such as working memory, planning and cognitive flexibility, but that these difficulties may be more apparent in older and more severely depressed patient populations. However, studies examining cognition in depression have provided more concrete evidence for decision-making deficiencies in the disorder.

1.3 Neuroimaging findings in depression

In order to understand the origins of the symptomatology and cognitive deficits and biases of MDD it may be important to utilise neuroimaging techniques, as many pharmacological treatments may lead to differences in brain structure/function (see Harmer and Cowen, 2013). Differences between depressed patients and healthy volunteers have been reported with

respect to structure, 'resting-state' neural blood flow and metabolism and task-related responses measured using blood-oxygenated-level-dependent functional magnetic resonance imaging (BOLD fMRI). Whilst not all results are consistent, a clearer picture of neural abnormalities in depression has emerged, with structures such as the amygdala, striatum, hippocampus and other limbic regions along with more cortical areas, particularly the prefrontal cortex (PFC) being implicated in the pathogenesis of this disorder.

1.3.1 Structural and resting-state functional studies implicate the amygdala, hippocampus and medial PFC in MDD

Many subcortical limbic structures have been implicated in depression, with one of the most consistent findings being that the amygdala displays abnormal functioning in MDD patients. For example, Drevets et al (2002a) have shown that this structure is overactive in melancholic depression, bipolar depression, and those at familial risk of MDD. However, such amygdala abnormalities are not always apparent in patients who meet a diagnosis for unipolar depression (see Drevets et al, 2003 for a review).

Abnormalities in the hippocampus have also been implicated in depression: MDD is recognised as a highly stress-sensitive illness (Kessler, 1997), whilst the hippocampus is a highly stress-sensitive brain-region (Thomas et al, 2007), and studies have argued that antidepressant treatments may ameliorate stress-associated changes in the hippocampus by inducing neurogenesis in this brain region (Duman et al, 2001, Santarelli et al, 2003). Many structural imaging studies have reported the hippocampus to be reduced in size in depressed patients, and a meta-analysis by Videbech and Ravnkilde (2004) reported a weighted average of an 8% and 10% reduction in left and right hippocampal volume, respectively. MacQueen and Frodl (2011) report that hyperactivity of the hypothalamus-pituitary-adrenal (HPA) axis (and the resultant increase in glucocorticoid levels) in the brain to be the cause of dendritic remodelling in the CA1 and CA3 hippocampal subfields and even decreased neurogenesis within the hippocampus. This, coupled with the fact that the hippocampus has been shown to provide negative modulation to the HPA stress hormone axis, means that dysregulation of the hippocampus can lead to sustained dysregulation of the stress hormone, which can lead to increased vulnerability of neuronal death in the CA3 field.

Studies have also shown abnormalities in the medial PFC in depression, with research showing that total PFC and anterior cingulate cortex (ACC; within the PFC) volume are decreased in depression (Drevets et al, 1997, 1998, 1998b). Results of George et al (1995) and Mayberg et al (1997) show that negative mood induction in healthy controls increases ACC activation, and results of Mayberg et al (1999) show that resting state ACC blood flow activity decreases following remission from a depressive episode. These results have led to the interpretation that once the amount of reduction in total PFC and ACC volume is accounted for, depressed individuals show a relative increase in ACC activity (Drevets et al, 1999), which Drevets (2001) argued could be the result of adaptation to the increased amygdala activity discussed above.

1.3.2 Other regions implicated in depression

A number of other areas have also been identified as displaying abnormal structure or function in depressed individuals, such as the lateral PFC, anterior thalamus, caudate and orbitofrontal cortex (OFC; see Drevets (2000) for a review). However, findings for these regions have not been as consistent as for the amygdala or medial PFC. In a meta-analysis of 143 fMRI and x-ray computed tomography studies, Kempton et al (2011) report that depressed patients display decreased caudate, putamen, globus pallidus, thalamus and OFC volume. Hamilton et al (2012) in a meta-analysis of 24 fMRI studies reported that, relative to healthy controls, depressed patients exhibited greater responses in the amygdala, insula and ACC, and decreased responses in the dorsal striatum and dorsolateral PFC when viewing negative stimuli.

1.3.3 Neural correlates of reward and punishment processing deficits in depression

Studies using neuroimaging techniques have provided a better understanding of the neural mechanisms underlying abnormal reward and punishment processing. For example, Elliot et al (1998) showed blunted responses in patients' medial caudate and OFC to both positive and negative feedback on the CGT. Steele et al (2007) report that patients displayed both reduced behavioural responses to both positive (win) and negative (lose) feedback on a novel gambling task (from Steele et al, 2004), and attenuated responses in the ACC and ventral striatum. In support of these results, Pizzagalli et al (2009) report both reduced behavioural responses and attenuated BOLD responses in the ventral striatum during reward feedback on

a monetary incentive delay task in medicated depressed patients, which in Steele's results correlated negatively with anhedonia. Further, Knutson et al (2008) reported increased dorsal ACC activity (but no differences in the striatum) to anticipated increasing gains using the same monetary incentive delay task in unmedicated depressed patients compared to controls. Finally, McCabe et al (2009) reported that remitted depressed patients displayed decreased responses to positive stimuli in the ventral striatum, and enhanced responses in the caudate and blunted responses in the lateral OFC to aversive stimuli. These differences in neural responsivity between groups were observed despite the fact that these patients displayed no clinical symptoms or differences in subjective ratings of the stimuli compared to controls. However, it must be noted that antidepressant use can have an effect upon neural responses in depression: McCabe et al (2010) administered 20mg of citalopram daily for 7 days to healthy volunteers, and discovered a reduction in the resting state connectivity between the dorsal nexus and the left hippocampus after such treatment. Connectivity between these two regions has been shown to be increased in depression (e.g. Frodl et al, 2010) and as such, it must be noted that some of the results of the above studies could be affected by antidepressant use; e.g. Steele et al (2007) administered their task to medicated MDD patients.

Few studies have examined the neural correlates of impulsive responding in MDD. However, Dombrovski et al (2013) recently examined the link between impulsivity, reward learning, depression and suicidality by administering a probabilistic reversal learning task to both elderly depressed individuals who had and had not attempted suicide, and healthy volunteers during fMRI. Measures of impulsivity were obtained from subscales of the Barratt impulsiveness scale, as well as the number of bets against the odds on the CGT, both of which the authors demonstrate to be linked to suicidality. The results of this study revealed that impulsivity and a history of suicide attempts were associated with a weakened expected reward signal on a probabilistic reversal learning task in the paralimbic cortex, and that severity of depression was associated with disrupted corticostriatothalamic encoding of unpredicted rewards in depression. As such, studies using neuroimaging techniques have been able to support the results of behavioural studies, highlighting deficient processing of rewards and punishments in medicated, unmedicated and even remitted depressed patients at the neural level.

1.4. Serotonin hypothesis of depression

Whilst the above imaging studies have helped to shed light on the neural circuits implicated in depression, theories of the neurochemical basis of this disorder have come from studies of the monoamine systems, which have long been thought to be important in the aetiology and particularly the treatment of depression (Baumeister et al, 2003). This emphasis on the monoamine systems began from the discovery that reserpine, a drug that was initially used in order to treat hypertension, induced depressive symptoms by depleting monoamine neurotransmitters in a small number of patients (Freis, 1954). There are relatively few neurons of the 5-HT, DA and NA systems in comparison to other neurotransmitter systems (e.g. the glutamatergic system) but the fact that they each have diffuse projections throughout the cortex means that they can greatly influence cognition and mood. Pharmacological treatments that block the re-uptake of one or more of these monoamines (e.g. selective serotonin reuptake inhibitors; SSRIs) are effective at alleviating the symptoms of depression (e.g. Anderson, 2000, Steffens et al, 1997). This led to the 'monoamine hypothesis' of depression, which posits that the disorder is caused by low monoamine levels (Everett and Toman, 1959). There is in fact good evidence that depleting the monoamines through pharmacological and dietary treatments can induce specific cognitive and mood effects seen in depression in both experimental animals and humans (e.g. Booij et al, 2003, McLean et al, 2004b, Riedel 2004). An overview of the current literature on the involvement of 5-HT in depression will be presented below, along with the effects of depleting 5-HT in both animals and humans. Whilst there is research indicating that DA and NA are involved in depression (e.g. Blier et al, 2003), these studies will not be discussed, as the focus of this thesis is the role of 5-HT in decision-making and mood.

1.4.1 Animal studies of 5-HT's role in depression

Animal models largely provide support for the notion that 5-HT is involved in depression, although not all results are consistent with this hypothesis. For example, the most widely used model of depression in rodents, the learned helplessness (LH) model (which is induced by exposing the rodent to inescapable and chronic stressors) has been shown to reduce 5-HT release within the PFC (e.g. Petty et al, 1994). Adell et al (1997) also report that acute exposure to a stressor can lead to increased firing in the dorsal raphe nucleus (DRN; the origin of the 5-HT system in the brain), resulting in 5-HT release in downstream projection

targets of the DRN, including the hippocampus (Amat et al, 1998) and the PFC (Yoshioka et al, 1995). Furthermore, both pre-treatment with an SSRI (Petty et al, 1996) or the selective 5-HT_{1A} receptor (an inhibitory receptor) agonist 8-OH-DPAT directly into the DRN (Remy et al, 1996, Hogg et al, 1994) can inhibit the development of LH. However, Perona et al (2008) report that 5-HT-transporter (5-HTT) knockout-mice performed no differently to wildtype (control) mice on tests of the forced swim test (to measure helplessness), tail suspension test (to measure behavioural despair) and sucrose consumption (to measure anhedonia), whilst differences in depressive-like behaviours were observed between DAT (dopamine transporter) knockout-mice and wildtype mice. The results of this study indicate that dopamine, not 5-HT, may play a more important role in animal models of depression.

As such, not all animal research supports a role for 5-HT in animal models of depression. However, it could be argued that animal models of psychiatric disorders are poor replicates of such disorders in humans: for example, the fact that administration of almost all antidepressants immediately reverses behaviours interpreted to replicate depression contrasts with the fact that antidepressants restore mood in humans only after many weeks of administration (Krishnan and Nestler, 2011). Further, it could be argued that decreased mobility on the forced swim test represents the animal learning that it cannot escape and that the best strategy is to conserve energy until it is rescued, rather than increased helplessness (e.g. Krishnan and Nestler, 2011). As such, it may be difficult to draw conclusions about depression in humans from animal models.

However, an important hypothesis regarding the involvement of 5-HT in depression stems from such animal research findings (Deakin and Graeff, 1991, Graeff et al, 1996). This hypothesis posits that there are two main 5-HT systems within the brain; one that projects from the DRN to the amygdala, and one that projects from the median raphe nucleus (MRN) to the hippocampus, with the former mediating adaptive responses to dangerous life events via 5-HT_{2C} receptors (influencing anxiety), and the latter mediating responses to life events in which loss is experienced, via 5-HT_{1A} receptors (influencing resilience to aversive events and thereby depression). Graeff et al (1996) provide empirical support for this hypothesis, by showing that decreasing DRN firing by injection of 8-OH-DPAT decreased conditioned fear (as shown by decreasing avoidance on the elevated T-maze test), whilst increasing DRN firing did the opposite. Furthermore, drug-induced increases in 5-HT within the extracellular space in the hippocampus attenuated the decrease in exploratory behaviours that occurs 24

hours after induction of a stressor, interpreted as reflecting resilience. Interestingly, this attenuation was also decreased by administration of the 5-HT_{1A} receptor antagonist WAY-100135; Graeff et al (1996). Taken together, the above studies provide some evidence to suggest that 5-HT is implicated in animal models of depression and provide convergent evidence supporting an important role for the hippocampus in the disorder.

1.4.2 Human studies of 5-HT's role in depression

The strongest evidence that 5-HT is involved in depression comes from studies using human participants, including studies showing the anti-depressant effects of drugs that increase extracellular 5-HT, the effects of acute tryptophan depletion (ATD) upon the mood of recovering depressed patients, measurements of 5-HT metabolites and tryptophan in the cerebrospinal fluid (CSF) and plasma of depressed patients, and post-mortem studies examining the expression and distribution of 5-HT receptors throughout the brain of suicide victims.

The fact that the most successful pharmacological treatments increase levels of extracellular 5-HT suggests that this neurotransmitter may be involved in the aetiology of depression. The most commonly prescribed antidepressants are the selective serotonin reuptake inhibitors (SSRIs), which have a high affinity for the 5-HTT, greatly reducing the extent to which this transporter can re-uptake 5-HT back inside the cell. These drugs have been shown to have a reliable antidepressant effects (e.g. Anderson et al, 2000), alleviating many symptoms of depression in some patients. However, it must be noted that the results of an extensive examination of the efficacy of many antidepressants showed that administration of citalopram (an SSRI) over a 12-14 week period led to roughly one third of depressed patients reaching remission, with a further 10-15% of patients exhibiting decreased symptoms without actually reaching remission. Further, it took roughly 6 weeks of daily administration for citalopram to reduce symptoms, with remission only being achieved at 7 weeks (Insel, 2006, Hierholzer, 2006). Roughly one third of the non-responders were then administered sertraline (another SSRI), following which one third of these patients became symptom free (Nelson, 2006). The results of this study therefore show that whilst administration of SSRIs improves mood in many patients, a substantial proportion of patients (30-40%) show no such improvement, indicating that 5-HT may not be the only neurotransmitter involved in this disorder.

As discussed above, there is a delay in the alleviation of depressive symptoms of anywhere between 2-6 weeks (Blier et al, 2003). This has traditionally been attributed to the 5-HT_{1A} inhibitory autoreceptors within the DRN; the autoreceptor hypothesis posits that after SSRI administration these inhibitory receptors experience increased binding by 5-HT which initially decreases DRN output. However, over time these receptors are hypothesized to become desensitized to the increased levels of 5-HT and thus cease inhibiting firing from this region, leading to increased 5-HT levels throughout the cortex. Selvaraj et al (2012) provide some support for this hypothesis by reporting that a single 10mg dose of citalopram leads to a decrease in 5-HT release throughout the cortex. However, Harmer et al (2009) recently reviewed behavioural and neuroimaging studies that have examined the effect of antidepressants, and argue that the effects of antidepressants upon patients' neural and cognitive processing of emotional and social stimuli in a more positive manner are apparent within only a couple of hours, but that mood takes longer to improve due to a resultant gradual change in social reinforcement that over time increases mood. As such, there are differing accounts of the true mechanism of antidepressant action, however the consensus is that these pharmacological treatments further indicate a role for dysfunctional 5-HT in depression.

Studies examining 3,4 methylenedioxymethamphetamine (MDMA) users have also indicated a role for 5-HT in mood. MDMA is a psychoactive stimulant that has been shown to affect the 5-HT system (Fitzgerald and Reid, 1990) by binding the 5-HT transporter (5-HTT) and reversing its function, leading to increased levels of extracellular 5-HT (Rudnick and Wall, 1992). Neuroimaging studies have shown this stimulant to affect the 5-HT system also: Kish et al (2000) report that MDMA exposure leads to decreased levels of 5-HT within the striatum, and Kish et al (2010) show decreases in 5-HTT binding throughout the cortex in chronic MDMA users (which Mcann et al (1998) showed is inversely related to the number of previous MDMA exposures). Curran and Verheyden (2003) reported that ecstasy users scored slightly less than 3 points more on the BDI than poly-drug users. However, it must be noted that neither MDMA-users nor poly-drug users scored within the clinical range for depression in this study, and Gerra et al 1998 report that MDMA-users score significantly higher on the Hamilton Rating Scale for Depression than non-drug users. Curran and Travill (1997) examined recreational MDMA users' self-reported mood levels both acutely and sub-acutely, and discovered that participants' rated their mood as elevated immediately after taking MDMA, but that their mood became progressively lower over the next 4 days, with

some participants recording mood within the range for clinical depression. The authors attribute this subsequent low mood to reflect MDMA-dependent 5-HT depletion that may occur days after administration.

Whilst not specifically related to depression, some related studies have examined levels of the 5-HT metabolite 5-hydroxyindoleacetic acid (5-HIAA) within the CSF and plasma of depressed patients, and have suggested a role for 5-HT dysfunction in suicide. For instance, 5-HIAA has been shown to predict suicide (Asberg et al, 1976) and suicidal intentions (Nordstrom et al, 1994, Samuelsson et al, 2006). Further, Jokinen et al (2007) report that 5-HIAA levels were correlated with scores on the Suicide Intent Scale (SIS) but not scores of depression or anhedonia. Further, Cremniter et al (1999) report that CSF 5-HIAA levels are lower in suicide attempters compared to healthy controls, with this effect being driven entirely by the impulsive suicide attempters. Roggenbach et al (2002) in a review of several often quoted reports of associations between aggression and levels of CSF 5-HIAA, argue that these reports perceive such a link due to an interpretation of suicide as an 'autoagressive behaviour', and that this is wrong due to the fact that aggresivity is insufficiently defined. Although, Stanley et al (2000) showed that 5-HIAA levels did correlate with self-reports of aggressive behaviour on a 6-item history of aggression scale, there was no relationship with aggressive feelings or thoughts. Consistent with the notion that self-control (impulsivity), and not aggression per se, mediates this effect, results linking 5-HIAA levels with suicidality, impulsivity and aggression are complicated by the fact that successful alleviation of depressive symptoms through the administration of SSRIs leads to a reduction in 5-HIAA (Backman et al, 2000). As such, levels of 5-HIAA within the CSF may be linked to some depressive symptoms, particularly suicide, but the exact nature of this relationship is not fully understood.

Finally, post-mortem studies of 5-HT receptor density suggest a role for altered 5-HT functioning in depression. For example, Owens and Nemeroff (1994) found reduced 5-HTT density in depressed patients, although Hrdina et al (1993) did not find such a result. However, the same study (along with McKeith et al, 1987) did find increased 5-HT_2 receptors in the post-mortem brains of depressed patients. Further, Shelton et al (2009) report that there are increased levels of 5-HT_{2A} (but not 5-HT_{1A}, or 5-HT_{2C}) receptors in the post-mortem prefrontal cortex tissue of patients with MDD, and that increased levels of 5-HT_{2A} receptors in their depressed sample was correlated negatively with levels of protein kinase A (PKA).

The results of this latter study suggest a mechanism by which depressed patients may exhibit altered levels of such receptors.

In summary, the above studies administering SSRIs to both animal and human subjects implicate a role for 5-HT in depression; and studies administrating various agonists/antagonists and post-mortem studies have provided further evidence for an involvement of specific 5-HT receptor subtypes in the aetiology of depression.

1.4.3 Mood and cognitive effects of ATD in patients recovered from depression and healthy volunteers

Many studies have attempted to link 5-HT and depression by examining the effect of ATD on both mood (see Ruhe et al (2007) for a review) and cognition (see Mendelsohn et al (2009) for a review). With regards to the mood effects, this dietary technique has been shown to have little or no effect on mood in healthy volunteers (e.g. Carpenter et al, 1998, Nishizawa et al, 1997, Riedel et al, 1999, Rogers et al, 1999a). Some studies however have suggested that ATD may decrease mood in individuals vulnerable to depression (e.g. those with a family history; e.g. Ellenbogen et al, 1999, Stewart et al, 2002), or may reinstate depressive symptoms in patients recovered from depression (Delgado et al, 1990; see Booij et al, 2002 for a review). Other studies report that patients considered to have a more severe form of depression (such as those who required the help of SSRIs to recover from a depressive episode, or those who have suffered from multiple episodes) are at higher risk of relapse following ATD (Booij et al, 2002, Delgado et al, 1999). However, O'Reardon et al (2004) found that ATD only produces this transient increase in symptoms in roughly 50-60% of patients who have recovered using SSRI treatment, and that those patients who have recovered by other means (e.g. cognitive-behavioural therapy) rarely experience any change in mood under ATD.

With regards to the cognitive effects of ATD, studies have shown differing effects upon different cognitive measures. For example, ATD has been shown to impair performance on verbal learning tasks of episodic memory (e.g. Riedel et al, 1999, Schmitt et al, 2000, Sambeth et al, 2009), and McAllister-Williams (2002) showed that ATD affected participants' episodic source memory on a word learning task. However, studies of the effect of ATD upon spatial memory, a specific type of episodic memory, have found conflicting

results, with Mendelsohn et al (2009) in a review concluding that sufficient evidence does not exist to claim ATD has an effect upon spatial memory. Further, ATD has been found to not affect declarative memory (Park et al, 1994, Porter et al, 2000, 2005) or semantic memory (Allen et al, 2006, Amin et al, 2006, Gallagher et al, 2003). However, taken together, the results of these studies indicate that ATD treatment in healthy volunteers leads to cognitive deficits that are similar to those observed in depressed patients, and as such provide convergent evidence for the serotonin hypothesis of depression.

ATD has been found to have effects on emotional processing and decision-making. For example, Murphy et al (2002) report that participants who had undergone ATD treatment displayed increased response times for happy but not sad targets. Further, Roiser at al (2008) report that ATD attenuated the bias towards positive words, and that this was accompanied by increased hemodynamic responses during the processing of these words in cortical structures including the striatum and ACC. Feder et al (2011) have also shown that relative to healthy controls who were at low familial risk for depression, those at high risk made a higher number of inappropriate responses to sad distractors on an affective go/no-go task following ATD. Munafo et al (2006) administered ATD to previously SSRI-medicated and unmedicated recovered depressed patients, along with healthy controls. The results of this study revealed that the previously medicated patients demonstrated an increase in selective processing of social threat cues on an emotional Stroop task after ATD compared to the control condition. With regards to decision-making, Rogers et al (2003) report that ATD led to reduced discrimination between magnitudes of expected gains on a gambling task, Crockett et al (2012) administered ATD to healthy controls and report a decrease in the sampling of costly information on the information sampling task, and Schweighofer et al (2008) report that ATD leads to an increase in the discounting of future rewards based upon their temporal delay; see 1.4.5 5-HT in decision-making, below).

However, studies using tasks that tap ‘executive functions’ such as working memory, attention, response inhibition, planning and cognitive flexibility have generally found no effect of ATD upon performance. For example, Luciana et al (2001) found no effect of ATD upon digit span or spatial span using the Sternberg Memory Scanning task (which measures working memory), Schmitt et al (2000) and Murphy (2002) discovered that ATD did not affect efficiency of planning (supported by Mendelsohn et al’s (2009) claim that studies have not found effects of ATD upon performance on the Tower of London task).

Similarly, studies examining effects of ATD upon attention report largely null results: Luciana et al (2001) report such null results during two types of letter cancellation tasks (although they do report that acute tryptophan loading (to increase levels of tryptophan; ATL) did decrease the number of omission errors on this task), and Harrison et al (2004) and Park et al (1994) report that ATD did not affect performance on a continuous performance task. Gallagher et al (2003) showing that ATD does not affect performance on a Stroop task, but Booij et al (2005), Schmitt et al (2000) and Scholes et al (2007) all reported that ATD improved focussed attention. However, Sobczak et al (2002) report no such improvement on the same task, which the authors claim is due to the testing of participants with a wider age-range. Therefore it seems unlikely that executive function deficits commonly reported in depression are directly related to altered 5-HT transmission.

In sum, studies administering ATD have provided support for the theory for a role of abnormal 5-HT functioning in the aetiology of depression. Both mood and cognitive effects have been observed in healthy controls, those at risk for depression and patients who have recovered from depression after administration of this dietary technique, and importantly in some cases these mimic the pattern observed in currently depressed patients, though not with respect to executive function. ATD has been shown to transiently lower mood in a proportion of patients vulnerable to the disorder, and also to lead to a temporary recurrence of some depressive symptoms in those patients who recovered using SSRI treatments, further indicating a role for 5-HT in the aetiology of depression.

1.4.4 Neuroimaging studies of 5-HT's role in depression

There have been many studies examining the role of 5-HT in depression using neuroimaging techniques. Commonly, studies have used positron emission tomography (PET) or single-photon emission computerised tomography (SPECT) in order to examine 5-HT receptor binding in depressed patients, whilst other studies have used fMRI or PET combined with a manipulation of participants' 5-HT levels in order to examine metabolism or haemodynamic responses in brain regions hypothesised to be involved in depression.

Studies using SPECT ligand ^{123}I-2 beta carbomethoxy-3 beta-(4-iodophenyl) tropane (^{123}I-β-CIT) have generally reported decreased 5-HTT binding potential in depressed patients (e.g.

Malison et al, 1998). However, Dahlstrom et al (2000) reported increased binding in depressed adolescents and children. Furthermore, a study examining patients recovered from depression revealed that these patients had a greater increase in binding than those who did not recover over a 6 month period (Laasonen-balk et al, 2004). Ruhe et al (2009) reported differences in 5-HTT binding potential between depressed and non-depressed males, but no such difference with regards to females, and intriguingly an effect of season upon 5-HTT binding potential within the midbrain. This latter result is consistent with the finding of Willeit et al (2000) who reported decreased 5-HTT binding potential in seasonal affective disorder (SAD) patients, and also the results of Neumeister et al (2000), in which it was reported that patients who were tested in the winter displayed reduced binding potential compared to those patients tested in the summer. The results of the above studies indicate that decreased 5-HTT binding potential is associated with depression, however, the results of Dahlstrom et al (2000) indicate otherwise. An interpretation of this could be that the patients studied in the latter experiment were children and adolescents, who would have experienced both fewer depressive episodes and fewer administrations of antidepressants. This could be an indication of a depression-related change in 5-HTT binding potential throughout adolescence. However, in order to better understand the results of Dahlstrom et al (2000), more research needs to be conducted. Finally, Selvaraj et al (2011), using the ligand [^{11}C]DASB in conjunction with PET, did observe decreases in the binding of this transporter in several brain regions, including the brain stem, thalamus, striatum, ACC and PFC, supporting the above SPECT studies.

Studies using PET to observe 5-HT$_{1A}$ receptor binding in depression have provided some evidence for its dysfunction in the disorder, although relatively few studies exist. Using the ligand ^{11}C-WAY100635, Drevets et al (1999) (replicated by Drevets et al, 2007) and Sargent et al (2000) have reported reduced binding in both the raphe and cortex in depressed patients, and Bhagwagar et al (2004) and Moses-Kolko et al (2003) report (using the same ligand) reduced 5-HT$_{1A}$ receptor binding potential throughout the cortex in recovered depressed patients. Moses-Kolko et al (2007) shed light on the effect of SSRI treatment upon 5-HT$_{1A}$ receptor binding potential in MDD; here the authors discovered that out of 22 depressed patients who were administered an SSRI for 9 weeks, the treatment non-responders had higher baseline 5-HT$_{1A}$ receptor binding potential in the OFC bilaterally than treatment responders. Further, the authors discovered that the antidepressant drug treatment did not significantly change cerebral 5-HT$_{1A}$ receptor binding potential in treatment responders,

which they argue supports findings that alterations in 5-HT functioning after SSRI treatment do not involve changes in 5-HT_{1A} receptor density. However, Shrestha et al (2012) report in a recent review that that not all studies support these results, highlighting the fact that Parsey et al (2006, 2010) reported an *increase* in 5-HT_{1A} receptor binding potential in MDD patients compared to healthy controls. However, Shrestha et al argue that these discrepant results were observed due to both heterogeneity between patient samples, and differences in methodology between studies, with those studies reporting an *increase* in MDD 5-HT_{1A} binding potential (Parsey et al, 2006, 2010) using the cerebellum as a reference regions, whereas the others above used radioactivity concentration in the plasma. As such, whilst the above studies indicate a role for decreased 5-HT_{1A} receptor binding potential in MDD (with such decreased levels in the OFC also being related to increased potential for treatment response), the methodological differences between studies need to be rectified in order to gain a better understanding of the role of this receptor in MDD.

Studies attempting to observe 5-HT_2 receptor binding in depression have provided less consistent results however, potentially due to the fact that more than one ligand has been deployed in these studies (only one receptor ligand was used to look at 5-HT_{1A} receptors – ^{11}C-WAY-100635 – as this was the only one that was available until relatively recently). Some studies have reported decreased 5-HT_2 receptor binding in depressed patients compared to controls (e.g. Attar-Levy et al, 1999, Messa et al, 2003, Larisch et al 2001), whilst others report no difference (Meltzer et al, 1999, Meyer et al, 1999). Meyer et al (2003) were able to show that 5-HT_2 binding was positively correlated with negative dysfunctional attitudes in a depressed sample, which is supported by results of Meyer et al (2001) that show decreased 5-HT_2 binding following recovery from depression (though results of this latter study are contrary to results of Massou et al, 1997 and Zanardi et al, 2001). As such, results regarding 5-HT_2 receptors in depression are conflicting.

Lastly, studies using PET and fMRI have also attempted to image the neural correlates of the relapse in patients recovered from depression observed after ATD. Firstly, Bremner et al (1997) reported that ATD both induced symptom increases in these patients and was linked to decreased glucose metabolism within regions of the dorsolateral and ventrolateral PFC, OFC, ACC and the thalamus, with the levels of metabolism correlating negatively with increased mood scores. This is supported in part by Smith et al (1999) who, using $H_2{}^{15}$O-PET, reported that increasing levels of depression after ATD were correlated with diminished blood flow in

ACC, OFC and caudate nucleus in patients recovering from depression. However, Neumeister et al (2004) report on a sample of depressed patients who were free from antidepressants (unlike the studies above), and report increased glucose metabolism in the OFC, posterior cingulate cortex, thalamus and right putamen following ATD treatment, which is the opposite to findings found in the above studies that used patients who *were* being administered antidepressants. Finally, Roiser et al (2009), using fMRI, discovered a differential effect of ATD upon BOLD responses in the dorsal ACC to emotional relative to neutral words in remitted depressed patients compared to healthy volunteers, despite no difference in mood between the groups following ATD.

As such, these studies provide further results that implicate 5-HT in depression, and show that a rather consistent set of neural circuits underlies mood changes following ATD treatment, through the precise direction of changes observed does appear to depend on medication status. Importantly, many of these brain regions within this circuitry have been implicated in depression from neuroimaging studies using fMRI (see 1.3 Neuroimaging findings in depression, above).

1.5 Serotonin in Decision-Making

Much research has been conducted into the role of 5-HT in decision-making (e.g. Dayan and Huys, 2008), but it must be noted that decision-making includes many facets including reward and punishment processing (e.g. Rogers et al, 1999b, 2003, Eshel and Roiser, 2010) and impulsivity (e.g. Schweighofer et al, 2008, Pine et al, 2009, Dalley and Roiser, 2012). Studies have implicated 5-HT in each of these areas of decision-making, and this section will discuss each in turn.

1.5.1 Serotonin and reward and punishment processing

1.5.1.1 Animal studies of serotonin in reward and punishment processing

Decision-making has been defined as computing predictions of future consequences of possible actions, and selecting a behaviourally appropriate response based upon those predictions (Dayan and Huys, 2008). A rich animal literature has measured reward-related behaviour in rodents using three main paradigms: place conditioning, intracranial self-

stimulation (ICC) and drug self-administration. The results of these studies have reported the processing of reward-related information to be regulated by the action of 5-HT at a range of receptors.

For example, Ahn et al (2005) report on the dose-dependent effects of the 5-HT_{1A} receptor agonist 8-OHDPAT in reward responding, by showing that injection of a low dose into the rodent raphe nucleus (which decreases 5-HT neuron firing) led to increased responding to rewards on a conditioned place preference (CPP) task, but that injection of a high dose into the same brain region led to a decrease in responding. This is supported by results reported by Harrison and Markou (2001), but Markou et al (2005) and Budygin et al (2004) found that 5-HT_{1A} receptor antagonists have no effect. Studies have also shown dose-dependent effects of 5-HT_{1A} receptor agonists upon drug self-administration, with Peltier and Schenk (1993) showing high doses to inhibit the rewarding effects of cocaine self-administration in rats.

In general, stimulation of 5-HT_{1B} receptors has been shown to decrease the rewarding effects of rewards such as intracranial self-stimulation, cocaine, ethanol and amphetamine (e.g. Hayes et al, 2009b, Harrison et al, 1999, Fletcher et al, 2002a). However, Hoplight et al (2006) report an *increase* in the rewarding effects of ethanol following increased 5-HT_{1B} receptor expression in the ventral tegmental area (VTA) of rodents, though this result may reflect effects on the dopamine system.

Results regarding 5-HT_2 receptors are less consistent, with administration of 5-HT_{2A} receptor antagonists having no effect on the rewarding effects of intracranial self-stimulation or cocaine self-administration (Moser et al, 1996), but having an attenuating effect of self-administration of MDMA (Fantegrossi et al, 2002) and ethanol (Ding et al, 2009) when injected directly into the VTA. However, Hayes and Greenshaw (2011) argue that this may be due to the fact that 5-HT_{2A} receptors in the VTA increase dopamine release. No effects of 5-HT_{2B} or 5-HT_3 receptor stimulation upon reward processing have as of yet been published (Hayes and Greenshaw, 2011).

Early animal studies focussed more on the role of 5-HT in punishment processing (see Soubrie (1986) for a review). Studies that have administered both 5-HT receptor antagonists and manipulations known to decrease 5-HT release in terminal areas, as well as lesion studies (using 5, 7 dihydroxytryptamine), have shown that decreased 5-HT release results in

attenuation of punishment-induced inhibition (e.g. Thiebot et al, 1982, Tye et al, 1977, Tye et al, 1979, Wise et al, 1970), whilst Wise et al (1973) report that these effects can be reversed by direct injections of 5-HT or 5-HT agonists. Further, studies have shown that lesions of the raphe nuclei or administration of para-chloropheylalanine (pCPA) to deplete 5-HT can cause a deficit in passive avoidance of footshocks, and that the effect of pCPA is attenuated by 5-HT administration (Thornton and Goudie, 1978).

In summary, animal studies have provided some evidence for a role for 5-HT in reward and punishment processing, with numerous findings that 5-HT depletion disinhibits punishment processing. However, different receptors have been shown to have different effects upon responding to rewards and punishments: 5-HT_{1A} receptor agonists have been reported to have a dose dependent increase upon the processing of the rewarding aspects of drug self-administration, and stimulation of 5-HT_{1B} receptors has been shown to decrease the rewarding effects of stimuli. However, less is understood about the role of specific 5-HT receptors in punishment processing.

1.5.1.2 Human studies of serotonin in reward and punishment processing

Studies using human participants have also implicated 5-HT in the processing of rewards and punishments. For example, Anderson et al (2003) administered a novel gambling task in which participants who had undergone ATD treatment could choose between option 'A' which provided a smaller but nearly certain win, and option 'B' which provided a win 2.5 times that of 'A' but with a varying probability of occurring. The results of this study revealed that those who had undergone ATD treatment were more likely to take a risk for smaller rewards, but that ATD treatment did not modulate probabilistic choice overall. Rogers et al (1999b) administered the CGT to participants who had undergone ATD treatment, and found that treatment increased the number of sub-optimal decisions made, and increased deliberation times before making a decision. Further, Talbot et al (2006) discovered that ATD treatment had no effect on set shifting or reversal learning on the ID/ED set-shifting task of the CANTAB, and even risk taking (contrary to the results of Anderson et al, 2003 above, which used a different task). However, the authors here did report increased choosing of the more probable outcomes after ATD compared to sham depletion. Rogers et al (1999a) report that healthy volunteers who had undergone ATD treatment exhibited a deficit in the ability to learn changed stimulus-reward associations. Further, Rogers et al (2003)

administered a decision-making task that allowed the authors to examine the amount healthy volunteers (who had undergone ATD) used information pertaining to wins, losses and the probability of which they would win or lose in making their decisions. The results of this study showed that ATD attenuated participants' sensitivity to wins (rewards), but not losses (punishments) or probabilities. By contrast, Robinson et al (2012) report that ATD improved healthy controls' ability to predict upcoming punishments (but not rewards) on a reversal learning task, which supported the results of Cools et al (2008), who reported that ATD abolished the bias towards prediction errors (the difference between expected and actual outcomes) for punishing stimuli compared to rewarding stimuli. Crockett et al (2009) showed that ATD abolished punishment-induced behavioural inhibition on a go/no-go task without affecting overall motor response inhibition. Furthermore, Crockett et al (2012) report that ATD removed the suppressive effects of small local costs on information sampling in healthy volunteers on an information sampling task. In summary, behavioural studies using ATD suggest that there may be some role for 5-HT in both reward and punishment processing in humans, though many discrepant findings have been reported, and it is difficult to draw firm conclusions from the extant literature due to the large range of tasks employed.

Neuroimaging studies in humans have also shown reward and punishment processing to be linked to 5-HT. Seymour et al (2012) administered a four-arm bandit task to healthy volunteers who had undergone ATD that allowed the authors to simultaneously examine participants' responses to both rewards and punishments, and found that ATD altered the exchange rate by which rewards and punishments were compared, decreasing the subjective value of rewards, which was related to an increase in striatal and PFC responses. Few studies using fMRI have examined the role of specific receptor subtypes in this form of decision-making however, although one recent study has done so: Macoveanu et al (2013) reported on the effects of the 5-HT_{2A} receptor in risky decision-making. The authors administered a probabilistic gambling task to participants who were either administered the 5-HT_{2A} receptor antagonist ketanserin or a placebo. The results of this study showed that blockade of the 5-HT_{2A} receptor made participants more risk-averse and selectively reduced the neural response of the PFC to negative outcomes that occurred on low risk trials. Furthermore, the ventral striatum displayed a stronger response to low-risk negative outcomes at baseline in risk-taking compared to risk-averse individuals, which was abolished after administration of ketanserin.

1.5.2 Serotonin and impulsive responding and the processing of delayed rewards

1.5.2.1 Animals studies of serotonin and impulsive responding and the processing of delayed rewards

Research into impulsive responding to rewards and punishments using animals has shown that rats who have undergone 5, 7-dihydroxytryptamine (5, 7 DHT) administration (to deplete 5-HT globally) exhibit increased premature responding on the 5-choice serial reaction time task (5-CSRTT; Harrison et al, 1997). In support of this, Masaki et al (2006) report that depleting 5-HT via administration of parachloroamphetamine (pCA) leads to impairments on a go/no-go paradigm. However, Eagle et al (2008) show that modulating 5-HT functioning only affects 'action restraint' in rodents, and does not affect performance on the stop-signal reaction time task (SSRT) or temporal discounting tasks (supported by Eagle et al, 2009, Bari et al, 2009). However, the role of 5-HT in delayed rewards is controversial. Initially Bizot et al (1988) showed that rats on a range of antidepressants chose delayed but larger rewards (e.g. decreased discounting). However, Winstanley et al (2003, 2004) reported no effect of 5-HT lesions in performance on a temporal discounting paradigm, which may be due to differences in experimental procedures (Winstanley et al, 2006a, Dalley and Roiser, 2012).

Further insights have come from studies examining the effects of 5-HT receptor stimulation/blockade on impulsive responding: Blokland et al (2005) and Evenden and Ryan (1999) found that administration of the 5-$HT_{2A/2C}$ receptor agonist DOI (1-(2, 5-dimethoxy-4-iodophenyl)-2-aminopropan) increases impulsivity on both reaction time and temporal discounting tasks, an effect that can be reversed by administration of 5-$HT_{2A/2C}$ receptor antagonists, and Winstanley et al (2004) reported an effect of DOI on performance on the 5-CSRTT. However, Talpos et al (2006) found no such effect by administering the 5-$HT_{2A/2C}$ receptor antagonist SER082, and Blokland et al (2005) showed that systemic administration of the 5-HT_{1A} receptor agonist 8-OH-DPAT decreased impulsivity on a choice reaction time task but increased temporal discounting. This latter result highlights the fact that impulsivity is not a unitary construct and that different tasks of impulsivity do not tap into the exact same decision-making processes (Dalley and Roiser, 2012).

1.5.2.2 Human studies of serotonin and impulsive responding and the processing of delayed rewards

Whilst research into impulsive responding in humans does not have the range of pharmacological interventions, there nonetheless exists a consistent body of work that implicates 5-HT in this form of decision-making. The dietary technique ATD has often been used in order to examine 5-HT's role in impulsive decision-making: with regards to response inhibition, several studies report no effect of ATD: Clark et al, (2005), and Evers et al (2006b), Rubia et al (2005) and LeMarquand et al (1998, 1999) found no change in error rates on a go/no-go task after ATD treatment. The results of studies using the SSRT have been very consistent with those using animals, finding no effect of ATD on behaviour (e.g. Cools et al, 2005a, Clark et al, 2005). However, Walderhaug et al (2002) did report disinhibited behaviour following ATD in healthy controls, and Crean et al (2002) found that ATD increased stop reaction times in participants who had a family history of alcoholism. With regards to temporal discounting, results using human participants have been much more consistent than those using animals: Tanaka et al (2007) report that ATD increased healthy volunteers' rate of discounting, and Schweighofer et al (2008) tested healthy volunteers who had undergone both ATD and ATL on a 'dynamic' delayed discounting paradigm that required a continuous update of reward value estimates. The results of this latter study showed an increase in the rate of discounting delayed rewards in the low tryptophan group compared to baseline and high tryptophan groups. However, it should be noted that Crean et al (2002) reported no effect of ATD upon temporal discounting in a sample of men both with and without a family history of alcoholism.

Studies using other methods have also provided support for a role of 5-HT in this form of decision-making. For instance, Lindstrom et al (2004) and Ryding et al (2006) report reduced 5-HTT levels as shown by PET in impulsive compared to non-impulsive suicide attempters, which the authors claim could be due to reduced density of 5-HT terminals. Further, Frankle et al (2005) report similarly reduced levels of 5-HTT in impulsive compared to non-impulsive aggressive individuals.

The psychoactive drug MDMA has been shown to affect the 5-HT system (e.g. Rudnick and Wall, 1992, Kish et al, 2000, 2010), and studies examining decision-making in human MDMA users have provided some, although often conflicting, evidence for the role of 5-HT

in the processing of delayed rewards and impulsive responding. For example, Quednow et al (2007) reported on the decision-making and impulsive responses of chronic but recently-abstinent MDMA users by comparing them with both chronic but recently-abstinent cannabis users and controls. The results of this study revealed MDMA users to display higher levels of impulsivity on the Matching Familiar Figures Test (MFFT; Kagan et al, 1966) and poorer performance on the IGT. These findings are supported by Morgan et al (2002) and Bickel and Marsch (2001), who were able to show that regular ecstasy users display increased impulsive responding compared to both poly-drug users and drug-naïve controls on the MFFT and a temporal discounting paradigm, respectively. However, Clark et al (2009) reported that a cohort of previous ecstasy users and current ecstasy users did not display disrupted reflection impulsivity compared to drug-naïve controls on an information sampling task, despite the previous and current ecstasy users scoring significantly higher on the impulsivity subscale of the self-report Eysenck Impulsivness-Venturesomeness-Empathy questionnaire (interestingly the previous users scored (numerically) higher on this subscale than current users). However, it should be noted that there are limitations of studies of MDMA users, specifically that these participants often ingest various other drugs that can confound results, and that such cross-sectional studies cannot exclude the possibility that any differences observed were pre-existing. This makes any attempt to draw conclusions regarding links between MDMA use, 5-HT and task performance difficult.

Relatively few neuroimaging studies have studied the neural correlates of 5-HT's influence on impulsive responding. However Tanaka et al (2007) were able to show a role of 5-HT in temporal discounting using fMRI: here they discovered that BOLD responses within the ventral striatum were correlated with reward prediction at shorter time scales, which was stronger at lower levels of 5-HT. Further, BOLD responses within the dorsal portion of the striatum were correlated with reward prediction at longer time scales, which was stronger at higher levels of 5-HT. Whilst the results of this study support a role for 5-HT in temporal discounting by altering striatal responding, more research needs to be conducted in order to gain a more complete understanding of the neural circuitry involved in the link between 5-HT and impulsive responding.

1.6 Computational models of decision-making and the theory of altered pruning in depression

The behavioural, psychopharmacological and imaging studies reported above all utilised descriptive approaches to their analyses of the data, using 'raw score summary statistics' that simply average over trials at the subject level in order to produce the outcome measures of interest, such as average reaction times or proportion of choices made. An alternative approach to understanding participants' performances is computational modelling. Such an approach involves building a mathematical algorithm that putatively describes how participants perform a task (often explaining changes in behaviour over time - e.g. learning) and summarises performance according to a set of parameters that have been estimated on the basis of participants' behaviour (Montague, 1995).

This approach has allowed recent studies to provide novel insights into decision-making differences between depressed and non-depressed individuals. Applying models that attempt to explain performance in this way may help researchers understand more about why participants made particular choices: if an extra parameter is added to a model, for example, a parameter that influences reward sensitivity (which indexes the subjective value placed upon a reward that was administered) and this parameter increases the likelihood that this new model will be able to predict participants choices, then the inference can be made that participants are indeed using reward sensitivity information to guide their choices. Additionally, parameter estimates from models that accurately explain participants' behaviour (e.g. provide a good fit to the data) can be compared between groups, allowing a computationally and theoretically precise analysis of specific cognitive processes. Thus computational models can be informative over and above traditional statistical models which can help researchers to understand why certain behaviours on certain tasks are observed. In this thesis, such a computational approach was utilised in two of the chapters to characterise decision-making on a 'pruning' task (see below).

Whilst the advent of such models was rather recent, meaning that a large body of work does not yet exist, computational theories of decision-making in depression have been proposed, and a particularly relevant theory for this thesis is the theory of altered 'pruning' in depression which is discussed below (Dayan and Huys, 2008, Huys et al, 2012).

Early behavioural studies using the computational modelling approach did not apply such methods to study depression. However, Huys and Dayan (2009) were able to show that certain computations are linked to specific depressive symptoms. Administering a range of decision-making tasks, the authors were able to compute measures of reward sensitivity (how much a previous reward affected future choices) and controllability (the participants' belief that their action will lead to a particular set of outcomes – the opposite of 'helplessness'). The results showed that reward sensitivity and controllability were negatively correlated with self-reported anhedonia and self-reported helplessness, respectively. Furthermore, Chase et al (2010) tested both MDD subjects and controls on a probabilistic learning paradigm, and found that learning rates (the extent to which stimulus value estimates are updated following feedback) were negatively correlated with self-reported anhedonia in both groups, although the groups did not differ overall. Thus, studies using computational modelling of decision-making behaviour are beginning to provide insight into the specific decision-making processes in depression.

1.6.1 Theory of altered pruning in depression

Dayan and Huys (2008) posit a theory based upon computational work that is important to the studies and hypotheses set out in the chapters below. This work highlights how computational modelling may be able to aid our understanding of the role of neurotransmitters in psychiatric conditions, and is based upon early work in rodents that implicates 5-HT in both the prediction of aversive events (e.g. Deakin, 1983) and behavioural inhibition (Soubrie, 1986; see section *1.5.1.1* above). This theory posits that 5-HT is involved in the prevention of ongoing actions or thoughts in light of aversive events. Dayan and Huys (2008?) argue that many of the decisions made on an everyday basis are not single-step decisions, but rather require evaluation of rewards and punishments at many stages. Beyond a few steps, these kinds of planning problems cannot be solved by evaluating all potential sequences one by one, since too many alternatives would need to be evaluated: instead strategies, or heuristics, must be employed that allow one to eliminate sequences in order to lessen the computational load. As such, possible alternative action sequences are 'pruned' away from a 'tree' of potential decisions, and 5-HT is posited to be involved in such pruning for such sequences that include highly aversive outcomes. For example, when deciding upon a holiday destination, one may prune based on a number of heuristics; one may wish to prune away regions that are too expensive to travel to (e.g. Australasia), or those that one has

already travelled to (e.g. North America), or which are currently experiencing social unrest and thus could be unsafe (e.g. the middle east). Once a decision has been made (e.g. Asia), countries within that continent that are less attractive to the holiday-maker may be pruned away, until a decision is made. To consider all of the hotels, in all the towns, in all the world would be computationally ruinous, and pruning away large portions of the tree of decisions lessens this load.

Dayan and Huys et al (2008) and Huys et al (2012) argue that 5-HT is involved in pruning this tree of decisions in light of potentially aversive events, and that a decrease in levels of 5-HT could result in a decrease in behavioural inhibition, leading to decreased pruning. This is then hypothesised to lead to an increase in large negative prediction errors as more unexpected, negative consequences are encountered, leading to a more pessimistic evaluation of the world, and a decrease in mood. However, healthy controls (with normal levels of 5-HT) should reflexively prune away choices with aversive expected outcomes, and would thus underexplore negative environments leading to the experiencing of (and thinking about) fewer negative experiences and a more optimistic view of the world. The authors thus argue that it would be important to test this theory in MDD patients and subjects with high scores on depression scales in order to better understanding the link between decision-making, 5-HT and low mood.

The specific heuristics that humans deploy when pruning are difficult to ascertain. However Huys et al (2012) administered to healthy controls a sequential decision making task that was designed to reveal such specific pruning strategies based on aversive outcomes. On this task, healthy participants had to devise a sequence of moves of pre-specified length, with each move being deterministically associated with either a large or small financial win or loss. Three separate conditions were performed in a between subjects design, with group specific large losses of either £1.40, £1.00, or 70p. Due to the financial rewards and punishment associated with the other 3 move types (win of £1.40, win of 20p, loss of 20p), it was increasingly financially disadvantageous to cut the ever expanding decision tree down to a manageable size by pruning sub-trees that begun with a large loss in each condition (e.g. most disadvantageous in the 70p loss condition). However, participants pruned such sub-trees equally in all 3 conditions, extensively reducing their search space by failing to consider sequences of moves that began with a large loss.

Computational modelling of participants' choices was applied to the data to confirm that they were indeed employing a heuristic in which planning to transition through large losses was avoided (even when this strategy was financially disadvantageous). The fact that participants continued with this pruning behaviour, even when it was highly disadvantageous (e.g. in the 70p loss condition) indicated that it was reflexive, elicited in response to large losses in a Pavlovian manner that is non-adaptive and inflexible to task demands. Further, the authors also discovered a correlation between sub-clinical depression scores on the BDI and pruning behaviours. This correlation was, however, in opposition to the direction predicted by Dayan and Huys (2008), with a higher depression score being correlated with a *higher* pruning score. As such, whilst the employment of computational models in this study allowed for a better understanding of the link between pruning, 5-HT and mood, more work needs to be performed, with Huys et al (2012) pointing out that future studies should examine pruning in clinically depressed patients.

1.7. Major questions and aims of this thesis

The main aim of this thesis is to examine the link between 5-HT, decision-making and mood. Due to the putative disruption of 5-HT functioning in depression, it will examine how dysfunctional 5-HT is linked to the altered processing of information pertaining to rewards and punishments that may lead to the observed dysfunctional decision-making in depression.

This thesis will present data from 4 experiments in which tasks tapping different aspects of reward and punishment processing were administered. The main task of focus is the pruning task used in Huys et al (2012) which was administered in every study. From this there followed 2 main aims: firstly, to replicate findings from Huys et al (2012) in which it was discovered that healthy controls make computationally complex, multi-step decisions by curtailing the search of a tree of potential decisions in light of potentially aversive events (pruning): secondly to test Dayan and Huys' (2008) hypotheses that 5-HT is involved in this pruning process, and that impaired pruning behaviours are linked to low mood. This was tested by examining the pruning behaviours of both healthy controls who have undergone administration of MDMA (chapter 4) and ATD (chapter 5), and unmedicated unipolar depressed patients (chapter 6). This thesis also presents data from a study that attempted to examine whether such pruning behaviours were related to the availability of a specific serotonin receptor, the 5-HT_{1A} receptor (chapter 3).

This thesis will also present data from two other tasks of reward and punishment processing; a gambling task described in Rogers et al (2003) and a temporal discounting paradigm described in Pine et al (2009). The first aim with regards to these studies was to confirm that performance on these tasks is dependent upon 5-HT functioning by replicating findings (chapter 5) from previous studies which, using ATD, have shown decreased levels of tryptophan to lead to a decrease in sensitivity to rewards (Rogers et al, 2003) and to increased discounting of rewards based upon their temporal delay (Schweighofer et al, 2008). The second aim was to extend these findings by showing a link between performance on these two tasks and availability of the 5-HT_{1A} receptor (chapter 3).

1.8 Summary of thesis chapters

Chapter 2 details the various techniques used in the experiments described in chapters 3-6. This chapter is set out into 3 sections, with the first detailing the psychometric questionnaires and interviews administered, the second describing the behavioural tasks that were administered, and the third detailing the experimental techniques used to examine the impact of 5-HT on behaviour - one imaging technique (PET) which allowed the observation of 5-HT_{1A} receptor binding and putative 5-HT release, and two 5-HT manipulation techniques, MDMA and acute tryptophan depletion administration.

Chapter 3 details the results of a study in which participants were given 2 PET scans in order to observe both availability of the 5-HT_{1A} receptor and SSRI-induced 5-HT release in healthy controls, which were then correlated with performance on 3 decision-making tasks from a separate testing session. These behavioural tasks allowed the examination of pruning behaviours (using a sequential decision-making task), risky decision-making (using a risky decision-making task) and delay aversion (using a temporal discounting paradigm). It was hypothesised that performance on the above tasks would be correlated with both baseline 5-HT_{1A} receptor availability, and 5-HT release due to infusion of the SSRI citalopram in both the striatum and hippocampus.

Chapter 4 details the results of a study in which participants were administered an acute dose of MDMA, and then 3 days later completed both self-report mood questionnaires and the pruning task. This was performed in order to examine the subacute effects of MDMA

administration (and the putative decrease in 5-HT thought to occur at this time) upon performance on this task. It was hypothesized that compared to placebo, participants would display decreased pruning behaviours due to the decrease in 5-HT, and that this would be linked to decreases in mood.

Chapter 5 details the results of a study in which participants underwent ATD (which is hypothesized to lead to decreased levels of 5-HT in the brain) and performed 3 decision-making tasks (the pruning task, and the risky decision making and delay aversion tasks administered in chapter 3). It was hypothesized that participants who had undergone ATD treatment would decrease pruning behaviours, decrease discrimination between magnitudes of wins on the gambling task, and increase participants' discounting of future rewards on the temporal discounting paradigm. It was also hypothesised that ATD treatment would not alter mood.

Chapter 6 details the results of a study in which the pruning task was administered to both depressed patients and healthy controls. It was hypothesized that depressed patients would display decreased pruning behaviours compared to controls, and that this would be more marked in more severely depressed patients.

Chapter 7 provides a summary of the major findings of each experimental chapter, in particular focussing on the relationship between decision-making, 5-HT and mood. It compares the effects of the 5-HT manipulations, variation in 5-HT_{1A} receptors, and effects of MDD upon performance on the above 3 decision-making tasks. Finally, it considers limitations of these studies and directions for future research.

2) EXPERIMENTAL METHODS

This chapter details the various techniques used to collect the data that are reported in chapters 3-6. It will first outline the various mood and personality rating questionnaires and that were employed, before describing the computerized cognitive tasks that were administered and the chapters in which they appear. The method of positron emission tomography used in chapter 3 will be described, after which the method of administering MDMA in order to manipulate the 5-HT system (as in chapter 4) will be outlined, as will the technique of acute tryptophan depletion (ATD), which is thought to selectively lower levels of 5-HT synthesis, that was used in chapter 5. In all studies participants were compensated for their time and provided written informed consent to participate. Studies were approved either by the UCL Ethics Committee or the London Queen Square NHS Ethics Committee.

2.1. Mini-International Neuropsychiatric Interview (MINI) – Sheehan et al (1998); Chapters 3, 5 and 6

The MINI is a short, structured clinically diagnostic interview developed for the DSM-IV and ICD-10. Each section asks the participant about different psychiatric conditions; the interviewer asked questions from sections on unipolar depression, bipolar depression, panic disorder, agoraphobia, obsessive-compulsive disorder, post-traumatic stress disorder, alcohol and substance abuse/dependence, psychotic symptoms, anorexia nervosa, bulimia nervosa and generalized anxiety disorder. Each section began with a simple question (i.e. for unipolar depression: 'Have you been consistently depressed or down most of the day, nearly every day for the past two weeks?'). If the participant responded yes to any of these first questions, follow-up questions, according to the specific criteria, were asked in order to assess whether they met criteria for the relevant condition. Kotwicki and Harvey (2013) report that the MINI substantially improves upon the stability of diagnoses compared to unstructured procedures relying on clinician diagnoses. Further, the authors report that the differences in rates of changes in diagnoses between the Structured Clinical Interview for DSM disorders are non-significant ($chi^2(1)=2.01$, $P=.16$).

2.2 Mood/Personality Questionnaires

2.2.1 Hamilton Rating Scale for Depression (HAM-D) – Hamilton (1960); Murphy et al (2002); Chapters 5 and 6

The HAM-D is conducted in a structured interview and provides a measure of the severity of depressive symptoms. The interviewer asks the participant questions and then rates their answers, from 0-3,4 or 5, depending on the question. There were 17 questions on this questionnaire, with a maximum score of 49. A score of 0-7 is considered normal; 8-13 means mild depression, 14-18 moderate depression, 19-22 severe depression, and 23+ very severe depression. This was administered to measure severity of mood disturbance. A sample question was 'Depressed Mood', with the possible answers being 0 (Absent), 1 (Sadness etc), 2 (Occasional weeping), 3 (Frequent weeping), 4 (Extreme Symptoms). Kobak et al (1999) report that this measure has an internal consistency of .90, and a test-retest reliability of .74.

2.2.2 Profile of Mood States (POMS) – Biehl and Landhauer (1975); Chapter 6

The POMS is a self-rating scale in which participants must rate themselves as either 'not at all', ' a little', 'moderately', 'quite a bit' or 'extremely' similar to each of the 65 adjectives presented to them. The items load onto 6 factors; 'tension-anxiety', 'depression-dejection', 'anger-hostility', 'fatigue-inertia', 'vigour-activity' and 'confusion-bewilderment', with participants having to score between 1 (not at all) and 5 (extremely). Wywrich and Yu (2011) report that the internal consistency of this measure is .84, and the test-retest reliability is 0.78.

2.2.3 Beck Depression Inventory (BDI) – Beck et al (1961); Chapters 3, 4, 5 and 6

The BDI is a 21-question self-rating scale used to determine depressive symptomatology. For each item the participant is asked to choose one of four statements that best suits their mood over the previous two weeks. This scale was modified for administration in chapter 4 to examine participants' mood over the past 3 days. Statements for each item are given a score, from 0-3, depending on their severity. The maximum score is thus 63, but the BDI manual (Beck and Steer, 1987) recommends the following classifications; 0-9 normal mood, 10-16 mild mood disturbance, 17-20 borderline clinical depression, 21-30 moderate depression, 31-40 severe depression, and 41+ extreme depression. A sample question from this measure was

to rate either 0 (I do not feel sad), 1 (I feel sad), 2 (I am sad all of the time and cannot snap out of it) and 3 (I am so sad or unhappy I cannot stand it). Storch et al (2004) report the internal consistency of this measure to be 0.90.

2.2.4 Neuroticism/Extraversion/Openness Scale (NEO) – Costa and McCrae (1985) Chapters 3, 5 and 6

The NEO is a 60-item, self-rating personality measure, measuring the five personality traits of Extraversion, Agreeableness, Conscientiousness, Neuroticism and Openness. Participants are presented with the 60 statements, and must decide whether they 'strongly disagree', 'disagree', are 'neutral' towards, 'agree' or strongly agree' with each. A sample question would be 'I am not a worrier' and I rarely feel alone or blue'. McCrae et al (2011) report that the internal consistency for the neuroticism scale is 0.55, for the extraversion scale is 0.64, for the openness scale is 0.59, for the agreeableness scale is 0.58 and for the conscientiousness scale is 0.50.

2.2.5 State/Trait Anxiety Inventory (STAI) – Spielberger et al (1983); Chapters 3, 5 and 6

The STAI is a 40-item, self-rating anxiety measure, with the first 20 items identifying the participants' state anxiety, and the second 20 identifying the participants' trait anxiety. Participants are presented with the 40 statements and must give a score of either 1 ('do not agree at all'), 2 ('agree somewhat'), 3 ('agree moderately') or 4 ('very much agree'). A sample question would be 'I feel calm', or 'I feel upset'. These scores are then summed to give their state and trait anxiety scores. Spielberger et al (1983) report the internal consistency of the state measure to be 0.83, and 0.89 for the trait measure.

2.3. Psychometric Measures

2.3.1 Digit-Span; Chapters 3, 5 and 6

In order to test working memory, participants were asked to listen carefully to a sequence of numbers, and then repeat them back to the researcher. Once the participant had given their answer, the next sequence was given. Sequences were presented in 'levels' depending on their length (3-8), with each level containing two same-length sequences. If a participant failed both sequences at any level, the task stopped. Once this section was completed,

participants performed the same digit-span task, but had to repeat the sequences backwards (length 2-7).

2.3.2 Wechsler Test for Adult Reading (WTAR) – Wechsler (2001) – Chapters 3, 5 and 6

Participants were given a list of 50 words, which they had to, in their own time, read out loud. Each volunteer was instructed to read each word out loud, regardless of whether or not they recognised the word. A point was given for each correct pronunciation, and none for an incorrect pronunciation. From this a verbal IQ score was calculated following conversion to standardised scores.

2.4 Computerised Cognitive Tasks

2.4.1 Pruning Tasks – Huys et al (2012) – Chapters 3, 4, 5 and 6

This task was administered in every experiment throughout the thesis, with two versions of this task being used: in chapters 3 and 6 an un-timed version was administered, and in chapters 4 and 5 a timed version was used. First, the general task will be described as it was used in chapters 3 and 6 and then the modifications made to the task due to time constraints in chapters 5 and 6 will be explained.

2.4.1.1 Pruning Task used in Chapters 3 and 6

First, participants learned how to move around the matrix during 'transition training'. This matrix contained 6 boxes, and participants could move between these boxes by pressing either the U or the I key on a keyboard. From each box it was possible to move to two other boxes, depending on which key was pressed. Participants had a schematic of the transition matrix (figure 2.1, left) in front of them so that they could learn the moves, which they could look at as much as necessary. Neither the order of these transitions, nor the keys that moved from box to box ever changed. Participants were instructed that they were the white box, and had to reach a green target box within a set number of moves (1-4 moves; figure 2.1, right). If they failed on a trial, they simply tried again without incurring a penalty. Once they had reached the end of this training, they were then given a test, whereby they had to reach the green box in a specific number of moves on at least 9 of the next 10 trials on their first

attempt, without the schematic to help them. All subjects completed this test within two attempts.

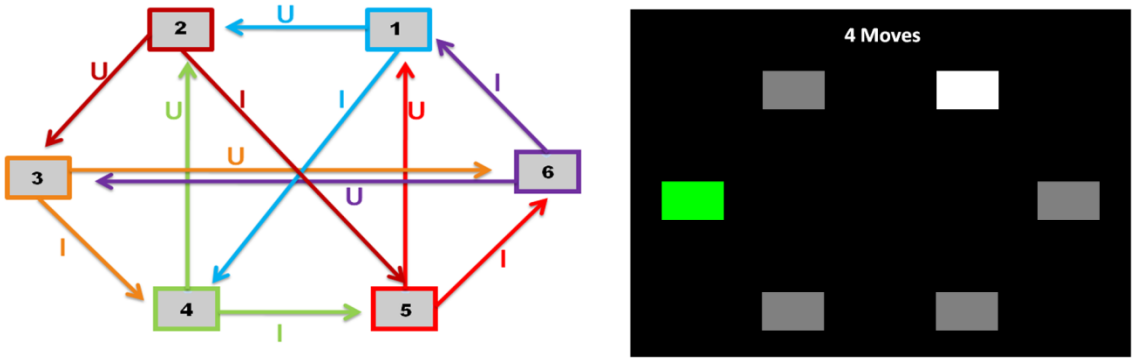


Figure 2.1. Left: Schematic of transition matrix presented to participants in order to aid the learning process. Right: Transition matrix as seen during the training phase – participants had to move the white box into the green box by using every move (precise number specified at the top of the screen on each trial)

Once this training was complete, participants began the task proper. This task began with a short further training phase. This instructed participants that each transition was associated with a deterministic financial outcome (£1.40 or 20p gain £1.40 or 20p loss; Figure 2.2). Participants were not told how much each transition would reward or punish, but instead would have to learn by themselves, by trial and error. Subjects completed 48 trials of varying length (2-8 moves). The first 24 trials were considered part of the reward transition matrix training and were discarded. Thus only the latter 24 trials were analysed. At the beginning of each episode subjects began in a random state, and had to make a sequence of transitions of a certain length in order to maximize financial gain. Relevant '+' or '-' signs were displayed beneath each box throughout the entire task to denote the deterministic rewards and punishments that would occur by *leaving* that box. The relevant deterministic rewards or punishments were displayed in the centre of the screen after each transition was made. On half of the trials, unless instructed otherwise, participants' decisions were displayed after each button press. However, on half of the trials participants were asked to plan ahead the remaining (2-4) moves. This involved subjects planning out their desired sequence, completing it, and only after the final button press was made would the sequence be played

out on the screen (figure 2.2, left). The reward matrix, denoting the financial rewards and punishments, can be seen in figure 2.2, right.

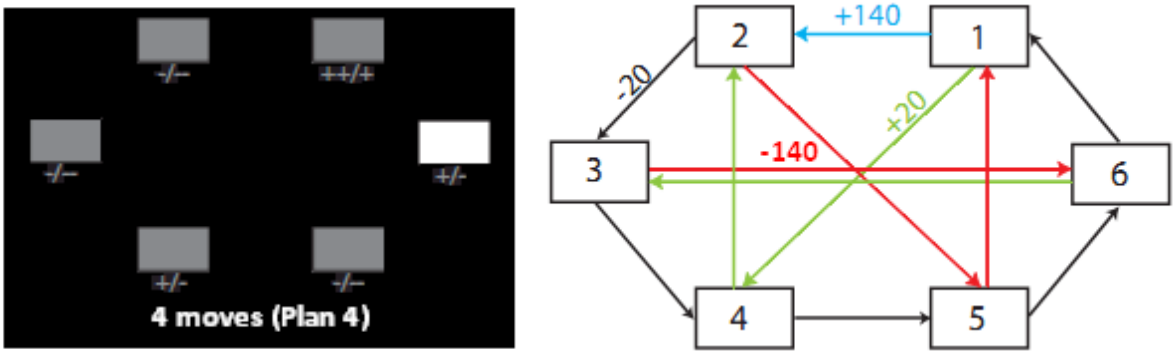


Figure 2.2. Left: Task as seen during performance: on this example participants have to enter 4 moves without immediate feedback, and Right: Reward matrix denoting the financial rewards and punishments for each transition. Participants never actually saw this schematic, but rather had to learn the value of each transition by trial and error

Participants were paid according to their earnings on the task, in order to motivate them to perform accurately, and were also financially compensated for their time. In chapter 3, this task was always administered first in the battery, with the order of the remaining tasks being switched so as to avoid any possible effects of fatigue due to a long testing session.

A set of increasingly complex computational models was fit to the data from this version of the task (chapters 3 and 6) using Bayesian model comparison approach by our collaborator, Dr Quentin Huys (senior research fellow, translational neuromodelling unit, ETZ Zurich and University of Zurich). Each successive model had extra parameters in order to explain the data, and was assessed according to its Bayesian information criterion (BIC_{int}), which is based on the likelihood function (the likelihood that the model can explain the data) but penalizes a model for extra complexity, so as to avoid model overfitting. The first model here was a simple 'look ahead' model that assumed subjects evaluated the entire decision tree. The second included a 'discount' factor parameter (termed γ_G), which represents the probability that participants will choose to continue to evaluate the next step of the sequence at any given point, and can also be termed the 'continuing probability'. The third, termed the 'pruning' model, is central to this study's hypothesis, and splits this 'discount' factor parameter into 'general' (γ_G) and 'specific' pruning parameter (γ_S), with the former describing discounting as

it is described above, and the latter being applied to transitions that immediately followed a large negative result. Both general and specific pruning denote continuing probabilities, and as such a higher score here reflects a lower occurrence of curtailment of the tree search. The final model included a learned Pavlovian component, which accounted for the attraction/repulsion to/from specific states after cumulative exposures. An additional set of parameters, 'rho', was assigned in order to capture sensitivity to each of the four transition types. A loss of a given amount may be more aversive than a win of that same amount is appetitive (loss aversion), and to test whether this was the case here the weights that each subject gave to each of the four possible reinforcements were inferred within this component 'rho'.

2.4.1.2 Pruning Task used in Chapters 4 and 5

A slightly different version of the pruning task was employed in chapters 4 and 5. The main differences were that the large negative transition resulted in a loss of 70 pence rather than 140 pence, the pre-specified sequence lengths were wither 3, 4 or 5 moves long, and participants had to plan their sequence of moves in a 9 second period, and then enter all of their moves in under 2.5 seconds. Furthermore, the experiments within chapters 4 and 5 were carried out using a within-subjects design, meaning that the training was slightly shorter in the second week as participants needed less training.

The initial 'transition matrix' training (including the end of training 'test') of week 1 was still the same as that used in chapters 3 and 6. However, this training phase was removed and replaced with the 'end of training test' in week two. In both weeks there was a second training session which was identical across weeks. This 'reward matrix' training involved participants having to learn that each move in the matrix was worth a win or loss of money. With each move, they could win £1.40 or 20p, or lose 70p or 20p, depending which move they made. Once again volunteers saw pluses or minuses at each box throughout the training and task (see figure 2.2, above), denoting the amount that could be won or lost by leaving that box. During training participants completed 10 of these trials without a time limit, and then completed 10 more which had time constraints. On the latter trials participants had 9 seconds to both look at the matrix and identify a sequence of moves of the length specified (3, 4 or 5 moves), and then 2.5 seconds to enter their moves. If they were too slow here, they would

lose £2. This latter point ensured that participants always attempted to enter moves and enough data was acquired. This training remained the same from week 1 to week 2.

Once the training was complete, participants played the game for real, which was exactly as the time-constrained training had been. The task included 90 trials: 30 of which the optimal sequence of transitions did not include a large punishment (termed non-large loss optimal (NLLO) trials), and the remaining 60 of which the optimal sequence of transitions did include a large punishment (termed large loss optimal (LLO) trials). From this, participants' pruning behaviours were estimated using two variables: the difference between the proportion of trials on each trial type in which participants made the optimal sequence of moves, denoted as the 'difference estimate'; and the proportion of trials of type LLO in which participants did not make the optimal sequence of moves because it contained a large negative, and instead took the next best sequence of moves because it did not contain a large negative, termed the 'proportion best remaining' score. Other variables of interest include the proportion optimal on each trial type, and at each depth, reaction times on each trial type, the number of trials missed (due to being to slow under time constraints) and the amount of money won (participants could win up to £20 at each session as compensation for their time which was added to the compensation for their time). Importantly participants were excluded from any analyses if they obtained a proportion optimal score of less than 40% on the NLLO trials, as this would indicate an inability to perform the task adequately.

Unlike in chapters 3 and 6 (described above) no computational models were applied to the data for the task administered in either chapters 4 or 5 due to the fact that these models were not fully developed and as such could not yet adequately explain the data. Given more time our collaborators would have been able to complete these models, but due to time constraints they could not be applied to the data in chapters 4 or 5.

2.4.2 Choice x Risk (CxR) – Rogers et al (2003), Chapters 3 and 5

Participants completed 80 trials, each of which required them to make a choice between two gambles. They were paid according to their winnings, with each point won being converted to 1 penny. Each gamble was represented as a histogram, the height of which conveyed the probability (0%, 25%, 50%, 75% or 100%) of winning or losing a number of points, the

amount of which was displayed at the top (in green) and bottom (in red) of the histogram, respectively (figure 2.3).

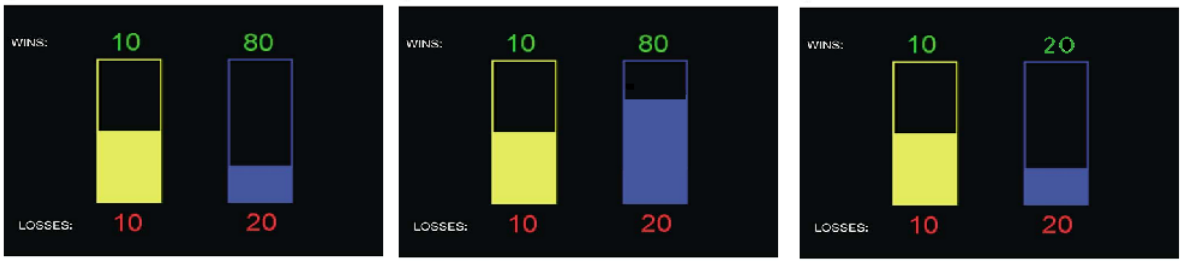


Figure 2.3. Example trials from the CxR task showing the 'control' gamble (left) consisting of a 50% chance of winning or losing 10 points, and the 'experimental' gamble (right). Possible wins are in green, and possible losses are in red

On each trial participants had to choose between the 'control' gamble, which consisted of a 50% chance of winning or losing 10 points, and an 'experimental' gamble, which varied in terms of probability, high (75%) or low (25%) potential gains (80 or 20 points) and potential losses (80 or 20 points), resulting in eight trial types. These two gamble types appeared randomly on either the right or left hand side of the screen. The dependent measure here is the proportion of choices of the 'experimental' gamble over the control gamble. Within these 8 trial types there are three main measures, specifically the proportion of choices of the experimental gamble over the control gamble as a function of 1) probability, 2) the size of expected gains and 3) the size of expected losses. These measures were calculated by measuring the difference between the proportion of experimental alternatives chosen when each of these three values were high, with the proportion of same choices when these values were low.

2.4.3 Temporal Discounting – Pine et al (2009); Chapters 3 and 5

Participants completed 220 trials, making hypothetical choices between options that varied in terms of the amount of money that could be gained and the delay associated with it. In 200 of these trials subjects were presented with 2 scenarios, one in which they would receive a smaller amount of money to be presented in the more immediate future, and one in which they would receive a larger amount of money in the more distant future (figure 2.4). The remaining 20 trials were 'catch' trials, and consisted of one scenario in which subjects could

receive a smaller amount of money presented in the more distant future, and one in which they would receive a larger amount in the more immediate future. These trials were administered to ensure that participants were engaged with the task. The choices ranged in magnitude (from £1 to £100) and delay (from 1 week to 1 year). Participants had to decide which of these scenarios they would rather experience, and were told to press left on a computer keyboard for the choice on the left, and right for the choice on the right.

Figure 2.4. Subjects had to choose between a smaller, more immediate reward (left side of blue bar) and a larger but more delayed reward (right side). The amounts of money differed in magnitude (£1 to £100) and in delay (1 week to 1 year). Subjects completed 220 trials, giving them different scenarios each time

Critical to subjects' choices in this task is the steepness of the discounting of expected reward values according to delay, denoted by V = D * U. Here, V is the subjective value (discounted utility) placed upon an expected, delayed reward; D is a reciprocal function of the delay of the reward, with each participant's 'temporal discount' factor, *k*, ranging from 0 to 1; and U is a negative exponential function of the magnitude of the reward, incorporating a 'diminishing marginal utility' parameter, *r*, that describes the concavity of the subject's utility function. This latter function examines the extent to which participants differ on how much value they place upon an amount (i.e. £1) in a total (i.e. £100), with the assumption that £1 is worth more in a £2 total than it is in a £100 total.. A steep rate of temporal discounting (higher *k*) results in more choices for the smaller, more immediate option being made, while a more shallow rate would result in the opposite, to the point that a 'flat' discounting rate (k=0) would result in no discounting at all, leading subjects to always choose the larger, more delayed reward, no matter how long the wait or how small the increase in magnitude. As such, subjects' choices were assessed in order to reveal the extent of discounting for both

magnitude and delay, and the best fitting parameter estimates for the discount rate (*k*) and utility concavity (*r*) were calculated. It is important to note that this modeling was performed by Dr Alex Pine (Weizmann Institute, Israel).

2.5. Positron Emission Tomography; Chapter 3

PET is a nuclear imaging technique which utilizes a radioactive compound (a molecule of interest bound to a radioactive tracer) that is introduced into the bloodstream in order to determine where it is taken up by the brain.

Subjects within chapter 3 were injected with the radioactive isotope ^{11}Carbon (^{11}C) bound to the molecule CUMI. This radioligand is a competitive agonist that binds preferentially to 5-HT_{1A} receptors, but also provides a better estimate of specific receptor occupancy than other noncompetitive ligands which may themselves have both a lower affinity for these receptors, and a higher affinity for others (Milak et al, 2011). Critically, this ligand was also chosen due to its likelihood of being sensitive to displacement from 5-HT_{1A} receptors by endogenous 5-HT, in an analogous manner to ^{11}C-raclopride's displacement from D2/D3 receptors by dopamine (Montgomery et al, 2003). In other words, since CUMI can be displaced from 5-HT_{1A} receptors by 5-HT itself, it may provide a method of indexing serotonin *release*, in contrast to all other 5-HT receptor ligands, which are only able to asses *binding*.

Subjects participated in two sessions, one of which included being administered 10mg placebo before ^{11}C-CUMI, and the other of which included being administered 10mg an SSRI (citalopram) before the same ligand. The purpose of the placebo condition was to measure baseline 5-HT_{1A} receptor binding values. The purpose of the citalopram condition was to image 5-HT release. In this condition, whilst ^{11}C-CUMI binds to subjects' 5-HT_{1A} receptors, citalopram binds with high affinity to the 5-HTT. The latter point means that this transporter cannot fulfill its normal function of removing excess 5-HT from the synaptic cleft, theoretically resulting in increased 5-HT in the synaptic cleft relative to placebo. Increased 5-HT availability in the synaptic cleft should result in increased binding to 5-HT receptors (including 5-HT_{1A}), and in doing so results in the displacement of ^{11}C-CUMI from the 5-HT_{1A} receptors. The resultant decrease in ^{11}C-CUMI binding signal (relative to placebo) caused by this displacement then provides a measure of serotonin release.

Subjects underwent both conditions with at least 5 days between each (mean inter-scan interval=12.5 days). Participants underwent testing in a randomized double-blind design in which they received a slow 30 minute intravenous infusion of either citalopram (10mg) or placebo before injection of the ^{11}C-CUMI, the latter of which was synthesized as described in Milak et al (2011).

PET scans were acquired from a GE Discovery RX PET/CT scanner with an axial field of view of 15.7cm. 22 frames in total were acquired, each with 47 slices at 3mm thickness. The dynamic PET scans were acquired over 90 minutes. Time frames were of increasing duration: 30 seconds pre-injection background, 1 x 15 seconds, 3 x 5 seconds, 1 x 30 seconds, 4 x 60 seconds, 7 x 300 seconds, and 5 x 600 seconds. The dynamic scans were de-noised using a level 2, order 64 Battle Lemarie wavelet filter (Turkheimer et al, 1999). Head movement in the dynamic PET acquisition was corrected for using frame-by-frame realignment using a mutual information algorithm (Studholme et al, 1997).

The whole-brain parametric images were acquired from the dynamic images using RPM (receptor parametric mapping) software using the SRTM (simplified reference tissue model; Lammerstma and Hume, 1996) method. Each subject's parametric image from one condition was then co-registered to their parametric image from the other condition, and one image was subtracted from the other. The resultant difference (displacement) image was then spatially normalised to the PET template within SPM8 software (statistical parametric mapping 8; www.fil.ion.ac.uk/spm/software/spm8/), as was the image from the placebo condition. All images were then smoothed with an 8mm full-width at half maximum Gaussian kernel. Participants' scores from each of the behavioural tasks (see *2.4.1.1, 2.4.2* and *2.4.3* above) were entered as second level covariates in analyses including 1) their placebo images and 2) their difference image to permit the calculation of correlations between their behaviour and baseline 5-HT$_{1A}$ binding, and between behaviour and the decrease in 5-HT$_{1A}$ binding (corresponding to 5-HT release) due to citalopram infusion, respectively.

Due to the importance of the dorsal raphe nucleus in the 5-HT system, we also conducted a secondary region of interest (ROI) analysis in order to observe any potential correlations between 5-HT$_{1A}$ availability in this region with behaviour. This was done by using signal extracted from the bilateral dorsal raphe nucleus ROI that was manually defined as a fixed

sized region (648 mm^3) on the summed PET images of each individual. These were each entered along with participants' behavioural scores into the Statistical Package for Social Sciences (SPSS 19, SPSS Inc., Chicago, IL) and a Pearson's *r* test of correlation was performed. We adopted a threshold of $P<0.01$ to adjust for the number of correlations conducted.

2.6 Serotonergic Manipulation Techniques

2.6.1 MDMA Administration; Chapter 4

MDMA is a psychoactive drug, leading to symptoms such as euphoria and visual hallucinations that reach a peak at roughly 90-120 minutes, and subside roughly 2.5-3.5 hours later. Administration of MDMA has been shown to acutely (at the time of administration) increase 5-HT levels (Rudnick and Wall, 1992), and subacutely (that is, typically, after mood change has subsided) decrease them (Stone et al, 1986, Kish et al, 2000). It has also been shown both to increase, and subacutely decrease mood (Curran and Travill, 1997). As such, administration of MDMA was used here as a method of 5-HT depletion, by examining participants during the sub-acute period.

Participants underwent administration of 100mg of MDMA or placebo (encapsulated ascorbic acid /vitamin-C) via injection into the antecubital vein, on acute day 1, and then the opposite on acute day 2 (at least one week later) in a within subjects, counterbalanced design. The same participants then returned 3 days later to participate in the behavioural testing session in which they performed the 3 behavioural tasks described in 2.4.1.1, 2.4.2 and 2.4.3 above. No drug was administered on this day, so that the subacute effects of prior MDMA administration upon decision-making could be assessed.

2.6.2 Acute Tryptophan Depletion (ATD); Chapter 5

ATD is an experimental technique used in order to decrease levels of 5-HT's precursor, the large neutral amino (LNAA) acid L-tryptophan (TRP). The synthesis of 5-HT within the brain is dependent on the availability of TRP, with the latter being firstly synthesised into 5-hydroxy-l-tryptophan (5-HTP) by tryptophan hydroxylase (TPH), and then into 5-HT by aromatic-l-amino acid decarboxylase (DDC; figure 2.6). Whilst 5-HT cannot cross into the

brain through the blood-brain barrier (BBB), both TRP and 5-HTP can. This means that ATD can be achieved through administration of the other LNAAs (of which there are 5), excluding TRP, via either capsules, or, as in this thesis, a drink. All LNAAs compete for entry into the brain via the LNAA transporter at the BBB. This transporter is non–LNAA specific, and thus competition exists between the LNAAs for entry. Administering LNAAs without TRP decreases the TRP:LNAA ratio in the blood and cerebrospinal fluid (Crockett et al, 2012), and as such TRP's competition increases, making 'Acute Tryptophan Depletion' something of a misnomer; this method actually increases levels of competing LNAAs, leading to a hypothesised decrease in levels of TRP (and thus 5-HT) in the brain (Crockett et al, 2012).

Due to the fact that until recently, there has been no way in which to image 5-HT release in the living human brain (see Selvaraj et al, 2012), there has been no way to show ATD's ability to decrease such a release. However, this method has been shown to reduce 5-HT levels in rodent brain tissue *in vivo* (Moja et al, 1989), reduces cortical 5-HT release in rats (Stancampiano et al, 1997), and has been shown Nishizawa et al (1997), using PET and the radiotracer α-^{11} C-methyl-tryptophan, to reduce 5-HT synthesis 5 hours after ATD.
As such, this technique was used here in order to manipulate levels of 5-HT and examine the effect of this upon participants decision-making behaviours.

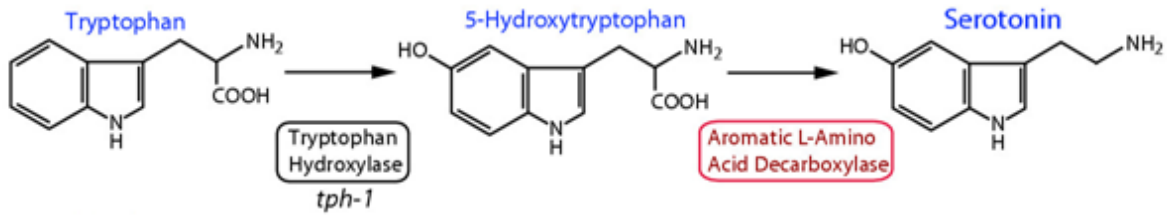


Fig. 2.6. The serotonin synthesis pathway. Tryptophan is converted in 5-Hydroxytrptophan by Tryptophan Hydroxylase, which is itself then converted to Serotonin (5-HT) by Aromatic-L-Amino Acid Decarboxylase

The study in chapter 5 was a double-blind, within subjects design, and as such participant's ingested either a tryptophan-depleted (TRP-) or sham-depletion (TRP+) mixture in week 1, and the converse in week 2 (for constituents, see table 2.1).

TRP -	TRP +	
4.1g	4.1g	L-alanine
2.4g	2.4g	L-glycine
2.4g	2.4g	L-histidine
6g	6g	L-isoleucine
10.1g	10.1g	L-leucine
6.7g	6.7g	L-lysine
4.3g	4.3g	L-phenylalanine
9.2g	9.2g	L-proline
5.2g	5.2g	L-serine
4.9g	4.9g	L-threonine
5.2g	5.2g	L-tyrosine
6.7g	6.7g	L-valine
3.7g	3.7g	L-arginine
2g	2g	L-cysteine
2.3g	2.3g	L-methionine
0g	3g	L-tryptophan
		TOTAL TRP- = 75.2g TRP+ = 78.2g

Table 2.1. Constituents of amino acid mixtures ingested by participants. TRP – indicates mixture without L-tryptophan (intended to deplete volunteer's tryptophan), whilst TRP + indicates mixture with L-tryptophan. These measurements were given to participants regardless of gender

The amino acid mixture was commercially mixed (Nutricia), coded according to a blinding protocol by another member of the laboratory who was otherwise not involved in the study (only unblinded after the final participant completed the study), and added to roughly 568ml (1 pint) of water, with either grapefruit, cherry vanilla, or lemon/lime flavouring to make the drink more palatable. The exact composition of amino acids was chosen based upon Roiser et al (2006) and Roiser et al (2007). Potential side-effects of the drink included nausea, which

two volunteers experienced and thus were withdrawn from the study (having completed only session 1).

Participants also had their blood taken (6ml) before ingestion of the amino acid drink, and 5 hours after ingestion (6ml again). Immediately after venepuncture, blood was centrifuged at 3000rpm for 10 minutes, and then frozen at -80°C. All samples were sent to the Department of Biology and Biomedical Sciences at Oxford Brookes University for amino acid analysis for both levels of total tryptophan, and the ratio of large neutral amino acids to tryptophan. This analysis was performed by Dr Michael Franklin of Faculty of Life Sciences, Oxford Brookes University.

3) DECISION-MAKING AND THE 5-HT$_{1A}$ RECEPTOR: A POSITRON EMISSON TOMOGRAPHY STUDY

3.1 Introduction

3.1.1 The 5-HT$_{1A}$ receptor and decision-making

The 5-HT$_{1A}$ receptor is an inhibitory G-coupled protein receptor found on the axon, soma and dendrites of serotonergic neurons. 5-HT$_{1A}$ receptors exist within the dorsal raphe nucleus (the origin of the brain's 5-HT system) as autoreceptors, where they respond to 5-HT released by the neurons in whose membranes they are embedded, and in projection sites throughout the rest of the cortex as heteroreceptors, where they respond to 5-HT released by adjacent neurons. These 5-HT$_{1A}$ receptors are the most widespread of all 5-HT receptors, and are found in high numbers within the raphe nuclei, hippocampus, amygdala, basal ganglia and thalamus, as well as throughout the cortex (Hannon and Hoyer, 2008). In the raphe, 5-HT$_{1A}$ autoreceptors play a critical role in 5-HT transmission, dampening down the firing of 5-HT neurons via an inhibitory feedback loop (Blier et al, 1998).

As discussed in the introduction, 5-HT is thought to influence numerous cognitive processes, in particular decision-making (e.g. Dayan and Huys, 2008, Seymour et al, 2012, Crocket et al, 2012). The 5-HT$_{1A}$ receptor may play an important role in this influence, with animal studies having also provided support for this: Miyazaki et al (2012) demonstrated that selective injection of the 5-HT$_{1A}$ receptor agonist 8-OH-DPAT into the dorsal raphe, which decreases 5-HT neuron firing rates, increased rats' number of waiting errors for delayed rewards but not immediate rewards on a temporal discounting paradigm. Similarly Carli and Samanin (2000) reported that administration of 8-OHDPAT into the raphe both depleted forebrain 5-HT by 90% and increased rats' discounting of future rewards based upon their temporal delay. Conversely, Cervantes et al (2009) reported that impulsive choice behaviours in adult male hamsters were reduced following systemic 5-HT$_{1A}$ agonist administration. The results of Carli and Samanin (2000) and Cervantes et al (2009) are thus complimentary due to the fact that the administration of 8-OH-DPAT into the raphe in the former study *decreases* transmission at the 5-HT$_{1A}$ autoreceptors in this region, whilst systemic agonist administration in the latter *increases* transmission at the 5-HT$_{1A}$ heteroreceptors throughout

the cortex. However, Liu et al (2004) reported that rats systemically injected with the 5-HT_{1A} receptor agonist buspirone at different doses (0.5, 1 and 2mg/kg) showed different effects upon temporal discounting depending on the dosing schedule, with acute buspirone dose-dependently increasing discounting, but chronic administration (over a 65 day period) reversing this pattern (decreasing discounting compared to baseline) at a dose of 1mg/kg. Further, these effects of buspirone were reversed by administration of the 5-HT_{1A} receptor antagonist WAY-100635.

Further, human studies of genetic polymorphisms have also provided support for an influence of 5-HT_{1A} receptors upon decision-making: Schmitz et al (2009) showed that participants who were homozygous for the 5-HT_{1A} C(-1019) G polymorphism, which is linked to increased expression of 5-HT_{1A} receptors (Czesak et al, 2012), exhibited both increased reaction times to potential rewards and decreased reaction times to potential punishments. Further, Gu et al (2013) report that schizophrenia patients with rs6295 polymorphisms in the HTR1A gene (which encodes the 5-HT_{1A} receptor) performed poorly on the 'ambiguity' trials of the IGT compared to controls, and Benko et al (2010) showed that healthy volunteers who were homozygous for the same polymorphism displayed significantly higher impulsiveness on both the impulsive subscale of the Eysenck impulsiveness, venturesomeness and empathy scale and the Barratt impulsiveness scale compared to heterozygotes and those without the polymorphism. Finally, Chamberlain et al (2007) reported no effects of administration of either 20mg or 30mg of buspirone upon impulsive responding in healthy human volunteers. The results of this latter study are in direct contradiction to those of Liu et al (2004) above, yet it must be noted that the task used in Chamberlain et al was the stop-signal task from the CANTAB rather than a test of temporal discounting. As such, whilst some studies into the effect of the 5-HT_{1A} receptor in decision-making and impulsivity using human participants have been performed, this area remains largely unexplored.

3.1.2 The role of the striatum and hippocampus' in decision making

Many brain regions innervated by 5-HT have been implicated in decision making, in particular the striatum, which has long been thought to play a central role in reward processing (e.g. Delgado et al, 2000, Kable and Glimcher, 2007, Pine et al, 2009; see Robbins and Everitt, 1996 and Everitt et al, 1999 for a review of the striatum's role in the processing

of rewards, and Cardinal et al, 2004 for a review of the role of the striatum in delayed reinforcements and temporal discounting).

Other studies highlight a specific role for 5-HT in the striatum in both reward and punishment processing. For example, Seymour et al (2012) used a decision-making task that permitted examination of the effects of rewards and punishments separately to show that pharmacologically decreasing levels of 5-HT through ATD altered the exchange rate by which rewards and punishments were compared, leading to an attenuation of the subjective representation of reward value, linked to an increase in haemodynamic responses in parts of the striatum and prefrontal cortex. Further, McCabe et al (2010) have highlighted the effect of SSRIs upon the neural processing of rewards and punishments within the striatum. Here, the authors administered citalopram for 7 days, and discovered that it reduced activation to appetitive stimuli (chocolate) in the ventral striatum whilst reboxetine (a selective norepinephrine uptake inhibitor) had no such effect. Further, Abler et al (2012) report that administration of the SSRI paroxetine bilaterally decreased activation within the nucleus accumbens during the processing of rewards (erotic videos). Finally, Tanaka et al (2007) administered ATD to participants who were performing a temporal discounting paradigm, and discovered that participants displayed activity within the ventral striatum that correlated with reward prediction at shorter time scales, which was stronger at low levels of 5-HT, and displayed activity in the dorsal striatum during the prediction of rewards at longer time scales, which was stronger at higher levels of 5-HT.

The hippocampus has also been implicated in decision making due to its role in contextual punishment processing in experimental animals. For example, Fanselow and Dong (2010) showed that rats with hippocampal lesions have a deficit in fear processing in a tone-shock association paradigm, but only when the context changed. It has also been argued that hippocampal place cells provide spatial evaluation functions that are involved in the planning and representation of location with goal proximity (e.g. Viard et al, 2011) in order to provide a basis for appropriate choice selection. Importantly, in humans Camara et al (2008) showed increased coupling between the ventral striatum and regions of the amygdala and hippocampus during the processing of gains and losses. Work on human participants has argued that the hippocampus and parahippocampal gyrus play a crucial role in the formation of past, present and future episodic representations (Schacter and Addis, 2009); and Peters and Buchel (2010) suggest that decision-making within a temporal discounting paradigm

depends on the subjective capacity for future episodic thought, with this 'mental time travel' (self-projection into the future) involving the hippocampus.

Research is beginning to highlight the involvement of 5-HT in the relationship between the hippocampus and decision-making. Acute tryptophan depletion (ATD) is associated with lower performance on hippocampal-dependent episodic memory tasks (Riedel et al, 1999), which is argued to be due to a resultant decreased activation of excitatory 5-HT receptors (Meeter et al, 2006). Further, 5-HT has also been linked with the hippocampus' potential role in temporal discounting: Mobini et al (2000) reported that rats whose 5-HT systems had been destroyed with 5,7-dihydroxytryptamine became more impulsive and exhibited increased choosing of smaller, sooner rewards in a temporal discounting paradigm, which correlated with a decrease of 5-HT in the hippocampus. However, little work has attempted to link 5-HT transmission in the hippocampus to decision-making in humans.

3.1.3 Present study and predictions

In order to assess the hypothesis that transmission at the 5-HT_{1A} receptor plays an important role in decision making, we measured individual differences in regional binding of this receptor by using PET and correlated this with participants' performance on three decision-making tasks at a separate testing session.

Initially, Selvaraj et al (2012) administered ^{11}C-CUMI-101, a partial agonist of the 5-HT_{1A} receptor, before either a placebo, or citalopram. The former allowed the researchers to observe baseline 5-HT_{1A} receptor binding, whilst the latter allowed the assessment of 5-HT release (see experimental methods chapter 2.6.1). For this first section of the study, it was predicted that citalopram infusion would lead to increased 5-HT within the brain and thus decreased binding of CUMI to the 5-HT_{1A} receptors, due to increased binding of endogenous 5-HT to these receptors throughout the cortex.

On a separate testing session, we administered the pruning task described in 2.4.1, the gambling task described in 2.4.4 and the temporal discounting paradigm presented in 2.4.5. Participants scores were then entered as second-level covariates in an SPM analysis in order to observe any correlation between performance on these tasks and both baseline 5-HT_{1A} availability, and the change in such availability due to citalopram infusion.

It is important to note that PET scans (to observe both the baseline and the change in 5-HT_{1A} receptor availability due to citalopram infusion) were performed by Selvaraj et al (2012), whilst the separate behavioural session, in which the 3 cognitive tasks were administered, was carried out by me, as was the subsequent statistical analysis in which participants's scores were correlated with their 5-HT_{1A} availability.

With regards to participants' baseline PET scans, it was predicted (based on theory and previous findings) that participants with low levels of 5-HT_{1A} (heteroreceptor) availability in projection sites would display decreased pruning (based on Dayan and Huys, 2008 and Huys et al, 2012), decreased sensitivity to rewards and probabilities on the gambling task (as elicited with acute tryptophan depletion: Rogers et al, 2003 and Rogers et al, 1999), and increased discounting (as elicited with acute tryptophan depletion: Schweighofer et al, 2008). It was also predicted that participants with low levels of 5-HT_{1A} (autoreceptor) availability within the dorsal raphe nucleus would have attenuated inhibition of 5-HT firing within the raphe, and therefore increased release in projection sites, and thus higher levels of pruning, greater sensitivity to rewards and probabilities on the gambling task, and decreased discounting.

With regards to participants' citalopram-induced change in 5-HT_{1A} receptor availability within projection sites, it was predicted that participants with low levels of 5-HT release would display low levels of pruning, poorer sensitivity to rewards and probabilities, and increased discounting.

3.2 Methods

3.2.1 Participants

Fifteen participants (13 males, mean age 50.9 years, range 35-63 years) were recruited via the website www.gumtree.com. Of these 15 participants, all underwent a PET scan in the placebo condition, and 13 underwent a scan in the citalopram condition. Scanning took place at the Cyclotron Unit at the Hammersmith Hospital campus of Imperial College London. All participants completed the behavioural session at the Institute of Cognitive Neuroscience, University College London. Participants were free of any psychiatric disorders, as determined by assessment on the MINI, HAM-D, BDI and STAI at the behavioural session. None of the participants had taken any psychotropic medication in the 12 months prior to participating, nor had they any previous history of alcohol/substance dependence. Informed written consent was obtained from all participants at both scan and behavioural sessions, and ethical approval was obtained from the Hammersmith, Queen Charlotte's and Chelsea Hospitals Research Ethics committee for the PET scan and from the UCL ethics committee for the behavioural testing.

3.2.2 Procedure

Participants arrived for their PET sessions at the Hammersmith Hospital at 9am, with scanning commencing at 11am for all participants at all sessions. Participants were scanned with at least 5 days between their sessions (mean inter-scan interval = 12.5 days). Participants were injected with either 10mg of placebo (saline) or citalopram 45 minutes before injection of the radioligand. This dose of SSRI was chosen as it has been shown to produce robust and lasting release of anterior pituitary hormones, which is considered to be a marker of activation of 5-HT pathways (Hinz et al, 2008 and Attenburrow et al, 2001). For details of the PET analysis methods, refer to chapter 2, section 2.5. Participants were compensated £50 for their time and effort during these scans. Details of the scanning procedure can be seen in Selvaraj et al (2012).

Participants arrived for their behavioural sessions (on a separate day to the PET session) at either 9am, or 1pm, depending on allocation. They performed the three tasks, in a counter-balanced, randomized order. The pruning task, however, was always administered first.

Participants were compensated for their time and effort in this portion of the study. This included receiving £20 to compensate for travel and time, and up to £10 on 2 of the 3 cognitive tasks, depending on performance, meaning each volunteer was compensated between £20 and £50 for this session.

3.2.3 Statistical Analyses

Our primary analyses were conducted using the whole-brain parametric PET images presented in Selvaraj et al (2012), using SPM8 (http://www.fil.ion.ucl.ac.uk/spm/software/spm8/) using MATLAB software (http://www.mathworks.co.uk/products/matlab/). Participants' scores from each of the four behavioural tasks (pruning task, choice x risk, temporal discounting and the Pavlovian to Instrumental-Transfer (PIT) – see experimental methods, chapter 2), were entered as second level covariates in analyses involving both their placebo PET images, and their difference images (calculated as the difference between binding in the placebo and citalopram conditions), in order to identify correlations between performance on the tasks and both their baseline 5-HT_{1A} availability, and the change in 5-HT_{1A} binding due to citalopram infusion, respectively.

We corrected for multiple comparisons, controlling the family-wise error rate. We were particularly interested in relationships between 5-HT_{1A} availability and behaviour in the striatum and hippocampus, and as such created specific a striatal mask (containing the nucleus accumbens, putamen and pallidum only, due to very low binding values within the caudate) and a hippocampal mask (including the parahippocampal gyrus to make a hippocampal complex mask). These masks were defined using the HamNET Atlas, and applied to constrain the search volume to adjust the correction for multiple comparisons.

Due to the importance of the dorsal raphe nucleus in the 5-HT system we also conducted a secondary analysis in order to observe correlations between 5-HT_{1A} availability in this region and participants' behaviour. This was done by using signal extracted from a bilateral dorsal raphe nucleus ROI that was manually defined as a fixed sized region (648 mm^3) on the summed PET images of each individual. These were each entered along with participants' behavioural scores into the Statistical Package for Social Sciences (SPSS 19, SPSS Inc., Chicago, IL) and a Pearson's *r* test of correlation was performed.

3.3 Results

3.3.1 5-HT$_{1A}$ receptor binding

Table 3.1 and Figure 3.1 summarize the regions of citalopram-induced change in 5-HT$_{1A}$ receptor binding as shown by the whole-brain analysis. The direction of this change in 5-HT$_{1A}$ receptor binding was surprising due to the fact that CUMI-binding was *increased*, implying decreased 5-HT release due to citalopram infusion, which was opposite to the direction predicted. The regions displaying change are consistent with those from the region of interest analyses reported in Selvaraj et al (2012), especially with respect to the frontal and temporal lobes, hippocampus bilaterally and the cingulate gyrus. Importantly, no effect was detected in any region at $P < .001$, uncorrected, for the opposite contrast (i.e. in the direction to that was predicted). The raphe nucleus (midbrain) did show a non-significant ($P>.05$, uncorrected) numerical decrease in CUMI binding however (dorsal raphe CUMI-binding mean (SD) in the placebo condition = 1.66 (0.28), and mean (SD) in the citalopram condition = 1.61 (0.30), t[12] = 0.57, $P = 0.58$).

Region	**Cluster size**	***Z* score**	**X**	**Y**	**Z**
Angular gyrus (L)	347 *	5.15 *	-26	-66	34
Lateral Orbital gyrus (L)	509 *	5.02	-28	40	-18
Isthmus cingulate gyrus (R)	326 *	4.99	8	-52	14
Medial frontal gyrus (R)	5187 *	4.97	48	26	32
Putamen (R)	221 *	4.44	20	8	16
Medial frontal gyrus (R)	50	4.4	18	10	42
Superior frontal gyrus (L)	832 *	4.36	-20	30	34
Cingulate gyrus (R)	308 *	4.33	12	-42	30
Precentral gyrus (R)	434 *	4.25	54	-10	44
Hippocampus (R)	517 *	4.19	18	-12	8
Postcentral gyrus (R)	33	4.17	36	-44	74
Putamen (L)	291 *	4.16	-20	14	2
Superior temporal gyrus R	422 *	4.12	72	-18	-8
Superior frontal gyrus (L)	71	3.9	-14	-8	64
Internal capsule (L)	61	3.89	-20	-24	-4

Supramarginal gyrus (L)	66	3.87	-40	-46	28
Hippocampal commissure (L)	142	3.84	-6	-36	20
Inferior temporal gyrus (R)	162	3.82	52	6	-48
Entorhinal cortex (R)	86	3.76	8	-8	-44
Medial temporal gyrus (R)	33	3.7	56	-32	-26
Entorhinal cortex (L)	155	3.68	-10	8	-26
Inferior temporal gyrus (L)	21	3.68	-44	-46	-12
Occipital gyri (L)	29	3.66	-22	-82	-14
Medial frontal gyrus (R)	29	3.61	34	-10	44
Medial frontal gyrus (R)	49	3.57	44	-10	62
Supramarginal gyrus (R)	29	3.57	28	-56	28
Medial temporal gyrus (R)	54	3.52	66	-32	18
Postcentral gyrus (R)	22	3.51	52	-22	48
Superior frontal gyrus(R)	20	3.47	10	-10	80
Superior frontal gyrus (L)	69	3.42	-18	14	48
Medial temporal gyrus (R)	21	3.33	42	-66	22

Table 3.1. Table summarizing the whole-brain analysis denoting regions of change in 5-HT_{1A} receptor binding from the contrast citalopram-placebo, the size of clusters in voxels and the peak co-ordinates in MNI space. Asterisks indicate where an effect survives whole-brain correction for multiple comparisons at the cluster level (Cluster size column) or voxel level (*Z* score column). Images were initially thresholded at *P* < .001, uncorrected, minimum cluster size 20

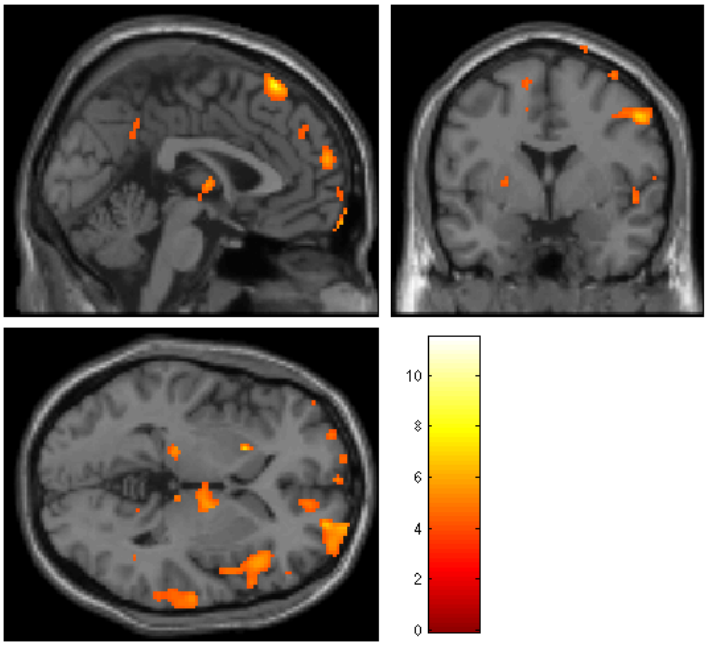


Figure 3.1. Statistical Parametric Map indicating citalopram-induced change in 5-HT$_{1A}$ receptor binding within brain regions (citalopram condition – placebo condition contrast, thus indicating an increase in CUMI binding). Whole-brain analysis presented at *p* < .001, uncorrected. Colour bars denote t-values

3.3.2 Relationship between 5-HT$_{1A}$ receptor binding and behaviour

Scores on each of the task variables were added as 2nd level covariates into SPM in order to identify correlations between both baseline 5-HT$_{1A}$ receptor availability, and the change in such availability due to citalopram infusion, and performance on the tasks. Results of these 2nd level analyses can be seen in table 3.2 below.

Task (Variable)	Condition	Direction	Region	Cluster size	*Z value*	X	Y	Z
Pruning (Specific)	**Change**	**Positive**	Inferior Temporal Gyrus (L)	10	3.35	-40	-64	-4
			Medial Temporal Gyrus (R)	12	3.28	54	-76	6
CxR (Win)	Baseline	Positive	Occipital Gyrus (R)	22	3.52	14	-58	16
		Negative	Medial Temporal Gyrus (L)	11	3.91	2	-28	-4
CxR (Loss)	Baseline	Positive	Inferior Temporal Gyrus (R)	15	3.41	42	-62	0
			Inferior Temporal Gyrus (L)	32	3.34	-54	-32	-36
		Negative	Inferior Temporal Gyrus (L)	29	3.92	-10	-50	36

CxR (Loss)	Change	Positive	Inferior Temporal Gyrus (L)	19	4.08	-44	-12	-32
			Medial Temporal Gyrus (L)	56	4.06	-30	26	58
			Parietal Operculum (R)	29	4.01	44	-36	26
			Nucleus Accumbens (R)	23	3.88 *	10	18	0
			Medial Frontal Gyrus (R)	20	3.55	46	20	56
			Inferior Temporal Gyrus (R)	12	3.47	42	-46	-12
			Piriform cortex (R)	23	3.36	26	0	-10
CxR (Prob.)	Baseline	Positive	Perirhinal Cortex (L)	55	4.06	-10	4	-36
			Perirhinal Cortex (R)	292	3.87 *	18	6	-38
CxR (Prob.)	Change	Positive	Brainstem	25	4.15	6	-16	-40
Temp. Disc. (*K*)	Baseline	Negative	Parahippocampal (L)	411	4.24 *	-26	-14	-42
			Paracentral Lobule (L)	23	4.00	-16	-42	46
			Striate Area (L)	49	3.54	-8	-88	8
			Inf. Temporal Gyrus (R)	31	3.29	46	-44	-34
			Occipital Gyrus (R)	15	3.26	26	-88	-6
			Fusiform Gyrus (R)	16	3.21	40	-18	-46
Temp. Disc. (*K*)	Change	Positive	Sub thalamic nucleus (R)	56	4.10	6	-16	-12
			Inferior frontal gyrus (R)	28	3.83	56	26	-16
			Parahippocampal gyrus (L)	13	3.40	-12	-30	-20
Temp. Disc. (*r*)	Baseline	Positive	Lateral Sup.Frontal Gyrus (L)	24	3.71	-34	40	50
Temp. Disc. (*r*)	Change	Positive	Medial frontal gyrus (R)	27	3.65	40	38	26
			Superior Frontal Gyrus (L)	32	3.61	-12	32	40

Table 3.2. Results of the covariate analyses. All images were initially thresholded at *P*<.001, uncorrected. Asterisks denote correlations that survived small volume correction for multiple comparisons

3.3.2.1 Pruning

3.3.2.1.1 Behavioural Analysis

Eight models were applied to the data in order to explain participants' choices, which are explained in 2.4.1.1. Figures 3.2 and 3.3 show the results of these models, each of which have been compared using the BIC method. Each of the four models with the 'rho' parameter (that examines participants' sensitivity to each of the four transition types) was increasingly better at explaining the data, even after being penalized for its added complexity. Furthermore,

when this parameter was added to each model, it significantly increased each model's ability to explain participant's choices. Figures 3.2 and 3.3 show that the model including the separate pruning parameters (general and specific pruning) along with the Pavlovian and loss aversion components ((rho) Prun. & Pav.) was the most parsimonious, supporting the presence of pruning in this sample.

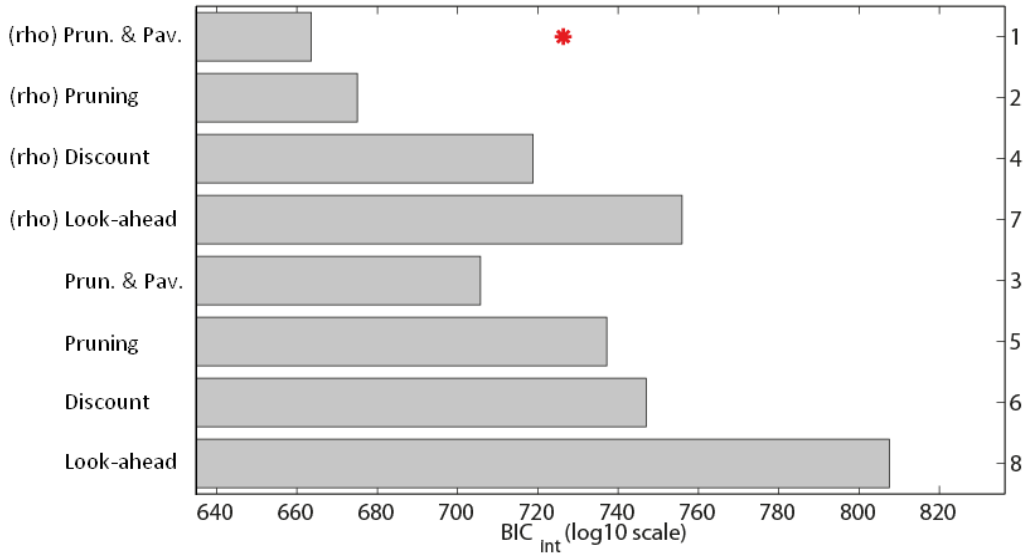


Figure 3.2. Results of BIC model comparison. The top four models are presented in descending order of complexity, as are the bottom four (bottom four without rho parameter). Model pruning and Pavlovian with the rho parameter provided the most parsimonious model. There is decisive evidence in favour of the most complex model (log10 BIC difference>10)

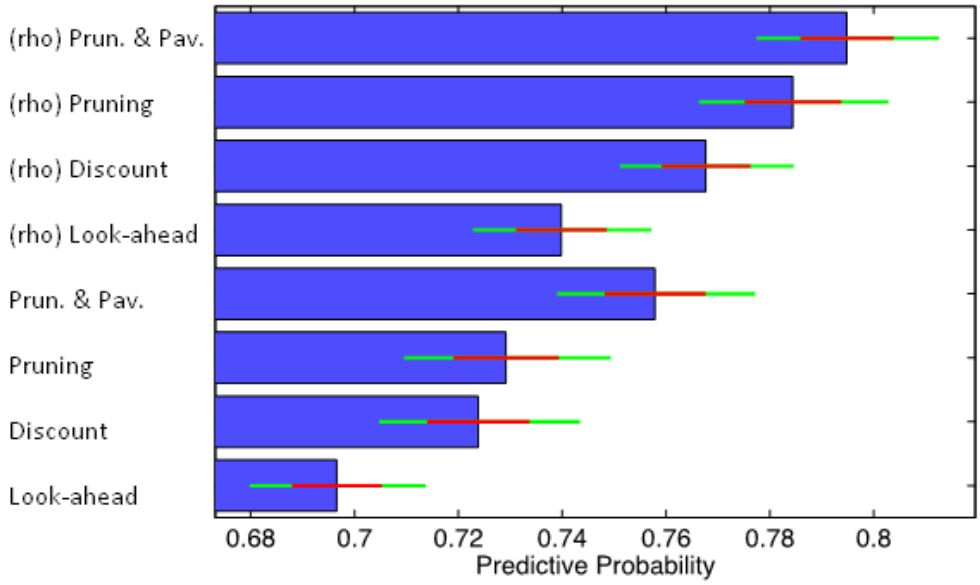


Figure 3.3. Predictive probabilities for each of the 4 models. This table shows that the most parsimonious model from table 3 (rho Prun. & Pav.) was best at predicting participants' choices

3.3.2.1.2 Correlation of specific pruning scores with PET data

From the SPM voxel-wise analyses, no relationship between the specific pruning parameter (γ_S) and baseline (placebo) 5-HT_{1A} receptor availability was detected, even at a threshold of $P < .001$, uncorrected. However, the specific pruning vs citalopram-induced change in 5-HT_{1A} availability contrast revealed positive correlations, though these did not survive correction for multiple comparisons. Table 3.2 summarizes the regions evident in this analysis.

The small volume correction applied to the data revealed no suprathreshold clusters.

3.3.2.2 Choice x Risk

3.3.2.2.1 Behavioural analysis

Participants chose the 'experimental' gamble significantly more often when its probability of winning was higher ($t[14] = 15.0$, $p < 0.001$), when the amount they could win was higher ($t[14] = 2.9$, $p = .011$) and when the amount they could lose was lower ($t[14] = 3.0$, $p = .009$).

The mean sensitivity to wins was 0.110 (SD 0.147), the mean sensitivity to losses 0.115 (SD 0.146) and mean sensitivity to probability was 0.694 (SD 0.180).

3.3.2.2.2 Correlation of sensitivity to wins, losses and probabilities with PET data

No correlations between sensitivity to win and 5-HT_{1A} availability survived correction for multiple comparisons (table 3.2).

The correlation of loss sensitivity with the change in 5-HT_{1A} availability due to citalopram infusion produced positive correlations detected at a threshold of $P < .001$ (table 3.2). The small volume correction that was applied to the data revealed two positive correlations from the contrast of sensitivity to loss with the change in 5-HT_{1A} binding due to citalopram infusion, although only the former survived correction for multiple comparisons; right nucleus accumbens ([x = 10, y = 18, z = 0], cluster size = 19, Z = 3.88, P_{SVC} = .038; figure 3.2), and the right putamen ([x = 26, y = 0, z = -8]), cluster size 3, Z = 3.30, P_{SVC} = .183). This indicates that greater sensitivity to information pertaining to losses was linked with increased 5-HT_{1A} availability (which indicates decreased citalopram-induced 5-HT release) in the right nucleus accumbens following citalopram infusion (figure 3.2).

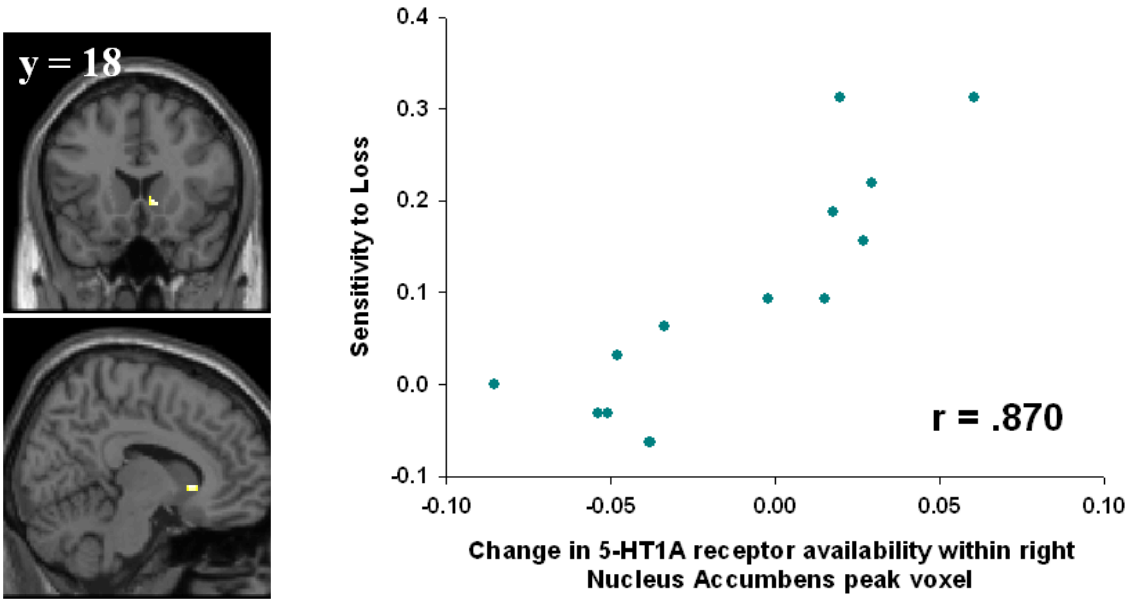


Figure 3.2. Left: SPM image depicting the change in ^{11}C-CUMI binding within the right nucleus accumbens ROI that positively correlates with participants' sensitivity to loss in the change

condition (10, 18, 0, small volume correction analysis presented at P_{SVC} = .038). Right: Scatterplot of correlation. This correlation shows that those participants who were more sensitive to information pertaining to loss also displayed greater increase in 5-HT_{1A} receptor availability (indicative of decreased 5-HT release) within the right nucleus accumbens due to citalopram infusion

The correlation of probability sensitivity with the baseline 5-HT_{1A} availability produced only positive correlations; both in the perirhinal cortex detected at a threshold of $P < .001$ (table 3.2).

The small volume correction that was applied to the data revealed correlations between baseline 5-HT_{1A} availability in these regions bilaterally, though only the correlation in the right survived correction for multiple comparisons; (right; [x = 20, y = 0, z = -38], cluster size = 45, Z = 3.63, P_{SVC} = .028, figure 3.3, left). This indicated that greater sensitivity to information pertaining to the probability of winning was linked with higher baseline 5-HT_{1A} receptor availability.

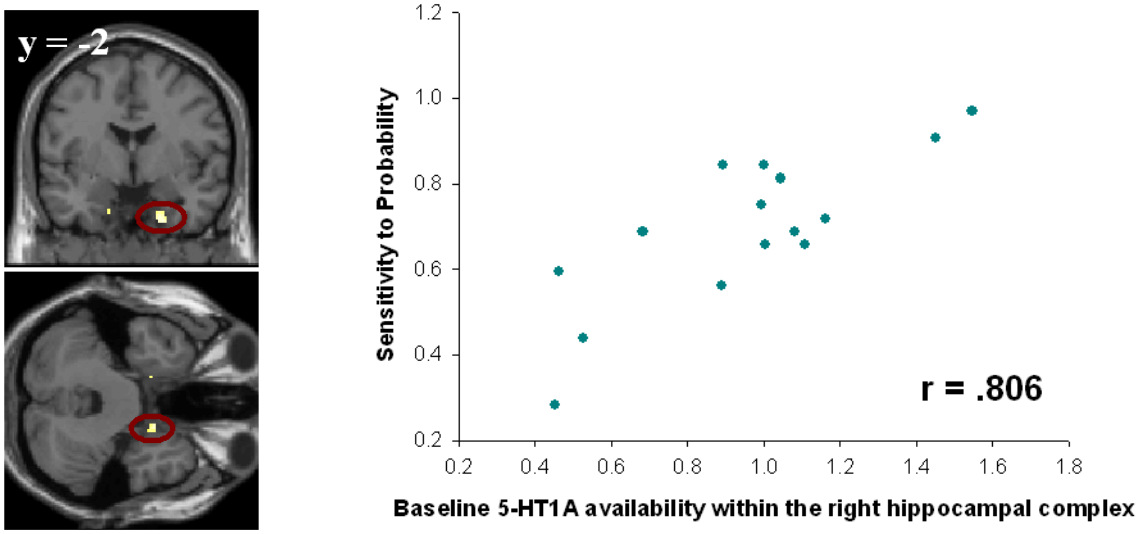


Figure 3.3. Left: Statistical Parametric Map (SPM) depicting ^{11}C-CUMI binding at baseline within the right hippocampal/parahippocampal ROI that positively correlates with participants' sensitivity to probability (20, 0, -38, small volume correction analysis presented at P_{SVC} = .028). Right: Scatterplot of correlation between participants' sensitivity to probability and baseline ^{11}C-CUMI binding within this cluster. This correlation shows that those participants who showed

greater sensitivity to information pertaining to probability had greater baseline 5-HT_{1A} binding within the right hippocampal complex

The correlation of sensitivity to probability with the change in 5-HT_{1A} availability due to citalopram infusion produced only a positive correlation detected at a threshold of $P < .001$ (table 3.2).

3.3.2.3 Temporal Discounting

3.3.2.3.1 Behavioural Analysis

All participants were found to be concentrating sufficiently on the task as they all chose the larger-sooner reward on above 90% of the 20 catch trials (mean 19.4, SD 0.83). On average participants chose the sooner, smaller option over the later option (mean 119.2, SD 48.05) times (out of 200).

Using the model of best fit from Pine et al (2009) it was shown that participants discounted the value of future rewards (mean $k = 0.099$, SD = 0.057) and also exhibited a concave utility function (mean $r = 0.0086$, SD = 0.017), comparable to results reported previously on this task (Pine et al, 2009).

3.3.2.3.2 Correlation of k values (discount factor) with PET data

The correlation of k values (discount factor) with baseline 5-HT_{1A} availability produced negative correlations detected at a threshold of $P < .001$, (table 3.2). The small volume correction applied to the data revealed a significant negative correlation that survived correction for multiple comparisons in the left parahippocampal gyrus ([x = -26, y = -14, z = -36], cluster size 14, Z = 3.54, $P_{SVC} = .037$; figure 3.4). This indicates that a lower discounting (i.e. an increased choosing of the larger, later rewards) was linked to higher baseline 5-HT_{1A} receptor availability.

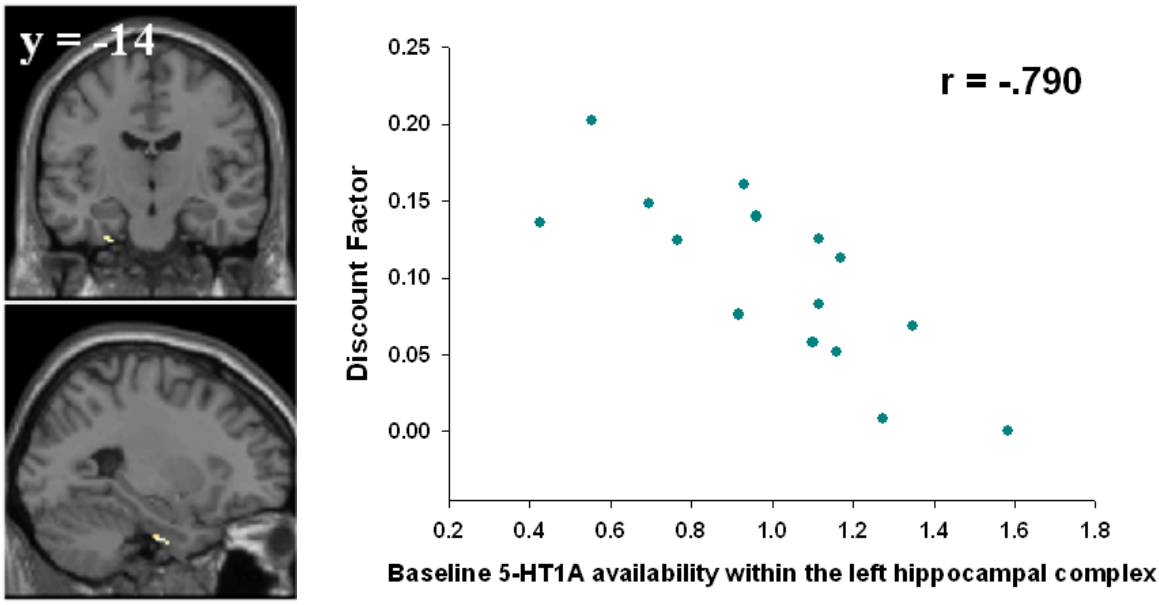


Figure 3.4. Left: SPM image depicting baseline ^{11}C-CUMI binding within the left hippocampal/parahippocampal ROI that negatively correlates with participants' discount factor (-26, -14, -36, small volume correction analysis presented at P_{SVC} = 0.037). Right: Scatterplot of correlation between discount factor and baseline 5-HT_{1A} receptor availability within this cluster. This correlation indicates that those participants who displayed increased discounting of rewards based upon their temporal delay had decreased baseline 5-HT_{1A} receptor binding within the left hippocampal complex

The correlation of k values with the change in 5-HT_{1A} availability due to citalopram infusion produced only positive correlations detected at a threshold of $P < .001$ (table 3.2). The small volume correction applied to the data revealed a positive correlation in the left putamen, however this did not survive correction for multiple comparisons ([x = -20, y = 14, z = -6], cluster size 8, Z = 3.30, P_{SVC} = .175).

3.3.2.3.3 Correlation of r values (utility concavity)

No correlations between participants' utility concavity and 5-HT_{1A} availability survived correction for multiple comparisons (see table 3.2).

3.3.3 Relationship between 5-HT$_{1A}$ receptor binding in the dorsal raphe nucleus and behaviour

Due to the importance of the dorsal raphe nucleus in the 5-HT system, a secondary analysis was also conducted, in which signal was extracted from a manually defined fixed size region (648 mm^3) on the summed PET image of each participant, and correlated (using a Pearson's *r* test of correlation) with variables from each of the above behavioural tasks. No significant correlations were observed (all $P > .2$).

3.4 Discussion

3.4.1 Citalopram challenge

The results of the citalopram challenge upon 5-HT_{1A} availability were surprising. It was originally hypothesized that, relative to placebo, citalopram infusion would lead to increased synaptic 5-HT (due to reuptake blockade) and thus decreased CUMI binding (due to displacement) in all brain regions. However, the inverse was found, with CUMI-5-HT_{1A} receptor binding numerically (but not significantly) decreasing within the raphe, and increasing significantly in projection sites.

One explanation for these unexpected effects relates to effects mediated at 5-HT_{1A} autoreceptors: activation of the 5-HT_{1A} receptors in the dorsal raphe has been demonstrated to inhibit both the firing of serotonin neurons and 5-HT release within the prefrontal cortex (Stockmeier et al, 1998). If citalopram increased the extracellular availability of 5-HT, and thus the binding to the 5-HT_{1A} autoreceptors in the raphe, this would then lead to decreased 5-HT firing in this region, and thus increased CUMI binding to the 5-HT_{1A} heteroreceptors in the projection sites (all areas other than the raphe), due to decreased 5-HT release in those regions. However, whilst Giovacchini et al (2005) were able to demonstrate similar results to these by using an intermediate-affinity antagonist [(18)F]FPWAY in anaesthetized monkeys, this explanation (Blier et al, 1998) remains speculative, and it will be necessary to confirm CUMI's sensitivity to 5-HT displacement using other 5-HT manipulations such as acute tryptophan depletion (which reduces levels of 5-HT) or methylenedioxymethamphetamine (MDMA, which releases 5-HT).

This theory has been proposed to explain why depressed patients who are administered SSRIs initially report no change, or even a decrease in mood, which subsides within roughly 1-2 weeks, by which time an improvement in mood starts to occur (Blier et al, 1998): desensitization of the 5-HT_{1A} autoreceptors within the raphe is thought to occur after this period of time, allowing for an increase in the levels of 5-HT within projection sites, and consequently an increase in mood. This theory could be tested via administration of citalopram over a 14 day period, and a citalopram challenge (using CUMI) at baseline and day 14. Nevertheless, these results have implications for studying 5-HT transmission in neuropsychiatric disorders such as depression, as they highlight the *in vivo* effects of a single

administration of citalopram for the first time. It is interesting to note that there is evidence for an immediate change in 5-HT_{1A} functioning, neuroendocrine functioning (increased adrenocorticotrophin hormone and prolactin release; Mondelli et al, 2006) and cognitive changes (Harmer and Cowen, 2013) following a single dose of citalopram, but that mood takes up to 2 weeks to change (e.g. Blier et al, 2003). As such, these findings could support the notion that neurological and behavioural changes are necessary in order to allow those experiencing low mood to make different decisions and interpret outcomes in a more positive manner, before mood can increase (Harmer and Cowen, 2013).

3.4.2 Pruning task

The behavioural results from the pruning task replicated previous findings (Huys et al, 2012), in that the model including a specific pruning and Pavlovian factor was best at explaining the data. However, in this study the addition of a 'rho' parameter (to identify participants' subjective valuations of each of the 4 monetary outcomes) also improved the parsimony of the model, unlike in the original data (Huys et al, 2012). All participants except one had high specific pruning values, suggesting that they were pruning branches of the decision tree due to anticipated large losses, as predicted. However, no correlations between participants' specific pruning scores and 5-HT_{1A} availability in either condition survived correction for multiple comparisons. Whilst Dayan and Huys (2008) posit a theory by which 5-HT transmission could be involved in pruning, the results of this study were unable to confirm this hypothesis with respect to the 5-HT_{1A} receptor.

3.4.3 Choice x Risk

Participants' sensitivities to wins and losses were lower than the values from participants within the original study describing this task (means of 0.110 and 0.115 compared to means of 0.300 and 0.250 respectively; Rogers et al, 2003). Furthermore, these values are much lower than the values for sensitivity to probability (mean of 0.694), which is also higher than the original study (original mean of 0.520). Taken together, these data suggest that participants were not adequately using information pertaining to the magnitudes of potential gains and losses when making decisions, and were instead over-relying on information regarding the probability of outcome. As such, this may have affected the results of the correlations of performance with PET data, and may help to explain why Rogers et al (2003)

discovered a decrease in sensitivity to wins on this task after acute tryptophan depletion, but here a link between 5-HT transmission and win sensitivity was not identified.

The involvement of the hippocampal complex in participants' sensitivity to probability supports previous research that has highlighted a role for this structure in reward and punishment processing (i.e. Camara et al, 2008). Furthermore it supports the notion that 5-HT may have a role to play in this relationship, since those participants with greater 5-HT_{1A} availability at baseline were more likely to use information about the probability of winning when making their decisions on the gambling task. This finding is consistent with studies in which ATD decreased participants' choices of probable gain outcomes on the CGT (Rogers et al, 1999b); however, opposing results have also been reported, with Talbot et al (2006) reporting that ATD led to *increased* selection of more probable gain outcomes. Further, the fact that indices of participants' baseline 5-HT_{1A} receptor were obtained from the PET session, and their behavioural (sensitivity to probability) indices were obtained at a separate testing session may make this result difficult to interpret.

The involvement of 5-HT_{1A} receptors within the striatal ROI in participants' sensitivity to losses supports previous research that has highlighted a role for the striatum in punishment processing (e.g. Seymour et al, 2012). However, due to the fact that participants were administered citalopram in the PET session (from which the index of the change in 5-HT_{1A} receptor availability was obtained), but did not receive this SSRI during the behavioural session (from which participants' sensitivity to losses were obtained) makes this result difficult to interpret. Another difficulty in interpreting these results is that the results of the citalopram challenge were counter-intuitive. In the present study, participants who exhibited higher sensitivity to information pertaining to losses demonstrated increased 5-HT_{1A} receptor availability (indicative of decreased 5-HT release) in the right nucleus accumbens after citalopram infusion. This is supported by the work of Schmitz et al (2009), who were able to show a role for the 5-HT_{1A} receptor in punishment processing by demonstrating that a 5-HT_{1A} C(-1019)G polymorphism-linked increase in 5-HT_{1A} availability is correlated with a greater sensitivity to punishments. However, it will be important to clarify the robustness and direction of this effect in future work as the overall levels of CUMI binding in the striatum were very low.

3.4.4 Temporal Discounting

As expected participants were found to discount future rewards, and their discount factors and utility concavity scores were found to be similar to that from the original study (0.099 compared to 0.033 and 0.0086 compared to 0.0089; Pine et al, 2009). Whilst no correlations between participants' utility concavities and 5-HT_{1A} availability survived small volume correction in either condition, there was a significant negative correlation between participants discount factors and 5-HT_{1A} availability in the left hippocampal complex in the placebo condition. This supports previous research that has highlighted a role for this region in temporal discounting (i.e. Mobini et al, 2000 and Schacter and Addis, 2009), and previous work using ATD that has suggested and involvement of 5-HT in delay discounting (i.e. Schweighofer et al, 2008). In the present study participants with greater 5-HT_{1A} availability were less likely to discount the value of rewards that were available further in the future. Although the hippocampus is typically associated with episodic memory processing and contextual learning, Peters and Buchel (2010) describe a manner in which this region may contribute to temporal discounting. These authors administered a standard discounting paradigm, but with the addition of a novel episodic condition which involved the presentation of relevant future episodes (i.e. vacation in Paris) that coincided with the later time point. They were able to show that these 'episodic tags' decreased participants' discount rates, and through connectivity analyses, that this tag effect was associated with increased coupling between the ACC and the hippocampus bilaterally. As such, it could be very interesting to extend the current study using a similar paradigm in order to understand whether such episodic tags mediate this relationship between temporal discounting and hippocampal complex 5-HT_{1A} receptor availability.

3.4.6 Limitations

Several limitations of this study merit comment. Firstly, PET scans of only 15 participants were used in the baseline analysis, and 13 participants in the change analysis. This means that only large effects could be identified reliably, and that smaller effects may have been missed. However, such low statistical power can also result in false positive findings (such as the small volume corrected significant correlations observed above, e.g. Button et al, 2013). As such, increased power from a larger subject population may help to find effects that this

number of participants could not, and would help to increase the reliability and generalizability of the results.

Secondly, the particular subject population and testing conditions within this study could have contributed to some of the differences between the above and original results; participants here were older than typically included in cognitive neuroscience studies (mean 50.9, range 35-63) with only one female subject.

Thirdly, study-specific factors may have had an effect upon the results. The pruning task was always administered first, since the prediction relating to pruning formed our primary hypothesis. This could have meant that length of this long, complex task, and entire behavioural testing session (3-4 hours) may have affected participant's motivation and engagement with the task, leading to a failure to identify significant PIT results and relative lack of win and loss sensitivities in the Choice x Risk task.

Fourthly, this study has observed correlations of decision making behavior with only one 5-HT receptor. Whilst this receptor has previously been shown to be involved in decision-making (i.e. Schmitz et al, 2009), the fact that it exists in such small numbers in a brain region (striatum) known to be heavily involved in reward and punishment processing, along with impulsivity, does not provide us with strong sensitivity to identify correlations. In order to gain a more complete picture of 5-HT's role in decision making, it will be necessary to measure other 5-HT receptors (e.g. the 5-HT_2 receptor subtype) which are present in far greater numbers in the human striatum (Joyce et al, 1993).

Finally, this study suggests that it is possible to observe the relationship between 5-HT and decision making by looking at a specific receptor subtype, rather than simply observing the effects of a global decrease or increase in 5-HT using, for example, acute tryptophan depletion or SSRI administration. However, the fact that CUMI is a competitive agonist of the 5-HT_{1A} receptor, meaning that it can be displaced from these receptors, means that both 5-HT_{1A} receptor density and levels of extracellular 5-HT contribute to measured ^{11}C-CUMI-101 signal at baseline, with more receptors increasing the signal, and more extracellular 5-HT decreasing it. As such, whilst this research does allow us to make predictions about the effects of increasing or decreasing the former or latter upon decision making, it is difficult to know the extent to which performance on the above tasks correlated with the amount of

extracellular 5-HT or receptor density at baseline. This question could be addressed using a non-competitive ligand such as [11C]WAY-100635 that would allow a more definitive conclusion to be made regarding the nature of the observed correlations.

3.4.7 Conclusion

In conclusion, we found that the influence of 5-HT on decision making can be observed at the level of the receptor. This has implications for psychiatric disorders in which 5-HT transmission is hypothesised to be compromised, such as depression, in which abnormalities in reward/punishment processing and impulsivity are also seen (Eshel and Roiser, 2010) and 5-HT_{1A} receptors may be reduced (Drevets et al, 1999). It would be of great interest to use the individual differences approach we adopted in the present study to investigate whether the 5-HT_{1A} receptor is linked to poor decision making in depression and other neuropsychiatric disorders.

4) DECISION-MAKING 3 DAYS AFTER ADMINISISTRATION OF 3,4 – METHYLENEDIOXYMETHAMPTHETAMINE (MDMA)

4.1 Introduction

MDMA is a psychoactive stimulant of the amphetamine family that has been shown to target the serotonergic system (Fitzgerald and Reid, 1990). Effects of administration include euphoria, increased extroversion, heightened sensual awareness, mild perceptual alterations and increased physical energy (Curran and Travill, 1997), which typically last for around 2-4 hours (Gamma et al, 2000). MDMA is a recreational drug typically taken at clubs or raves in pill form, with each dose containing anywhere between 0-150mg of MDMA (Cole et al, 2002). However, many pills taken in social settings may also contain other psychoactive substances, and many users will also knowingly ingest other drugs and alcohol, meaning that much human MDMA research may be better characterised as MDMA-polydrug investigations (Cowan, 2007).

4.11 Pharmacology and action of MDMA

MDMA has been shown *in vitro* to increase levels of 5-HT by entering neurons and binding to the 5-HT transporter (5-HTT), reversing its normal function. Due to this rather than being able to re-uptake 5-HT from outside the cell and transport it back inside, the 5-HTT actively increases levels of 5-HT within the extracellular space (Fleckenstein et al, 2007, Rudnick and Wall, 1992). Stone et al (1986) demonstrated the subacute effects of MDMA administration (i.e. the effects of the drug once the cognitive and perceptual effects have subsided), reporting that MDMA decreases both TPH and 5-HTP, the latter of which is then itself synthesized into 5-HT by aromatic-L-amino acid decarboxylase) and 5-HT within the rat hippocampus, striatum and cortex 3 hours after a single sub-cutaneous injection of MDMA. Furthermore, the authors discovered that repeated sub-cutaneous injection of MDMA (5 injections at 6 hour intervals) led to a 75% decrease in TPH and a 30% decrease in 5-HT within the same brain regions. It is important to note here that metabolism in rats is much faster than that of humans, and that any effects of MDMA 3 hours after administration should be considered subacute effects equivalent to those occurring over a longer time period (days) in humans (Stone et al, 1986). O'Shea et al (2006) report that repeated MDMA administration produces

long term damage to the axons of serotonergic neurons, without a compensatory increase in 5-HT synthesis. Studies in humans have reported decreases in 5-HTT binding throughout the cortex (Kish et al, 2010) (which has also been shown to be inversely correlated to the number of previous MDMA exposures; Mcann et al, 1998) and (Ricaurte et al, 1990) report reduced cerebrospinal fluid 5-HT metabolite levels in chronic MDMA users. Finally, Kish et al (2000) report that MDMA exposure leads to decreased levels of 5-HT within the striatum.

4.1.2 Cognitive effects of MDMA administration

A number of studies have reported on the effects of MDMA on cognition, yet results are mixed. The most consistent cognitive effects of MDMA use are memory deficits (Kalechstein et al, 2007). However, some studies report no differences between MDMA users and controls, especially when groups are well matched for cannabis use (Croft et al, 2001, Dafters et al, 2003). For example, Raj et al (2010) observed no difference in MDMA users and controls who were well matched for cannabis use on a semantic verbal recognition task. However, the review by Kalechstein et al (2007) concludes that chronic MDMA users display deficits in attention, nonverbal learning and memory, motor/psychomotor speed and executive functions. However, none of the studies discussed above could exclude the possibility that any differences observed were pre-existing, since they all adopted cross-sectional designs.

The evidence for deficits in decision-making processes in ecstasy users is, however, less well understood. Quednow et al (2007) examined the decision-making and impulsivity behaviours of chronic but recently-abstinent MDMA users by comparing them with chronic but recently-abstinent cannabis users and controls on three decision making tasks: the Matching Familiar Figures Test (MFFT; Kagan et al, 1966 – Information processing in the child:), a Go/No-Go task (Newman et al, 1990 – Passive avoidance in psychopaths) and the Iowa Gambling Task (IGT; Bechara et al, 1994). MDMA users exhibited higher impulsivity on the MFFT and poorer performance on the IGT (but did not make more errors on the Go/No-Go task) when compared to both groups. These findings are supported by results of other studies that show regular ecstasy users to display increased impulsive responding compared to both poly-drug users and drug-naïve controls on the MFFT (e.g. Morgan et al, 2002 – see Clark et al, 2009) and the Stroop task (i.e. Halpern et al, 2004 – see Clark et al, 2009) and tests of delay discounting (e.g. Bickel and Marsch, 2001 – see Clark et al, 2009). However, Clark et al

(2009) reported that a cohort of previous ecstasy users and current ecstasy users did not display disrupted reflection impulsivity (the tendency to make decisions before gathering sufficient information to make a well-judged decision) compared to drug-naïve controls on an information sampling task. These results occurred despite the previous and current ecstasy users scoring significantly higher on the impulsivity subscale of the self-report Eysenck Impulsivness-Venturesomeness-Empathy questionnaire (interestingly the previous users scored (numerically) higher on this subscale than current users). As such, research findings on decision-making deficits in ecstasy users are mixed.

Many studies have examined the effect of MDMA use on scores on depression scales, comparing MDMA users with both polydrug users (e.g. Gamma et al, 2000, Parrott et al, 2000) and drug-naïve controls (i.e. Morgan et al, 2002, Gerra, et al, 1998). For example, Curran and Verheyden (2003) reported that ecstasy users scored nearly 3 points more on the BDI than poly-drug users. However, it must be noted that neither MDMA-users nor poly-drug users scored within the clinical range for depression in this study. Finally, Gerra et al 1998 report that MDMA-users score significantly higher on the Hamilton Rating Scale for Depression than non-drug users.

The above studies typically required an abstinence period of several weeks in order to assess medium-to-long term effects. However, Curran and Travill (1997) examined recreational users' mood levels both acutely and sub-acutely, and reported that users rated their mood as elevated immediately after MDMA administration, but that this became progressively lower over the next 4 days, with some recording mood within the range for clinical depression. The authors attribute this subsequent low mood to reflect MDMA-dependent 5-HT depletion that may occur days after administration. Few studies, however, have examined changes in cognition during this sub-acute period, and even fewer studies have been able to control for non-ecstasy drug use and sleep deprivation. This latter fact is due to the data being collected from drug users who self-administered a range of drugs in a naturalistic setting. As such, the present study, which examined the subacute effect of MDMA administration on cognition in a controlled design, should be more straightforward to interpret.

4.1.3 Study design and experimental hypotheses

The aim of the present study was to test the hypothesis from Dayan and Huys (2008) that pruning decision trees is dependent upon 5-HT transmission. Healthy volunteers were administered administered either MDMA or a placebo in a within subjects, counter-balanced, randomised design, as part of a larger study involving the administration of tasks inside an MRI scanner on the acute administration day, and the administration of a large battery of tasks on the subacute day (not inside such a scanner). The only data presented in this chapter are the data obtained from the pruning paradigm explained in chapter 2 (2.4.1.2), which was administered 3 days after they had been administered either MDMA or a placebo. Participants were administered pure MDMA in a clinical setting, meaning that this should not be considered to be an MDMA-polydrug investigation (Cowan, 2007). It was predicted that participants would display both decreased pruning and decreased mood 3 days after MDMA administration compared to 3 days after administration of a placebo, which would be due to the subacute 5-HT-depleting effects of MDMA. It was also predicted that the MDMA-induced decrease in mood would be associated with the MDMA-induced decrease in pruning, based upon Dayan and Huys (2008) hypothesis that low levels of serotonin cause lower mood by decreasing pruning.

4.2. Methods

4.2.1 Participants

Nineteen participants (12 males, mean age 31.2 years, range 21-46 years) were included in the study. One subject (female) was subsequently excluded from the analyses due to a failure to follow task instructions, leaving data from 18 participants. The within subjects design of this task, with an *N* of 18 gives this study 79% power to detect an effect size of 0.80 or above (which is classified as a 'large' effect size; Cohen, 1988), and 52% power to detect an effect size of .50 or above (which is classified as a 'medium' effect size). Subjects were free of psychiatric disorders as determined by the MINI (Sheehan et al, 1998) at acute visit 1. Although past exposure to MDMA was an inclusion criterion for the study, participants had also taken no MDMA for at least 7 days prior to the study and no other drugs for at least 48 hours prior to the acute visits, confirmed by a urine screen. A breathalyser test confirmed that no participants had consumed alcohol prior to the acute visit. Participants had used MDMA an average 35 times (range 1-200) before entering the study, and the median time since last usage was 260 days (range 7 to 7300 days). Informed written consent was obtained at the beginning of both acute visit 1 and subacute visit 1, and ethical approval was provided by the West London Research Ethics Committee, Imperial College Healthcare NHS Trust and Imperial College London's Faculty of Medicine, and the study was conducted in accordance with Good Clinical Practice guidelines. A Home Office Licence was obtained for the storage and handling of a Schedule 1 drug. Ethical approval was gained from the UCL ethics committee for the subacute visits.

4.2.2 Procedure

All participants underwent administration of either 100mg of placebo (encapsulated ascorbic acid/vitamin-C) or 100mg of MDMA at session one (acute visit 1), and then the alternative at session two (acute visit 2), at the Hammersmith Hospital campus of Imperial College London, in a within-subjects, counterbalanced, double-blind, placebo-controlled design. This part of the study was conducted by Prof. Val Curran (University College London) and Prof. David Nutt (Imperial College London). Participants completed the two acute sessions at least a week apart. All participants then participated in the behavioural session at The Psychopharmacology Unit, University College London 3 days after both acute visit 1 and 2

(subacute visit 1 and 2, respectively; one participant completed their first subacute visit 4 days after acute visit 1 due to illness). This behavioural session was completed by myself.

Participants began the subacute visit by completing the BDI and Visual Analogue Mood scales, and then began behavioural testing. This involved administration of 4 cognitive tasks: a self-referential encoding task, a self-referential recognition task, an angry stories task and the pruning paradigm. Only data from the latter task were analysed as part of this study, with the data from the other tasks being analysed by the research group of Prof. Val Curran (University College London). The pruning task was administered either first or last in the task sequence, in a counter-balanced manner.

The pruning task was administered as reported in chapter 2 (experimental methods), with the transition matrix training being first, followed by the reward matrix training and then the main task. As described in the methods section 2.4.1.2, this version of the pruning task contained large punishing transitions that cost the participants 70p (-70 transitions), and was timed. The transition matrix training was omitted in week 2 (subacute visit 2). Participants were compensated up to £20, depending on performance on this task.

4.2.3 Statistical Analyses

The variables of interest for this task were the measures of 'proportion best remaining' (defined in the methods section as the proportion of LLO trials in which participants did not take the optimal sequence of transitions due to the fact that it contained a large negative, and instead took the next best sequence that did not contain a large negative), and the 'difference estimate' (defined in the methods section as difference between the proportion of trials in which the optimal sequence of moves was made on NLLO compared to LLO trials. Other variables of interest include the proportion of optimal choices made on each trial type calculated for 3, 4 and 5 move problems. The reaction times on each trial type were also analysed. For a more complete description of task variables, see page 50. Due to certain variables suffering from deviations from parametric assumptions, a set of transformations was applied to the data: an arcsine (square root) transformation was applied to proportion optimal scores on both NLLO and LLO trials from each treatment condition, a log transform was applied to all reaction time data, and a square root transform was applied to all data concerning the number of missed trials in each condition.

Repeated measures ANOVAs were run on the data. Firstly, in order to assess the effect of prior MDMA administration on proportion optimal scores on both NLLO and LLO trials, and therefore the difference estimate, a 2x2x3 factor ANOVA was conducted, with prior treatment (MDMA or placebo), trial type (NLLO or LLO), and depth (3, 4 or 5 moves) as factors. Secondly, in order to assess the effect of MDMA on proportion best remaining scores, a 2x3 ANOVA was calculated, with prior treatment (MDMA or placebo), and depth (3, 4 or 5 moves) as factors. Thirdly, in order to assess the effect of MDMA on reaction times on each of the two trial types (without difficulty factor), a 2x2 ANOVA was performed, with treatment (MDMA or placebo) and trial type (NLLO or LLO) as factors The order of treatment administration (i.e. MDMA in week 1 or week 2) was added as a between-subjects factor in all ANOVAs so as to examine any possible practice effects.

In order to analyse the mood data, a series of analyses was performed. Firstly, paired t-tests were performed in order to examine any differences between conditions in mood scores on the BDI and subscales 2 (Discontented-Contented), 3 (Amicable-Antagonistic), 4 (Annoyed-Composed), 5 (Happy-Sad), 6 (Calm-Anxious), 10 (No Euphoria-Extreme Euphoria), 11 (Lethargic-Energetic), 20 (Compassionate-Indifferent), 24 (Shy-Self Confident) and 27 (Not at all high-Extremely High) on the VAS. Secondly, Pearson's correlation analyses were run on both the data from the MDMA, and the difference between the placebo and MDMA conditions in order to observe any relationships between the above psychometric mood scores and both the proportion best remaining and difference estimate scores from the pruning task.

4.3 Results

4.3.1 Practice Effects

Participants performed more optimally on both NLLO and LLO trials in week 2 compared to week 1: main effect of order ($F(1,16)=11.644$, $P=.004$). There was also a trend towards a practice effect (increase) on participants proportion best remaining scores: main effect of order ($F(1,13)=3.737$, $P=.075$), but no significant practice effect on participants' difference estimate scores (trial type*order interaction ($F(1,16)=.079$, $P=.783$). Finally, participants' reaction times were significantly shorter in week 2 compared to week 1: main effect of order ($F(1,16)=8.057$, $P=.012$). There was however no significant trial type*order interaction, indicating that these practice effects did not affect reaction times on each trial type differently ($F(1,16)=.184$, $P=.674$). For a full summary of these practice effects, see table 4.1.

Variable	Week 1	Week 2
Proportion Optimal (NLLO)	0.71 (.23) *	0.84 (.19) *
Proportion Optimal (LLO)	0.39 (.14) *	0.52 (.15) *
Proportion Best Remaining	.42 (.23)	.51 (.26)
Difference Estimate	.32 (.22)	.32 (.23)
Reaction Times (NLLO)	490ms (104) *	428ms (110) *
Reaction Times (LLO)	501ms (134) *	438ms (93) *

Table 4.1. Summary of means and standard deviations (in brackets) of each variable in week 1 and 2. Red asterisks denote a statistically significant difference between week 1 and week 2

4.3.2 Treatment Effects

4.3.2.1 Difference Estimate

Participants performed significantly better (attaining a higher proportion of optimal choices) on NLLO relative to LLO trials ($F(1, 17)=39.256$, $P=<.001$), confirming the presence of a significant difference estimate. This difference estimate was significantly more marked at higher depths, as shown by a significant trial type*depth interaction ($F(2,34)=5.127$, $P=.012$). However, prior MDMA exposure did not affect participants' overall proportion optimal

scores (F(1, 17)=.001, P=.978), confirming that participants were not simply worse at planning overall. There was a trend towards MDMA reducing the difference estimate (see figures 4.1 and 4.2), as indicated by the treatment*trial type interaction (F(1,17)=3.305, P=.088). Prior MDMA exposure however did not affect the difference estimate differently at each depth, (treatment*trial type*depth interaction F(2,34)=1.434, P=.253). For a summary of difference estimate scores see table 4.2.

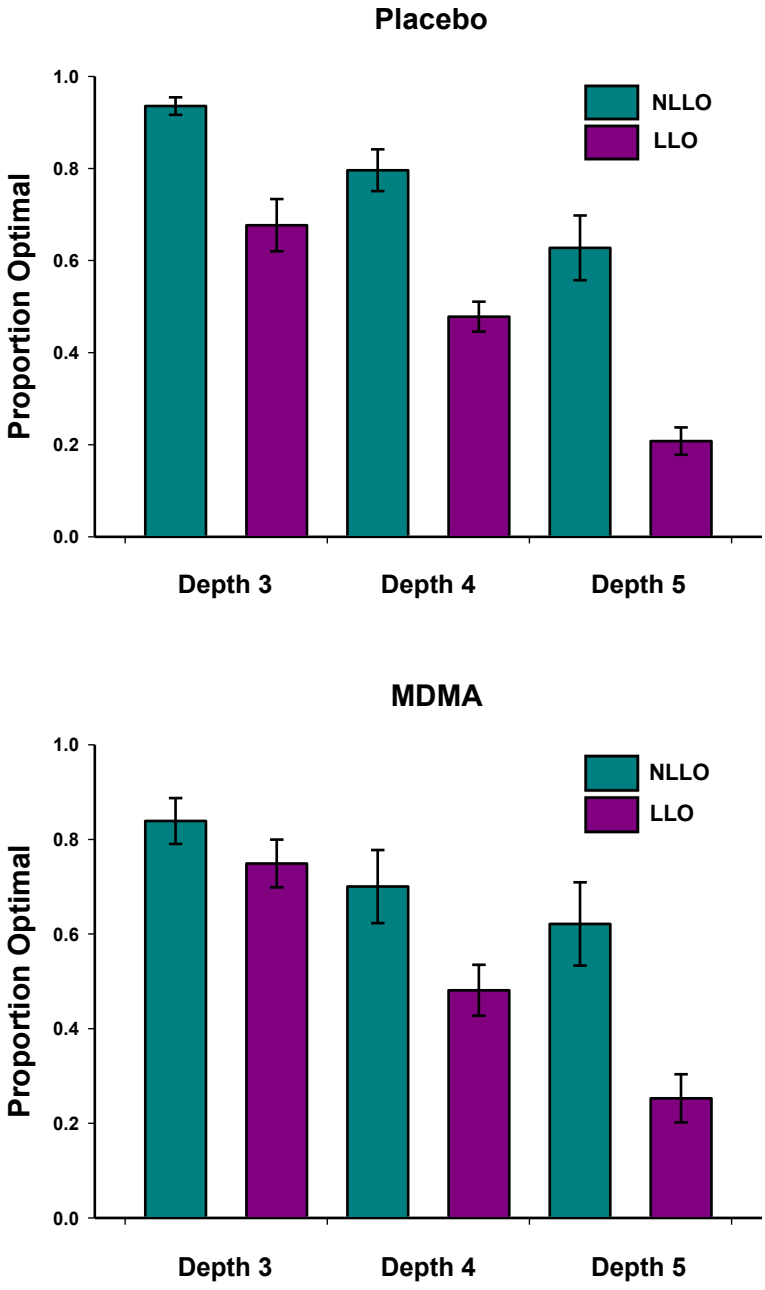


Figure 4.1. Proportion optimal scores on NLLO and LLO trials within the placebo (top) and MDMA conditions (bottom). Note that the difference between proportion optimal scores on each trial

type at all depths is smaller in the MDMA plot, signifying a decrease in the difference estimate after MDMA administration

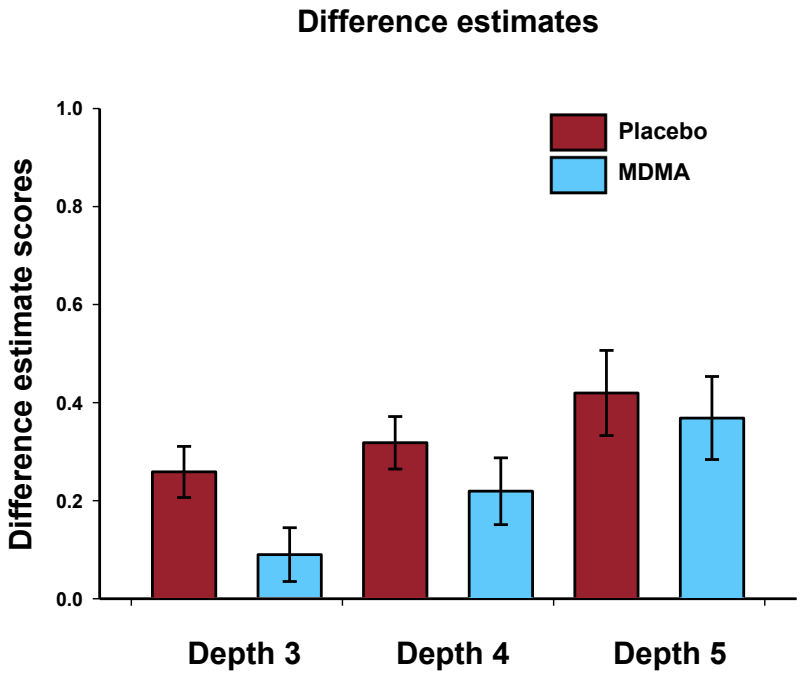


Figure 4.2. Difference estimates between proportion optimal scores on each trial at each depth

4.3.2.2 Proportion Best Remaining

Prior MDMA exposure was found to reduce proportion best remaining scores (see figure 4.3; $F(1,17)=7.478$, $P=.017$). However, MDMA had a similar effect upon proportion best remaining scores at each depth, as shown by no treatment*depth interaction ($F(2,12)=.819$, $P=.464$). For a summary of proportion optimal data see table 4.2, and figure 4.3.

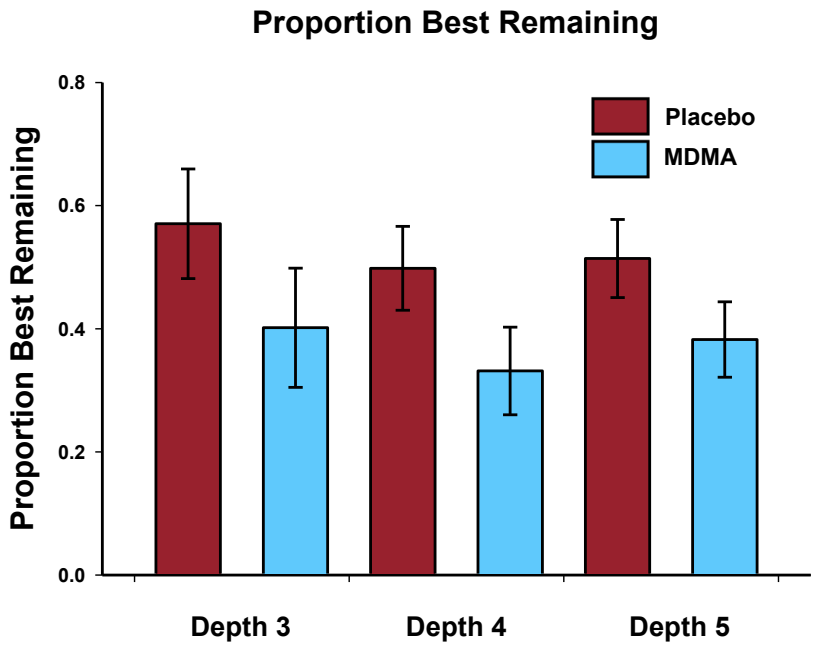


Figure 4.3. Proportion best remaining scores in each condition at each depth

The MDMA-induced change in proportion best remaining scores (i.e. post-MDMA scores minus post-placebo scores) and the MDMA-induced change in difference estimate scores were significantly positively correlated (r =.486, P=.048).

4.3.2.3 Reaction Times

There was no main effect of trial type (NLLO vs LLO; (F(1,16)=.842, P=.372).

Furthermore, there was no significant effect of treatment on reaction times overall (F(1,16)=.097, P=.760). Further, there was no significant effect of prior MDMA administration upon reaction times on either trial type (F(1, 17)=.007, P=.936). For a summary of reaction time data see table 4.2.

Variable	Placebo	MDMA
Proportion Optimal (NLLO)	0.81 (.14)	0.74 (.27)
Proportion Optimal (LLO)	0.43 (.13)	0.48 (.19)
Proportion Best Remaining	.55 (.23)*	.38 (.24)*
Difference Estimate	.38 (.18)	.26 (.25)

Reaction Time (NLLO)	457ms (108)	461ms (116)
Reaction Time (LLO)	469ms (100)	470ms (138)

Table 4.2. Summary of means and standard deviations (in brackets) of each variable in the placebo and MDMA conditions. Red asterisk denotes a statistically significant difference between conditions

4.3.2.4 Mood data

Analysis of mood scores from the BDI and VAS scales with repeated measures ANOVA (including drug order as a between subjects factor) revealed no order effects (all $P>.116$), and no effects of prior MDMA exposure (all $P>.149$) upon participants mood scores. For a summary of the psychometric data, see table 4.3 below.

Variable	Placebo	MDMA
BDI	1.53(2.50)	1.29(2.26)
Discontented-Contented	8.17(1.67)	8.53(1.17)
Amicable-Antagonistic	2.00(2.45)	1.59(1.23)
Annoyed-Composed	8.47(1.55)	9.12(1.05)
Happy-Sad	2.06(2.08)	2.12(1.45)
Calm-Anxious	1.77(1.82)	1.88(1.36)
No Euphoria-Extreme Euphoria	0.76(1.75)	1.00(1.90)
Lethargic-Energetic	6.88(1.87)	6.76(2.05)
Compassionate-Indifferent	2.81(1.83)	3.59(2.43)
Shy-Self Confident	7.06(1.91)	7.06(2.30)
Not at all high-Extremely high	0.13(0.34)	0.24(0.56)

Table 4.3. Summary of means and standard deviations (in brackets) of each psychometric variable in the placebo and MDMA conditions. For all variables except BDI, a low score denotes being more of the first descriptive of the variable (e.g. Discontented), and a higher scores denotes the opposite (e.g. Contented).

Correlational analyses were performed with mood scores from the BDI and VAS and both proportion best remaining scores and the differences estimate scores from the MDMA

condition. There was a significant negative correlation between proportion best remaining scores and the scale 'Annoyed-Composed' (i.e. higher annoyance correlated with higher proportion best remaining scores: r=-.510, P=.037), a trend positive correlation between the scale 'Amicable-Antagonistic' and the difference estimate (i.e. higher antagonism correlated with higher difference estimate scores: r=.472, P=.056), and a trend negative correlation between the scale 'Annoyed-Composed' and the difference estimate (i.e. higher annoyance correlated with higher difference estimate scores: r=-.478, P=.052).

Further, correlational analyses were performed in order to examine the relationship between MDMA-induced change (i.e. the differences between treatment conditions) in the proportion best remaining, difference estimate, and MDMA-induced change in BDI and VAS scores. Trend positive correlations were found between the scale 'Amicable-Antagonistic' and the difference estimate (i.e. higher MDMA-induced antagonism correlated with higher difference estimate scores: r=.452, P=.068) and the scale 'Happy-Sad' and the difference estimate (i.e. higher MDMA-induced sadness correlated with higher difference estimate scores: r=.470, P=.057). The direction of these correlations is counter to that which was hypothesised: increased negative mood is positively correlated with increased pruning (see figure 4.4). No correlations were found between the difference in proportion best remaining scores and any variables on the BDI or VAS.

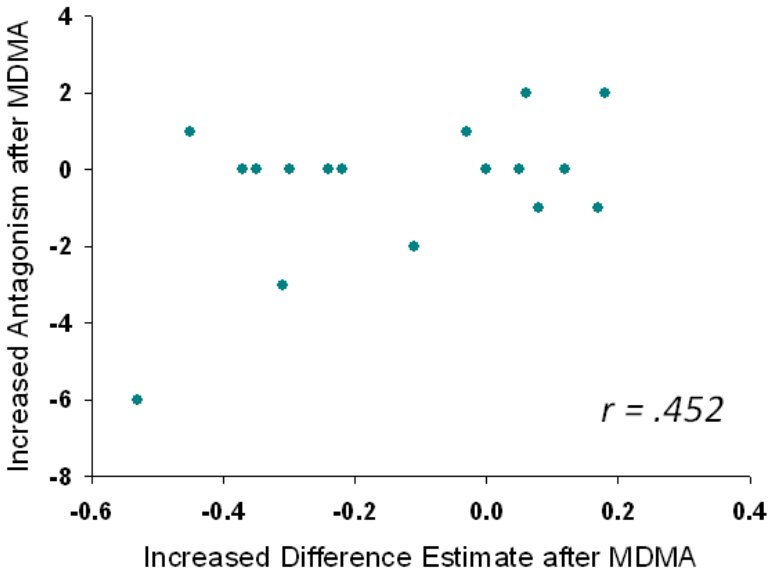


Figure 4.4. Positive correlation between negative affect and pruning: increased negative mood (antagonism) following MDMA is related (at trend level) to increased pruning

4.4 Discussion

This study used a within subjects, randomised, placebo-controlled design in order to investigate the impact of prior MDMA administration, thought to depleted levels of 5-HT, upon pruning. Healthy volunteers were administered either a placebo or a dose of MDMA 3 days before they were administered the pruning task described in the methods section 2.4.1.2, with the hypothesis that participants would have depleted levels of 5-HT 3 days after MDMA administration, and as such would exhibit decreased pruning. The results indicated that MDMA administration 3 days prior to performance on the pruning task did weaken participants' pruning compared to placebo. However, participants' mood was not decreased 3 days after MDMA administration. There was a trend correlation between participants' negative mood and their pruning as shown by the difference estimate, yet this was in the opposite direction to that which was hypothesized. This study will be discussed and considered in light of previous work by Huys et al (2012) and Dayan and Huys (2008).

4.4.1 Pruning Data

As expected participants displayed greater proportion optimal scores on NLLO trials compared to LLO trials (which is consistent with pruning) and made more optimal choices at lower depths, indicating that they found these depths easier to compute. Assessment of the treatment effects showed that MDMA decreased both participants' proportion best remaining scores and difference estimate scores (although the latter was only decreased at the trend level), which could be interpreted as being due to the expected low levels of 5-HT experienced by participants 3 days after administration of MDMA, and thus providing support for the theory that pruning is dependent upon normal 5-HT functioning (Dayan and Huys, 2008). Neither the proportion best remaining or difference estimate scores were affected differently at each depth. Further, prior MDMA treatment did not affect reaction times on any trial type. Finally, whilst participants did display improved performance on both NLLO and LLO trials in week 2, they displayed only a trend effect upon their difference estimate, and no such effect on their proportion best remaining scores, indicating that practice did not significantly influence pruning.

4.4.2 Mood Data

Examination of participants' mood scores revealed no significant differences between the conditions, indicating that MDMA did not reduce mood 3 days after administration. A number of correlations between pruning and mood data were identified. Firstly, in the MDMA condition annoyance correlated positively with both proportion best remaining and difference estimate scores, and antagonism correlated positively with the difference estimate. Secondly, MDMA-induced increase in antagonism correlated positively with MDMA-induced increase in the difference estimate. Dayan and Huys (2008) and Huys et al (2012) propose a theory that links low mood with abnormal 5-HT functioning (e.g. Everett and Toman, 1959, Coppen 1967) and 5-HT functioning with pruning. However, no correlation between BDI scores and pruning was seen in the current sample, possibly due to the facts that the range of BDI scores was relatively low, and that BDI scores did not differ between each condition. The above correlations with annoyance and antagonism are in direct opposition to the original theory (posited by Dayan and Huys, 2008) which states that negative affect is linked to decreased pruning. However, Huys et al (2012) reported a significant positive correlation between scores on the BDI and pruning scores, again indicating that the more participants displayed negative affect, the more they pruned. However, it is important to note that these correlations (both significant and at trend level) were not corrected for multiple comparisons and should thus be considered as uncorrected. As such, the mood data of this study not only fail to provide evidence for Dayan and Huys (2008) theory, but also provide a little support the mood data from Huys et al (2012) which opposes it.

4.4.3 Limitations and further work

Several limitations of this study merit comment. Firstly, the number of participants included in this within subjects design was low compared to other behavioural studies, especially those using a between subjects design (i.e. studies of MDMA users). However, whilst simply increasing the number of participants in a study can often lead to an increase in significance levels (Wagenmakers et al, 2011), the fact that MDMA was found to affect pruning with such a low number of participants increases confidence in this effect.

Secondly, the participants included in the study all had a history of abusing drugs such as heroin, marijuana, cocaine, nicotine and even MDMA (the latter of which was taken by some

participants over 2000 times in their lifetime). These drugs have been shown to affect the 5-HT, DA, norepinephrine and cholinergic systems, meaning that these participants may have damage of an unknown nature to these systems, which could affect their pruning abilities due to alterations in cognitions such as reward and punishment processing. As such, one direction for future study could be to administer the pruning paradigm to participants who have been administered MDMA 3 days prior, but who have never taken any illicit drugs. One caveat of this however is that such a study may raise ethical problems; participants in this study were only accepted if they had prior experience with MDMA without any adverse experiences in order to decrease the chances of such experiences occurring during testing. This would be impossible to ensure in those who have never taken any form of illicit drug before.

Thirdly, it was expected that participants would display decreased subacute mood due to a putative decrease in 5-HT; however, this was not observed. This prediction was made based on work by Curran and Travill (1997), which showed that participants who recreationally administered MDMA at the weekend reported low mood in midweek. However, this prior study may be confounded by the possible multitude of drugs/alcoholic beverages that may have also been ingested by the participants, along with the fact that these participants may have had disturbed sleep patterns. Furthermore, the concentration of MDMA ingested by these participants was not measured, and could have been much lower than expected, and even mixed with other drugs/substances. All of this could have affected both 5-HT and mood levels in these participants 3 days after their Saturday night self-administration. The mood data from the present study reflect the mood of participants who have not taken MDMA or any other drug/alcohol for at least 7 days, and had taken no other drugs or alcohol at the same time, unlike the users in the Curran and Travill (1997) study above. Furthermore, the participants in the current study were not sleep deprived as many of the users in the above study would have been; participants in this study were administered MDMA at roughly 9-11am in the morning, and returned home at roughly 5-6pm, meaning that they had ample rest on the day of administration, which could affect subacute mood levels. The fact that subacute effects of MDMA on pruning were observed indicates that these effects occurred over and above mood effects, and that 5-HT transmission may not have recovered on day 3.

Fourthly, whilst this study does provide some support for Dayan and Huys (2008) theory that pruning is dependent upon normal 5-HT function, the data within the study fail to support the notion that low mood is related to decreased pruning. Indeed, the opposite was observed;

negative affect was positively correlated with pruning, which is interestingly in the same direction as the correlation between negative affect (depression scores) and pruning observed in Huys et al (2012).

Finally, exploring the role of genetic influences upon the cognitive effects of MDMA may be useful in better understanding the effect of this drug upon pruning behaviours. It has been shown, for example, that regular users of MDMA who carry a COMT *val/val* polymorphism or the 'short' allele of the serotonin transporter (SERT *s/s*) displayed poorer performance on tasks of visuospatial attention and memory (Cuyas et al, 2011), and Roiser et al (2006) report that the increase in attention to differences in the probability of winning on a risky decision-making task that occurred in healthy controls carrying the short allele of SERT was attenuated in ecstasy users who carried this same allele. Studies such as these highlight a role for individual variability in the effects of drug use upon cognition, and as such examining certain polymorphisms may help improve our understanding of the subacute (and even acute) effects of MDMA upon decision-making behaviours. However, this would require testing a much larger sample than was included in the current study.

4.4.4 Conclusion

In conclusion, the results of this study suggest some evidence for an effect of MDMA administration on pruning 3 days later. However, no subacute effect of MDMA on mood was observed. These results are hypothesized to be due to a decrease in 5-HT, which lowered participants' abilities to prune efficiently without decreasing mood 3 days after administration. Surprisingly, pruning was positively correlated with negative mood. The results of this study provide some evidence to support Dayan and Huys (2008) and Huys et al (2012) hypothesis that pruning is dependent on normal 5-HT functioning, though they contradict the notion that this leads to negative mood.

5) THE INFLUENCE OF ACUTE TRYPTOPHAN DEPLETION ON THE DECISION-MAKING ABILITIES OF HEALTHY VOLUNTEERS

5.1 Introduction

5.1.1 Acute Tryptophan Depletion

Acute tryptophan depletion (ATD) is a dietary technique used experimentally in order to attempt to decrease levels of 5-HT within the brain. It is thought to do this by decreasing levels of 5-HT's precursor, the large neutral amino acid (LNAA) L-tryptophan (TRP). Synthesis of 5-HT in the human brain is dependent upon availability of TRP, which is firstly synthesised into 5-Hydroxy-L-Tryptophan (5-HTP) by Tryptophan Hydroxylase (TPH), and then into 5-HT by Aromatic-L-amino acid decarboxylase (DDC) in the brain (van Donkelaar, 2011). Whilst 5-HT cannot cross the blood brain barrier (BBB), TRP can, meaning that decreased levels of 5-HT in the brain can be achieved by increasing levels of the remaining 5 LNAAs in the blood. These LNAAs then act as 'competition' to TRP, with all 7 LNAAs (leucine, isoleucine, valine, methionine, tyrosine and phenylalanine) competing for entry into the brain via the LNAA transporter at the BBB. Administering LNAAs without TRP decreases the TRP:LNAA ratio in the blood leading to a hypothesised decrease in levels of TRP (and thus 5-HT) in the brain (Crockett et al, 2012).

5.1.2 Mood and cognitive effects of Acute Tryptophan Depletion

Administration of ATD has been shown to reinstate depressive symptoms in patients who have recovered from depression (Delgado et al, 1990, Smith et al, 1999). Patients with a more severe form of depression (i.e. those who required administration of SSRIs, or those who experienced many episodes) are at increased risk of mood change following ATD treatment (Booij et al, 2002, Delgado et al, 1990), as are those who are vulnerable to depression but have never been depressed themselves (i.e. those with a family history: Ellenbogen et al, 1999, Stewart et al, 2002). Furthermore, ATD has also been shown to affect neural metabolism and responsivity in similar regions to those implicated in depression. For example, Morris et al (1999) examined the neural correlates of such transient depressive symptom increases caused by ATD treatment using PET, and identified decreased perfusion

within the orbitofrontal cortex (OFC) and increased co-variation between the habenula and the dorsal raphe nucleus (DRN), with this coupling correlating negatively with plasma tryptophan levels, and positively with mood scores on the Hamilton Rating Scale for Depression (HAM-D). However, O'Reardon et al (2004) were able to show that ATD only produces this transient recurrence in symptoms in roughly 50-60% of patients who have recovered using SSRI treatment, and that those patients who have recovered by other means (e.g. cognitive-behavioural therapy) rarely experience such symptoms due to ATD treatment.

Much research has also been conducted into the cognitive effects of ATD treatment. For example, ATD treatment has been shown to affect the processing of emotional stimuli (e.g. Klaassen et al, 2002, Murphy et al, 2002), and performance on tasks of verbal learning tasks of episodic memory (e.g. Riedel et al, 1999, Schmitt et al, 2000). Mendelsohn et al (2009) argue that ATD treatment does not affect spatial memory, and studies have argued ATD to have no effect on tasks of declarative memory (e.g. Park et al, 1994, Porter et al, 2000, 2005) or semantic memory (Allen et al, 2006, Amin et al, 2006, Gallagher et al, 2003). Further, tasks that tap executive functions such as working memory, attention, response inhibition, planning and cognitive flexibility show either no detrimental effect of ATD upon performance, or in some cases improved performance (e.g. Talbot et al, 2006, Luciana et al, 2001, Schmitt et al, 2000, Clark et al, 2005, Cools et al, 2005a: see Mendelsohn et al (2009) for a review).

5.1.2 Effects of Acute Tryptophan Depletion upon decision-making

Whilst ATD has been found to have little effect on the performance of various cognitive tasks (excluding emotional processing and episodic memory) this dietary technique has been shown to influence decision-making in healthy volunteers. For instance, Schweighofer et al (2008) reported that participants who had undergone ATD displayed increased discounting of future rewards relative to those who had undergone acute tryptophan loading (to increase levels of 5-HT). However, whilst the results of this study are supported by those of Tanaka et al (2007), Crean et al (2002) reported a lack of ATD effects upon temporal discounting in both males with and without a family history of alcoholism.

Further, ATD has also been shown to influence decision making by altering the manner in which participants respond to information pertaining to both rewards and punishments;

Rogers et al (2003) reported that participants made more decisions based on rewarding information than sham-treated controls on a gambling task, whilst Cools et al (2008) claim that ATD affects punishment (but not reward) prediction. Cools et al (2005) also report that ATD induced faster but less accurate responses to cues that were predictive of high reinforcement certainty compared to cues that were predictive of lower reinforcement sensitivity on the cued-reinforcement reaction time task (CRRT), thus modulating the coupling between motivation and action. Further, Roiser et al (2006) administered ATD to both *ll* and *ss* homozygotes of the 5-HTT gene before they performed the CRRT. The authors report that ATD abolished reinforcement-related speeding on trials with a higher probability of reinforcement compared to trials with a low probability of reinforcement in the *ss* genotypes (similar to the result reported by Cools et al, 2005), whereas this reinforcement-related speeding was still present following ATD in the *ll* genotypes. The results of this study thus demonstrate that the cognitive effects of ATD may only be observed in specific subtypes of individuals.

Whilst results do not completely agree on whether ATD affects solely reward or punishments (or both), Seymour et al (2012) administered a decision-making task that allowed the authors to examine both rewards and punishments separately, and discovered that ATD altered the exchange rate by which rewards and punishments were compared, leading to an attenuation of the subjective representation of reward values. Finally, Crockett et al (2012) administered an information sampling task in which participants could sample as much information as they wanted, at a small local cost for each piece of information, in order to avoid making an incorrect decision which incurred a larger global cost, and discovered that ATD reflexively removed the suppressive effect of smaller local costs on information sampling behaviours. The authors explained this finding in terms of work by Dayan and Huys (2008) and Huys et al (2012) that characterises 5-HT as promoting reflexive avoidance of negative outcomes.

5.1.3 Study design and experimental hypotheses

The aim of the present study was to examine the role of 5-HT in decision-making by administering a battery of tasks to participants who had undergone ATD or sham-depletion in a within subjects, counter-balanced, randomised design.

Participants' total TRP and LNAA levels were measured in order to observe the effectiveness of the ATD procedure, and related to performance on the three decision-making tasks below between conditions.

Firstly, the pruning task described in the methods section *2.4.1.2* was administered. Based upon theoretical work by Dayan and Huys (2008), which proposes that 5-HT provides a Pavlovian inhibitory signal in the face of punishment, and the experimental work by Huys et al (2012), it was predicted that participants would display decreased pruning behaviours after ATD compared to sham.

Secondly, the gambling task described in the methods section *2.4.2* was administered. Based on previous work (Rogers et al, 2003) it was predicted that participants would display altered processing of information pertaining to both rewards and punishments after ATD compared to sham.

Finally, the temporal discounting task described in the methods section *2.4.3* was administered. Based on previous work it was predicted that participants would display a more impulsive decision-making style, choosing the smaller, sooner option when administered ATD compared to sham.

5.2. Methods

5.2.1 Participants

Thirty-one healthy volunteers (13 male, mean (SD) age 30.82 (7.45) years, range 22-51 years) were included in the study. Four participants (all female) withdrew from the study, failing to return for the second session (1 no-show, 1 due to having already completed the pruning task in another study, and 2 due to experiencing adverse effects of the drink), meaning that analyses were performed on 27 volunteers. A within subjects design with an *N* of 27 gives this study 97% power to detect an effect size of .80 (which is defined as a 'large effect size'; Cohen et al, 1988), and 71% power to detect an effect size of .50 (a 'medium' effect size). Participants were free of psychiatric disorders as determined by administration of the MINI (Sheehan et al, 1998). Inclusion criteria included no past or present psychiatric disorders or alcohol/substance dependence (alcohol/substance abuse was allowed if not within the past 6 months) as assessed by MINI and HAM-D (Hamilton, 1960). Participants were also free of any medication at the time of participating. Informed written consent was obtained from all subjects at the beginning of the session, and ethical approval was obtained from the UCL ethics committee. Participants were compensated for their time, which included being paid £35 for their time and inconvenience plus up to £20 (depending on performance) on the pruning task per session, meaning that participants could leave with between £70 and £110 for the entire study.

5.2.2 Procedure

All participants underwent administration of either ATD or sham depletion (both described in methods section *2.6.2*) at session one, and the opposite at session two at the Wellcome Trust Centre for Neuroimaging, University College London in a within subjects, counterbalanced, double-blind, placebo-controlled design. Participants completed the two sessions at least a week apart. Participants were initially asked questions from the MINI over the phone, in order to identify their potential suitability for participation. At the testing session, participants firstly answered questions from the entire MINI interview, along with questions from the HAM-D, Beck Depression Inventory (BDI; Beck, 1961), Profile of Mood States (POMS) the personality scale Neuroticism, Extraversion, Openness (NEO; Costa and McCrae, 1985), the State/Trait Anxiety Inventory (STAI; Speilberger et al, 1983) and the verbal section of the

Weschler Test for Adult Reading (WTAR; Weschler, 2001), as described in the methods sections *2.2 – 2.3*. Once these were complete, suitable participants then had 5ml of their blood taken at time 1 by medically qualified members of staff so that baseline levels of the LNAAs could be identified. Once this was complete, the relevant LNAA mixture (see 5.2.2.1) was mixed with 5g of flavouring (Grapefruit, Lemon-Lime or Cherry-Vanilla, depending on the participants' preference) and 450ml of water, which was then ingested. Participants were told that they could take as long as they wanted to drink this unpalatable drink, but that they needed to finish the entire drink. Of the participants who did not withdraw, only 2 (both female) did not drink the entire drink (one ingested ~75%, and the other ~66%). Of these two participants, only one ingested less drink than the other participants (~66% total) in the TRP- condition, but was one of the five participants from whom a blood sample could not be obtained at each of the four time points, meaning that they were excluded from the amino acid analyses and thus did not affect results (see *5.3.1* below). Task data from these two participants were not excluded from the below analyses.

Once this was complete, testing began 5 hours later in order to ensure that the levels of LNAAs would be altered maximally. As such, participants were trained on the pruning task immediately after drink ingestion, and then allowed to rest in the waiting area of the imaging department until 4 hours 45 minutes had passed since ingestion. Participants then answered the state anxiety section of the STAI, a second blood sample was taken, (in order to examine levels of the LNAAs again; time 2), and then began the battery of tests. All participants completed the battery of tests (the pruning paradigm, gambling task and temporal discounting paradigm described in the methods section *2.4*) within 100 minutes.

5.2.2.1 Amino acid mixtures

The quantities of amino acids in each drink were based on those used in (Roiser et al, 2006), in which robust reductions in plasma tryptophan levels were observed. Amino acid mixtures were:

TRP+ : L-alanine, 4.1g; L-glycine, 2.4g; L-histidine, 2.4g; L-isoluecine, 6g; L-leucine, 10.1g; L-lysine, 6.7g; L-phenylalaine, 4.3g; L-proline, 9.2g; L-serine, 5.2g; L-threonine, 4.9g, L-tryosine, 5.2g; L-valine, 6.7g, L-arginine 3.7g, L-cysteine, 2.0g;, L-methionine, 2.3g; L-tryptophan, 3g – total: 78.2g

TRP- : L-alanine, 4.1g; L-glycine, 2.4g; L-histidine, 2.4g; L-isoluecine, 6g; L-leucine, 10.1g; L-lysine, 6.7g; L-phenylalaine, 4.3g; L-proline, 9.2g; L-serine, 5.2g; L-threonine, 4.9g, L-tryosine, 5.2g; L-valine, 6.7g, L-arginine 3.7g, L-cysteine, 2.0g;, L-methionine, 2.3g – total: 75.2g

The same amino acid levels were administered to all participants, regardless of gender or height/weight.

5.2.2.2 Analysis of plasma amino acid levels

Following blood draws using the BD Vacutainer system ® using EDTA tubes (http://www.hdft.nhs.uk/EasysiteWeb/getresource.axd?AssetID=4865&), the plasma was separated by centrifugation and stored at -20^{o}C. Concentrations of total LNAA levels (tyrosine, leucine, isoleucine, valine, phenylalanine, methionine and tryptophan, from which the LNAA:TRP ratio was also calculated) were measured by means of HLPC using fluorescence end-point detection and pre-column sample derivatisation adapted from the methods used by Furst et al (1990). The limit of detection used 5nmol/ml using a 10µl volume sample. The inter-assay and intra-assay coefficients of variation were <15% and <10% respectively. These analyses were carried out by Dr Michael Franklin of Faculty of Life Sciences, Oxford Brookes University.

5.3.3 Statistical analyses

All data were analysed in the Statistical Package for Social Sciences (SPSS 19, SPSS Inc., Chicago, IL). The main outcomes for the pruning task were the same as in chapter 4 (proportion best remaining, the difference estimate, proportion optimal scores and reaction times on each trial type). The main outcomes for the gambling task include sensitivity to probabilities, sensitivity to wins and sensitivity to losses, and for the temporal discounting task were participants' discount factor (k), utility concavity (r) and the number of sooner choices made.

Due to certain variables not meeting normality criteria some of them were transformed: an arcsine (square root) transformation was applied to proportion optimal scores from each trial

type, and a log transform was applied to reaction time data from the pruning task. Further, an arcsine transform was applied to the variables from the gambling task. No variables from the temporal discounting were transformed.

The statistics performed on the pruning task data mirror those in chapter 4 exactly. Briefly, they include a 2x2x3 ANOVA to observe treatment effects, with treatment (ATD or sham), trial type (NLLO and LLO) and depth (3, 4 or 5 moves) as factors, a 2x3 ANOVA to assess the effect of treatment upon proportion best remaining scores, with treatment as two levels and depth as three, and a 2x2 ANOVA to assess reaction times on each of the two trial types, with both treatment and trial type constituting two levels. Regarding the gambling task, a 2x2x2x2 ANOVA was computed, with treatment constituting two levels (ATD and sham), and each of probability, win and loss constituting two (high and low). With regards to the temporal discounting task, ANOVAs were computed including participants' discount factors, utility concavities and the number of sooner, smaller choices made, with treatment (ATD of sham) as the only within-subjects factor. The order of treatment administration (i.e. sham in week 1 or week 2) was added as a between-subjects factor into all ANOVAs in order to assess any order effects. Finally, in order to further assess the relationship between decreased tryptophan levels and performance on the above decision-making tasks, the difference between the LNAA:TRP ratio at time 1 and time 2 in the TRP- session was added as a covariate in exploratory analyses.

5.3 Results

5.3.1 Plasma amino acid concentrations

Due to difficulties in taking blood, plasma samples were not available at all four time points for five subjects (2nd time point TRP- x2, 2nd time point sham x2, and 1st time point sham x1). As such, the below analyses pertain to the remaining 22 participants (14 males, mean (SD) age = 31.24 (8.12) years, range = 22-51 years).

Total tryptophan concentration (nmol/ml) in the plasma was lower overall on the depletion day (mean = 38.26, SD = 9.19) than the sham day (mean = 91.70, SD = 21.45, $F(1, 20) = 161.889$, $P < .001$) and was lower overall at time point 2 than time point 1 ($F(1, 20) = 9.650$, $P = .006$). As expected treatment had a different effect upon total tryptophan concentration at the 2nd time point in the sham session and TRP- session ($F(1, 20) = 130.700$, $P < .001$). Post-hoc analyses indicated that tryptophan concentration in the plasma increased 126.5% from time 1 to time 2 on the sham day (mean time 1 = 56.16, SD = 12.50, mean time 2 = 127.24, SD = 40.42, t[21] = 7.994, $P < .001$), and decreased 73.3% from time 1 to time 2 on the depletion day (mean time 1 = 60.39, SD = 13.97, mean time 2 = 16.12, SD = 5.66, t[21] = 19.239, $P < .001$). Importantly, there was no difference between mean tryptophan concentrations between sham and depletion days at time 1 (t[21] = 1.343, $P = .194$). Furthermore, there was no effect of order of drink administration ($F(1, 20) = .782$, $P = .388$) or gender ($F(1, 20) = .685$, $P = .419$) on tryptophan levels. It should be noted that all interactions with gender were non-significant (all $P>0.1$).

The tryptophan:LNAA ratio was also lower overall on the depletion day (mean = 0.096, SD = 0.031) than the sham day (mean = 0.172, SD = 0.043, $F(1, 20) = 94.823$, $P < .001$) and was lower overall at time point 2 than time point 1 ($F(1, 20) = 27.105$, $P = < .001$). There was a significant treatment x time interaction ($F(1, 20) = 105.583$, $P < .001$), and post-hoc analyses indicated that there was a 17.2% increase in the tryptophan:LNAA ratio from time 1 to time on the sham day, which showed a trend towards being significant (mean time 1 = .1560, SD = .046, mean time 2 = .1885, SD = .015, t[21] = 1.931, $P = .067$), and a significant 85.1% decrease in this ratio from time 1 to time 2 on the depletion day (mean time 1 = .1661, SD = .054, mean time 2 = .0247, SD = .012, t[21] = 13.860, $P < .001$). Importantly, there was again no difference between this ratio between days at time 1 (t[21] = .937, $P = .359$). Finally,

again there was no effect of order of drink administration ($F(1, 20) = 1.319$, $P = .266$) or gender ($F(1, 20) = 1.042$, $P = .321$) on the TRP:LNAA ratio. It should again be noted that all interactions with gender were non-significant (all $P>0.1$).

5.3.3 Pruning task

Five participants obtained proportion correct scores of less than 40% on the NLLO trials in both weeks, indicating either that they were not concentrating, or that they could not perform the task adequately. As such, these 5 were excluded from the analyses, meaning that the below analyses were performed on the remaining 22 participants (13 males, mean (SD) age = 30.59 (6.71) years, range = 22-46 years).

5.3.3.1 Practice effects

Participants performed more optimally on both NLLO and LLO trials in the second week compared to the first, ($F(1, 20) = 28.419$, $P<.001$). There was also a practice effect on participants' proportion best remaining scores ($F(1, 19)=10.104$, $P=.005$), but no significant practice effect upon participants' difference estimate scores, ($F(1, 20)=.003$, $P=.958$). Finally, participants' reaction times were not significantly shorter in week 2 compared to week 1: $F(1, 20)=.342$, $P=.565$. For a summary of these data see table 5.1.

Variable	Week 1	Week 2
Proportion Optimal (NLLO)	0.75 (.17) *	0.84 (.15) *
Proportion Optimal (LLO)	0.33 (.15) *	0.45 (.17) *
Proportion Best Remaining	.43 (.22) *	.62 (.27) *
Difference Estimate	.42 (.22)	.39 (.21)
Reaction Times (ms: NLLO)	450 (104)	422 (75)
Reaction Times (ms: LLO)	440 (137)	447 (81)

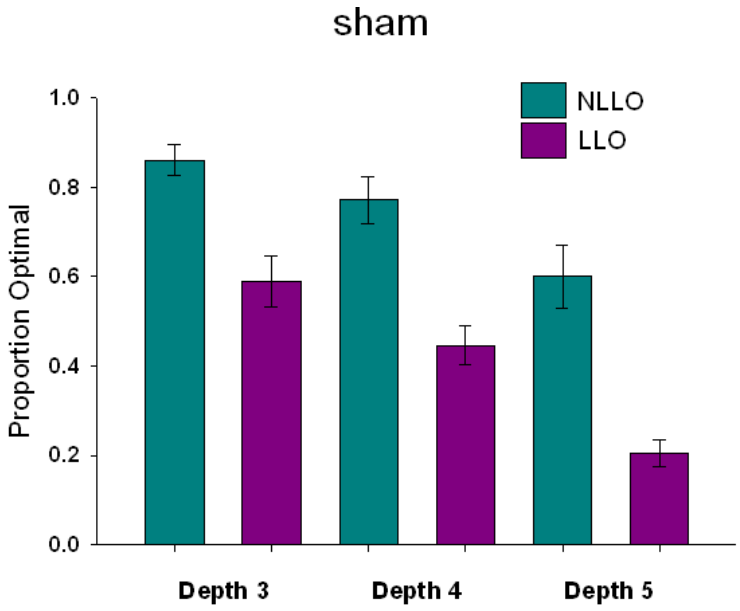
Table 5.1. Summary of means and standard deviations (in brackets) of each variable in week 1 and 2. Red asterisks denote a statistically significant difference between week 1 and week 2

5.3.3.2 Treatment effects

5.3.3.2.1 Difference estimate

When examining proportion optimal scores, participants made significantly more optimal decisions on NLLO trials than LLO trials, consistent with pruning ($F(1, 20)=78.437$, $P=<.001$). Participants also performed more accurately at lower depths, as shown by a significant main effect of depth ($F(2, 19)=80.860$, $P<.001$). The difference estimate was however no more marked at higher depths, as shown by a lack of a significant trial type*depth interaction ($F(2, 19)=1.309$, $P=.281$).

ATD treatment had no effect upon participants' proportion correct scores ($F(1, 20)=1.718$, $P=.205$). Treatment also had no effect on the difference estimate as shown by a lack of a significant treatment*trial type interaction ($F(1, 20)=1.136$, $P=.299$; see figures 5.1 and 5.2). Further, treatment did not have a significantly different effect at different depths as shown by the lack of a significant treatment*depth interaction ($F(2, 19)=.548$, $P=.582$). Finally, treatment did not affect the difference estimate differently at each depth, shown by the lack of a significant treatment*trial type*depth interaction ($F(2, 19)=.552$, $P=.580$). For a summary of participants' proportion optimal scores on each trial type in each condition see table 5.2.



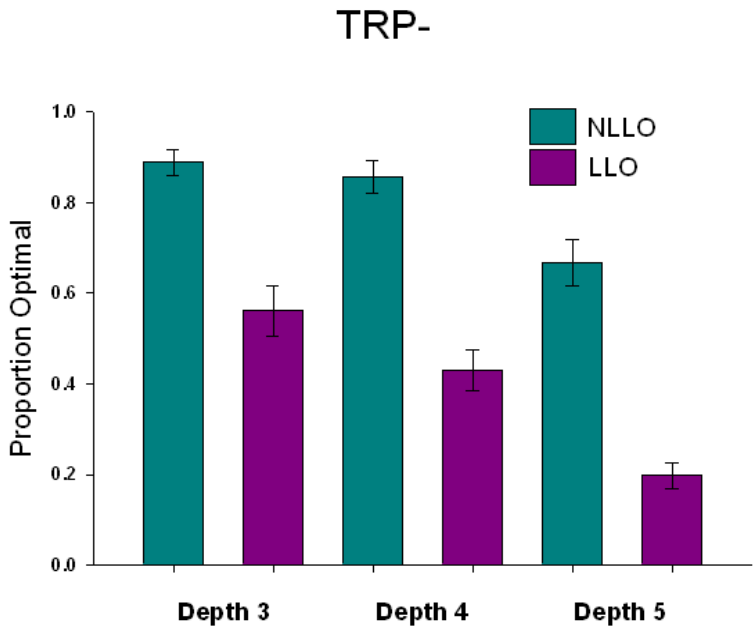


Fig. 5.1. Proportion optimal scores on NLLO and LLO trials within the sham (top) and TRP- conditions (bottom)

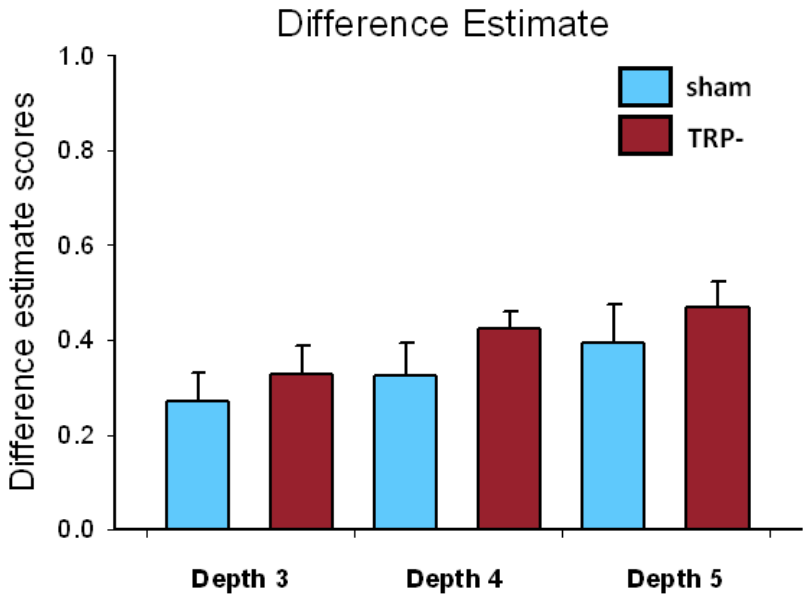


Fig. 5.2. Difference estimates between proportion optimal scores on each trial at each depth

5.3.3.2.2 Proportion best remaining

ATD treatment was found to have no effect upon participants' proportion best remaining scores ($F(1, 19)=.302$, $P=.589$), (figure 5.3). Furthermore, ATD treatment was found to not

have a significantly different effect at each depth ($F(2, 18)=.164$, $P=.850$). For a summary of proportion best remaining scores see table 5.2.

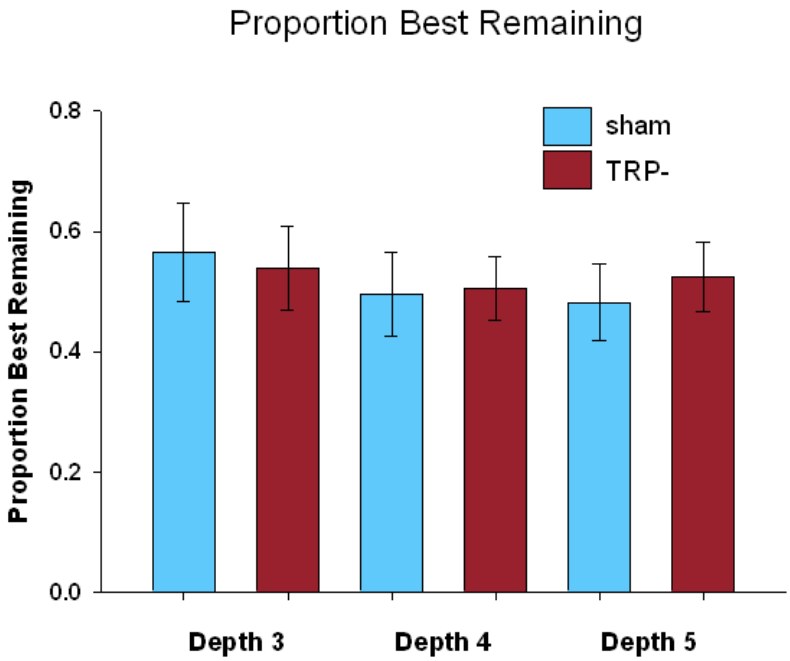


Figure 5.3. Proportion best remaining scores in each condition at each depth

5.3.3.2.3 Reaction times

There was no significant effect of treatment upon participants' reaction times across NLLO and LLO trial types ($F(1, 20)=.317$, $P=.579$). However, there was a trend towards a difference in the effect of treatment upon participants' reaction times on each trial type ($F(1, 20)=2.414$, $P=.051$), with ATD treatment numerically *decreasing* participants' reaction times on NLLO trials and numerically *increasing* reaction times on LLO trials (see figure 5.2 below). However, neither change in reaction times was significant in post-hoc analyses (both $P>.176$). For a summary of participants' reaction times, see table 5.2.

Variable	Sham	TRP-
Proportion Optimal (NLLO)	0.76 (.18)	0.82 (.14)
Proportion Optimal (LLO)	0.39 (.18)	0.38 (.17)
Proportion Best Remaining	.53 (.28)	.52 (.25)
Difference Estimate	.37 (.25)	.44 (.16)
Reaction Time (ms: NLLO)	443 (105)	428 (76)

Reaction Time (ms: LLO)	426 (130)	461 (88)

Table 5.2. Summary of means and standard deviations (in brackets) of each variable in the sham and TRP- conditions. There were no statistically significant differences between treatment conditions

5.3.4 Gambling task

For a summary of participants' sensitivities to probability, wins and losses across both conditions, see table 5.3.

Variable	Both Conditions
Sensitivity to Probability	.1510 (.141)
Sensitivity to Wins	.1777 (.168)
Sensitivity to Losses	.6267 (.276)

Table 5.3. Summary of participants' mean (SD) sensitivity to probability, wins and losses across both conditions

5.3.4.2 Practice effects

Participants displayed no practice effects (meaning that they did not display a significantly increased choosing of high or low probability ($F(1, 25)=.188$, $P=.668$) win ($F(1, 25)=.112$, $P=.741$) or loss ($F(1, 25)=2.126$, $P=.157$) gambles in week 1 or week 2). Further, there were no practice effects upon participants' reaction times to probabilities, wins and losses, as shown by the lack of main effects of order upon each of these variables ($F(1, 25)=.107$, $P=.747$, $F(1, 25)=.312$, $P=.582$, $F(1, 25)=893$, $P=.354$, respectively). For a summary of these data, see table 5.4.

Variable	Week 1	Week 2
Sensitivity to Probability	.6123 (.221)	.6412 (.326)
Sensitivity to Wins	.1586 (.151)	.1412 (.132)
Sensitivity to Losses	.2001 (.193)	.1551 (.140)

RT high probability (ms)	2176 (1067)	1576 (774)
RT low probability (ms)	2379 (1022)	1846 (821)
RT high win (ms)	2271 (1000)	1713 (813)
RT low win (ms)	2284 (1070)	1709 (729)
RT high loss (ms)	2376 (1112)	2179 (961)
RT low loss (ms)	1777 (752)	1645 (775)

Table 5.4. Summary of means and standard deviations (in brackets) of each variable during week 1 and week 2

5.3.4.3 Treatment effects

As expected, participants chose the experimental gamble significantly more often when its probability of winning was higher ($F(1, 25)=165.261$, $P<.001$), when the amount that could be won was higher ($F(1, 25)=31.986$, $P<.001$) and the amount that could be lost was lower ($F(1, 25)=40.929$, $P<.001$).

Following ATD participants showed a trend towards choosing the experimental gamble less often overall than following sham (($F(1, 25)=3.542$, $P=.072$). For participants' sensitivity to high and low probabilities, wins and losses in each treatment session, see figure 5.4. Examining each sensitivity separately, there was no effect of treatment upon sensitivity to win or loss, as shown by the treatment*win and treatment*loss interactions ($F(1, 25)=.047$, $P=.830$ and $F(1, 25)=.043$, $P=.838$, respectively). However, there was a trend towards an effect of treatment upon participants' sensitivity to probability ($F(1, 25)=3.099$, $P=.091$), with participants' sensitivity to probability decreasing after ATD treatment (mean (SD) sham = .666 (.226), TRP- = .588 (.318)). For these treatments effects see figure 5.4.

Post-hoc analyses revealed that ATD treatment reduced participants' sensitivity to high probabilities (mean (SD) sham = 3.365 (.579), TRP- = 3.065 (.728), t[26]=2.061, $P=.049$), reduced participants' sensitivity to small wins (mean (SD) sham = 1.741 (.461), TRP- = 1.574 (.497), t[26]=2.360, $P=.026$), and produced a trend towards a reduction in participants' sensitivity to small losses (mean (SD) sham = 2.380 (.361), TRP- = 2.245 (.407), t[26]=1.815, $P=.081$; see figure 5.5).

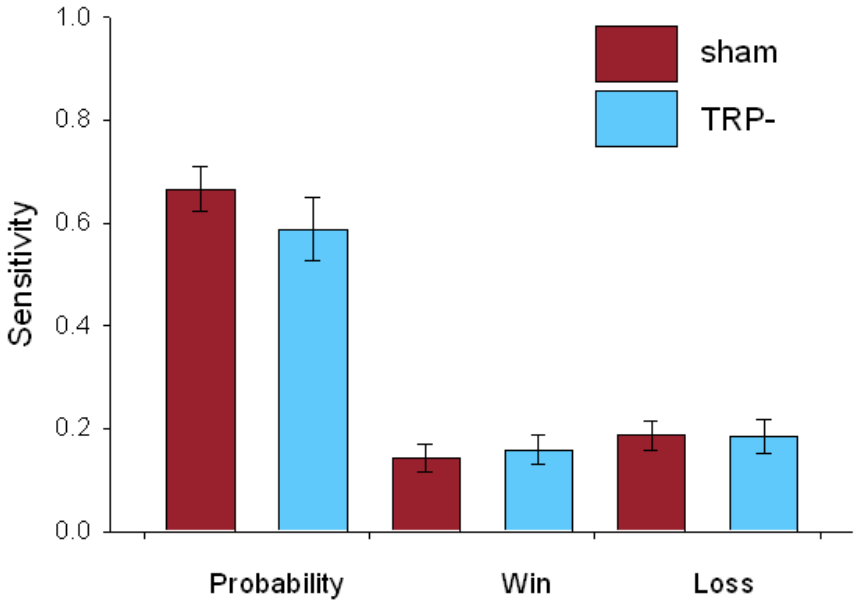


Figure 5.4. Mean sensitivities to probabilities, wins and losses in each condition

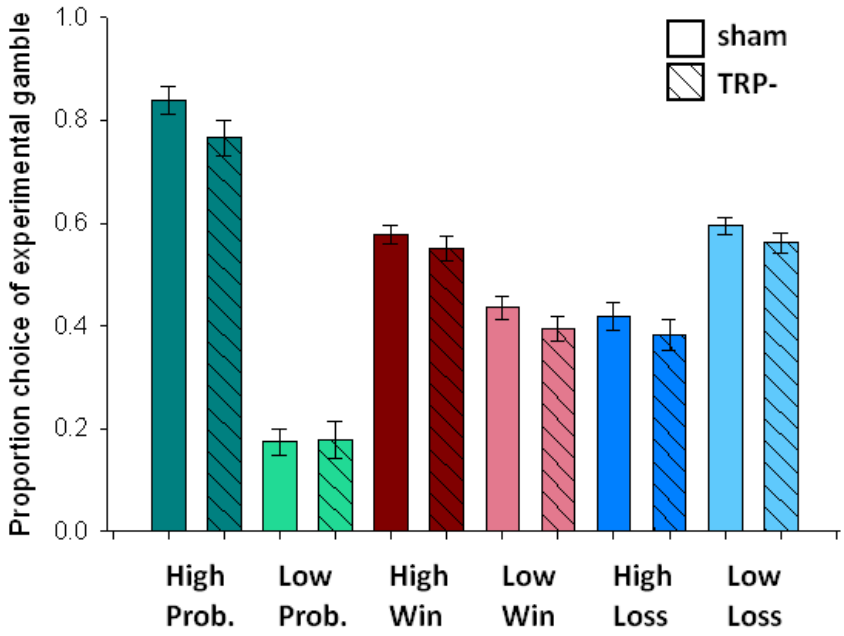


Figure 5.5. Mean choice of the experimental gamble at high and low probabilities, wins and losses in each condition. ATD treatment produced a significant reduction in participants' sensitivity to high probabilities and low wins, and a trend towards a reduction in sensitivity to low losses

Finally, treatment did not affect participants' choosing of the experimental gamble differently between weeks, as shown by non-significant treatment*order interactions with regards to

their sensitivity to probabilities ($F(1, 25)=.014$, $P=.907$), wins ($F(1, 25)=1.135$, $P=.297$) or losses ($F(1, 25)=1.163$, $P=.291$).

5.3.4.4 Reaction times

For a summary of participants' reaction times to high and low probabilities, wins and losses, see table 5.5 below.

There was a main effect of probability ($F(1, 25)=8.742$, $P=.007$), with participants reacting quicker to information pertaining to high probability, and a main effect of loss ($F(1, 25)=11.770$, $P=.002$), with participants reacting quicker to information pertaining to a smaller loss. There was however no main effect of win ($F(1, 25)=.007$, $P=.932$).

ATD treatment did not have an effect upon participants' reaction times ($F(1, 25)= 882$, $P=.357$), and specifically there were no effects of treatment upon participants reaction times to probability, win or loss as shown by a lack of significant treatment*RT_probability, treatment*RT_win and treatment*RT_loss interactions ($F(1, 25)=.087$, $P=.771$, $F(1, 25)=.473$, $P=.498$ and $F(1, 25)=1.339$, $P=.258$, respectively).

Reaction Times	*Sham*	*TRP-*
Probability (ms)		
High	1789 (980)	1963 (973)
Low	2040 (946)	2184 (979)
Wins (ms)		
High	1895 (957)	2089 (941)
Low	1934 (948)	2058 (969)
Loss (ms)		
High	2019 (983)	2134 (1007)
Low	1810 (918)	2013 (898)

Table 5.5. Participants' mean (SD) reaction times to information pertaining to high and low probabilities, wins and losses on the gambling task

5.3.5 Temporal Discounting

Data for this task were lost for the first two participants, and two participants were found to be not concentrating sufficiently on the task, choosing the larger, sooner option on a mean of 14 (SD = 2.83) trials in the sham condition, and on a mean of 14.5 (SD = 0.71) trials in the TRP- condition. As such, these participants were excluded from the analyses, meaning that the below analyses were conducted on 23 participants (13 males, mean (SD) age = 30.38 (7.28) years, range = 22-51 years).

5.3.5.1 Practice effects

For a summary of the means for each variable in each week, see table 5.5.

Participants were found to be concentrating sufficiently on the task in both weeks, choosing the larger sooner option on over 97% of the 20 catch trials in both weeks 1 and 2. There was no significant difference between performance on these catch trials between weeks as shown by a lack of a treatment*order interaction ($F(1, 23)=.112$, $P=.741$).

There were significant order effects upon participants' discount factors ($F(1, 23)=6.638$, $P=.017$), with participants discounting future rewards more in the second week (mean (SD) week 1 = .068 (.038), week 2 = .103 (.071)). There were however no significant order effects upon participants utility concavities ($F(1, 23)=1.909$, $P=.180$) or the number of sooner choices made (excluding catch trials; $F(1, 23)=.436$, $P=.516$). For a summary of participants' performance on this task in both weeks see table 5.6.

Variable	Week 1	Week 2
Catch trials (/20)	19.52 (0.73)	19.48 (0.59)
Sooner, smaller choices (/200)	121.09 (47.17)	119.22 (52.85)
K	.0687 (.038) *	.104 (.071) *
R	.027 (.033)	.019 (.037)

Table 5.6. Summary of means and standard deviations (in brackets) of each variable during week 1 and week 2. Red asterisk denotes a statistically significant difference between weeks

5.3.5.2 Treatment effects

For a summary of participants' scores on each of the task variables in each session, see table 5.6 below.

Participants were found to be concentrating sufficiently on the task in both conditions, choosing the larger, sooner option on over 97% of the 20 catch trials in the both the sham and TRP- conditions, and treatment had no effect upon participants' performance on these catch trials ($F(1, 23)=.112$, $P=.741$).

Using the model of best fit from Pine et al (2009), it was discovered that participants discounted the value of future rewards in both conditions: however, ATD treatment had no effect upon participants' discount factors, meaning that participants did not discount the value of a future reward any differently after ATD than after sham ($F(1, 23)=.030$, $P=.864$). Using the same model it was discovered that participants also displayed a concave utility function. However, treatment had no significant effect upon participants' utility concavities ($F(1, 23)=.898$, $P=.353$). Finally, ATD treatment had no effect upon the number of sooner, smaller choices made by participants ($F(1, 23)=1.086$, $P=.308$). For a summary of participants performance on this task in each condition, see table 5.7.

Variable	sham	TRP-
Catch trials (/20)	19.52 (0.59)	19.48 (0.73)
Sooner, smaller choices (/200)	116.92 (48.91)	119.72 (49.38)
k	.084 (.059)	.081 (.061)
r	.027 (.051)	.034 (.047)

Table 5.7. Summary of means (SDs) of each variable during sham and TRP- depletion

5.3.2 Mood data

Participants' scores on the BDI and on each subscale of the POMS were analysed in separate ANOVAs. Firstly, there was no effect of treatment upon participants' BDI scores ($F(1, 24)=.041$, $P=.842$).

Secondly, there was no effect of treatment upon participants' state anxiety scores on the STAI ($F(1, 24)=.002$, $P=.965$). There was however a trend towards a significant difference between participants' state anxiety scores between each time point ($F(1, 24)=3.757$, $P=.064$). There was also a trend towards a significant difference between the different time points in each treatment session, as shown by the treatment*time interaction ($F(1, 24)=3.119$, $P=.090$). Post-hoc analyses revealed that whilst these scores were very similar between time points in the sham condition (mean(SD) time 1 = 10.35(8.65), time 2 = 10.62(7.71)), participants' state anxiety scores increased significantly from time point 1 to time point 2 in the TRP- condition (mean (SD) TRP- time 1 = 9.42, time 2 = 11.54, $t[25]=2.948$, $P=.007$).

Thirdly, there was no effect of treatment upon participants' scores on the depression, tension/anxiety or anger/hostility subscales of the POMS (all $P>.144$). There were no differences between participants' scores on the subscales between each time point (all $P>.259$). Finally, there was no difference between these scores at different time points in each treatment session (all $P>.163$).

Finally, there were found to be no significant order effects on any mood scores (all $P>.412$). For a summary of participants' mood data at each time point within each session see table 5.8 below.

Variable	Sham (time 1)	Sham (time 2)	TRP- (time 1)	TRP- (time 2)
LNAA:TRP ratio	.1462 (.053)	.1779 (.077)	.1688 (.055)	.0352 (.035)
BDI	1.88 (2.57)	-	1.96 (2.39)	-
STAI (state)	10.35 (8.65)	10.62 (7.71)	9.42 (6.34) *	11.54 (6.42) *
POMS (tension-anxiety)	10.36 (3.88)	10.57 (3.89)	10.16 (2.72)	9.57 (2.79)
POMS (depression)	17.28 (6.59)	17.17 (6.40)	16.84 (3.80)	16.35 (3.69)
POMS (anger-hostility)	14.24 (4.68)	13.96 (4.09)	14.17 (4.75)	13.83 (4.12)
POMS (vigour-activity)	24.32 (6.34)	23.32 (5.87)	21.44 (5.19)	21.09 (4.63)
POMS (fatigue)	9.76 (3.06)	9.92 (3.00)	10.87 (3.51)	11.78 (3.77)

POMS (confusion)	8.04 (2.21)	8.48 (1.92)	8.39 (2.17)	9.00 (2.17)

Table 5.8. Summary of LNAA :TRP ratio and mood data at each time point. Data are displayed as a means (SDs). Red asterisks denote a significant difference

Due to the fact that the only observed significant difference was between participants' STAI state scores at time point 1 and 2 in the TRP- condition, the difference between STAI state scores at each time point was calculated and correlated with performance on the above decision-making tasks. However, these correlations were all non-significant (all $P>.141$).

5.3.3 LNAA:TRP ratio covariate

5.3.3.1 Pruning task

There was no significant correlation between participants' difference estimate and the difference between the LNAA:TRP ratio at time point 1and 2 on the TRP- session ($F(1, 19)=.109$, $P=.745$). Further, there was no significant correlation between this ratio and participants' proportion best remaining scores ($F(1, 18)=.933$, $P=.347$), and finally there was no correlation between this ratio and participants' reaction times ($F(1, 19)=1.078$, $P=.312$).

5.3.3.2 Gambling task

There was no significant correlation between participants' sensitivity to probability, win or loss and the difference between the LNAA:TRP ratio at time point 1 and 2 on the TRP- session ($F(1, 19)=1.706$, $P=.207$). Further, there was no correlation between this ratio and participants' reaction times to information pertaining to probability, win or loss ($F(1, 19)=.070$, $P=.795$).

5.3.3.3 Temporal discounting task

There was no significant correlation between participants' discount factor on the TRP- session and the difference between the LNAA:TRP ratio at time point 1 and 2 in this session ($r=-.137$, $P=.564$). Further, there were no significant correlations between this ratio and

participants' utility concavity (r=.145, P=.541) or the number of sooner, smaller choices made on the task (r=.088, P=.711).

5.4 Discussion

This study used a within subjects, randomised placebo-controlled design in order to investigate the impact of depleted levels of 5-HT on decision-making. Healthy volunteers were administered either acute tryptophan depletion (ATD) or a sham depletion 5 hours before they were administered the pruning task described in the methods section *2.4.1.2*, the gambling task described in *2.4.2* and the temporal discounting task described in *2.4.3*. The hypotheses were that participants would exhibit decreased pruning on the pruning task, altered processing of information pertaining to both rewards and punishments on the gamble task, and an increased choosing of the smaller, sooner option after being administered ATD compared to the sham depletion. The results indicated that, contrary to both the hypotheses and previous work, ATD administration had no significant effect on participants' performance on any of the behavioural tasks administered, although a trend towards treatment both decreasing participants' sensitivity to information pertaining to probabilities and decreasing their overall inclination to gamble was observed on the gambling task. The results of this study will be discussed in light of previous studies that have argued that 5-HT may modulate performance on these decision-making processes.

5.4.1 Pruning data

Participants displayed improved performance on both NLLO and LLO trials in week 2 compared to week 1. Further, these practice effects increased their proportion best remaining scores, indicating that whilst there were no such effects upon the difference estimate, practice increased participants' pruning behaviours. Furthermore, as expected participants displayed greater proportion optimal scores on NLLO trials compared to LLO trials (which is consistent with pruning) and made more optimal choices at lower depths, indicating that they found these depths easier to compute. Assessment of the treatment effects showed that ATD did not affect either the difference estimate or the proportion best remaining scores, indicating that decreased 5-HT did not decrease pruning behaviours, and the addition of the LNAA:TRP ratio as a covariate in the ANOVA revealed that these variables did not correlate with participants' tryptophan levels. As such, these results provide no support for the theory that pruning is dependent upon 5-HT functioning (Dayan and Huys, 2008). Finally, prior ATD treatment did not affect reaction times on any trial type.

5.4.2 Gambling task data

Participants chose the experimental gamble more often when the probability of winning was high, the amount that could be won was high, and the amount that could be won was low, supporting the results of Rogers et al (2003). As expected there were no practice effects upon participants' choices on this task, but contrary to expectations there was also no effect of treatment, nor were there any correlations between task variables and the LNAA:TRP ratio. However, ATD showed a trend towards decreasing participants' sensitivities to probability, specifically reducing proportionate choice of high probability gambles.

Whilst this trend result is supported by Rogers et al (1999b) who also observed a reduction in high probability choices after ATD on the CGT, they conflict those of Talbot (2006) who report an *increase* in high probability choices after ATD on the same task. Further, the fact that no significant effects were observed on sensitivity to wins on this task is in direct contradiction to the results of the original study using this task (Rogers et al, 2003). The authors of this original study used a between subjects design but used more participants (18 per group) than the current study, and observed an effect of ATD decreasing sensitivity to wins with an effect size of 0.76. The current study, using 27 participants in a within subjects design had 96.7% power to detect such an effect size and thus had adequate power to detect effects observed in Rogers et al (2003). However, whilst these results were not replicated in the current study, this chapter did provide some evidence of a role of 5-HT in decision-making.

5.4.3 Temporal discounting task

Participants discounted future rewards based upon their temporal delay somewhat more than the original study (.084 in the sham condition and .081 in the TRP- condition compared to .033 in Pine et al, 2009), and also displayed increased utility concavities (.027 in the sham condition and .034 in the TRP- condition compared to .0089 in Pine et al, 2009). However surprisingly there was no effect of treatment upon participants' discount factors, utility concavities, or the number of sooner, smaller choices made, which was contrary to expectations based upon the results of previous studies (i.e. Tanaka et al, 2007, Schweighofer et al, 2008). However, it must be noted that these two previous studies did not use the same model to analyse the data as the used here, which was taken from Pine et al (2009);

specifically this form of analysis included examination of participant's utility concavities. As such, this form of analysis has never been tested in participants who have undergone ATD treatment, and so these results present the first evidence of a lack of an effect of this dietary technique upon participants' discount factors and utility concavities as defined by this model. However, the fact that ATD had no effect upon the number of sooner, smaller choices made (which is a 'raw' score of impulsivity on this task) does not support the two previous studies, or indeed research in the animal literature (e.g. Soubrie et al, 1986). Further, practice effects are not typically observed on this task (e.g. Tanaka et al, 2007, Schweighofer et al, 2008). However, the fact that this task is hypothetical (which could have been avoided if participants were actually awarded the money stated on each trial), means that in the second session participants gain a greater sense that this is hypothetical, perhaps magnifying the observed difference in discount rates that has sometimes been shown to occur between real vs hypothetical intertemporal choice in humans (Bickel et al, 2009).

5.4.4 Mood data

Participants did not display differences between scores on the BDI or POMS, which supports previous findings in this area (e.g. Carpenter et al, 1998, Nishizawa et al, 1997, Riedel et al, 1999, Rogers et al, 1999a). However, surprisingly differences were seen in participants' state anxiety scores as shown by the STAI, with such scores increasing on the TRP- day after ingestion of the drink. This is in direct contradiction to previous research, which have shown there to be no effect of ATD upon STAI state measures (e.g. Hood et al, 2010). Further, Altman et al (2010) report no effect of ATD upon (non STAI) anxiety questionnaires, even in participants vulnerable to depression. However, there is much research indicating that 5-HT is involved in anxiety disorders (e.g. Mosienko et al, 2012), and it may be that in the current study ATD led participants to be more prone to increased anxiety due to their knowledge of an upcoming stressful situation (a 2 hour testing battery including a complex pruning paradigm in which financial compensation could be received). However, as no effects of ATD upon state anxiety scores on the STAI have been observed in prior studies, this interpretation is purely speculative and more research would provide a better understanding of these results.

5.4.5 Limitations and further work

Several limitations of this study merit comment. For example, the fact that no significant effects of ATD upon performance on the pruning and temporal discounting paradigms could be due to the decreasing of statistical power due to the need to exclude certain participants during the analysis stage. For instance, 5 participants were excluded from the analyses of the pruning data due to an inability to perform the task. This meant that the power to discover a 'medium' effect size of .50 (Cohen et al, 1988), or even a 'small' effect size of .30 dropped 70% and 32% to 61% and 26% respectively. Four participants also had to be excluded from the temporal discounting analysis (2 due to lost data, and 2 due to poor performance on the 'catch' trials). This reduced the power to 63% and 28% for a 'medium' and 'small' effect size. With regards to the temporal discounting paradigm, the results of Schweighofer et al (2008), who reported a decrease in temporal discounting after ATD treatment, observed such an effect at a size of .87 (large; Cohen et al, 1988). The current study had 99.1% power to detect an effect size of this magnitude, and as such a decrease in power due to the exclusion of the 4 and 5 participants (for the temporal discounting and pruning tasks) cannot be the sole reason for the failure to find any treatment effects on these two tasks. Further, no subjects were excluded from the analysis of the gambling task data, and as described above the results of the original study, in which ATD decreased sensitivity to wins, were not replicated, despite the current study having sufficient power to do so. However, increasing the sample size here would increase the statistical power and improve the chances of observing an effect of ATD upon participants' performance on these decision-making tasks.

Further, the fact that a within subjects design was utilised here is important. Whilst this design was used in order to increase the statistical power (as an increased *N* is required in a between subjects design in order to obtain the same power as would be achieved with a smaller sample size in a within subjects design; Wagenmakers et al, 2012), studies using this design have been shown to discover treatments effects in week 1, but nothing in week 2 (see McCabe et al, 2010 for a discussion). If this was the case here, the repeated measures may have masked the ability of this study to uncover treatments effects. However, this was not the case, as treatment was found to have no effect in both week 1 and week 2.

Finally, whilst participants' LNAA:TRP and tryptophan concentration nmol/ml were significantly reduced following ATD treatment, the fact that participants were allowed no

food from midnight the night before testing until the end of the session at roughly 5:30pm (in order to ensure that participants ingested no tryptophan) may have affected task performance. This fasting (including no tea or coffee) will have increased the effects of fatigue upon participants' performance. However, whilst this rigorous approach to fasting has been used in many studies that have found effects of ATD (e.g. Robinson et al, 2012), others have allowed a small tryptophan-free diet at lunchtime in order to reduce the effects of fatigue upon performance (i.e. Robinson et al, 2013), and this study may have been improved by the introduction of such a diet.

5.4.6 Conclusion

In conclusion, this study administered 3 decision-making tasks to healthy volunteers who had undergone ATD: one which allowed for the observation of pruning, one of which allowed for the examination of participants' gambling behaviours (taken from Rogers et al, 2003), and the final one of which examined the amount to which participants discount the value of a reward based upon its temporal delay. The results of this chapter revealed no effect of ATD upon participants' pruning behaviours or their temporal discounting behaviours. However, the results did reveal a trend towards ATD decreasing participants' sensitivity to different probabilities on the Rogers' (2003) gambling task. As such, the results of this study failed to support the hypotheses from Dayan and Huys (2008) that 5-HT depletion would result in decreased pruning behaviours, and further failed to replicate previous results from Rogers et al (2003) and Schweighofer et al (2008), the former of which showed ATD to decrease sensitivity to different magnitudes of wins, and the latter of which showed ATD to increase the discounting of rewards based upon their temporal delay. Finally, whilst ATD increased participants state anxiety scores, no other effects upon mood were observed, and no significant correlations between performance on any of the above decision-making tasks and mood were observed.

6) PRUNING ABILITIES OF PATIENTS DIAGNOSED WITH MAJOR DEPRESSIVE DISORDER

6.1 Introduction

6.1.1 Evidence for dysfunction of the 5-HT system in depression

MDD is a neuropsychiatric disorder hypothesized to be linked to disruptions in 5-HT transmission primarily to the fact that certain drugs that alleviate depressive symptoms act upon the 5-HT system, such as selective serotonin reuptake inhibitors (SSRIs) e.g. fluoxetine and citalopram (Everett and Toman, 1959, Coppen, 1967, Blier, 2003). Petty et al (1994) were able to show that the classic animal model of depression, learned helplessness, leads to a reduction in 5-HT within the prefrontal cortex, whilst Petty et al (1996) reported that this can be inhibited by pre-treatment of SSRIs. Furthermore, depleting levels of 5-HT via acute tryptophan depletion (ATD) has been shown to cause a transient reappearance of depressive symptomatology in medicated depressed patients in remission (Delgado et al, 1990) with those who have experienced multiple depressive episodes being at increased risk (Booij et al, 2002). Furthermore, Ruhe et al (2007) report that ATD lowers mood in healthy volunteers with a familial risk for depression and MDD patients in remission.

Owens and Nemeroff (1994) report data of post-mortem examinations of depressed patients and suicide victims, which includes reduced cerebrospinal fluid (CSF) concentrations of the main metabolite of 5-HT, 5-HIAA, decreased concentrations of 5-HT and 5-HIAA throughout the cortex, decreased plasma tryptophan (the precursor to 5-HT), increases in 5-HT_2 receptor density and reductions in 5-HTT throughout the cortex. Whilst there have been some contradictory results with regards to post mortem data (e.g. Hrdina et al, 1993), Stockmeier (2003) argues that this may be due in part to flaws such as poor characterisation of medication histories, substance abuse histories, specific psychiatric diagnoses, disorder remission and specific cytoarchitechtonic region co-ordinates, and that on balance the post-mortem literature does support the existence of 5-HT dysfunction in depression. Furthermore, neuroimaging studies have also provided support for the notion that 5-HT dysfunction is involved in depression: Malison (1998) compared levels of the 5-HT transporter (5-HTT) in depressed patients and controls by measuring binding with the radioligand ^{123}I-β-CIT, and

discovered a reduced binding potential in patients, however Dahlstrom et al (2000) report an increase in binding potential in unmedicated depressed children and adolescents. Drevets et al (2007) also report a 26% and 43% decrease in levels of the inhibitory 5-HT_{1A} receptor in the mesiotemporal cortex and raphe nucleus, respectively.

6.1.2 Decision-making abilities of depressed patients

Decision-making depends on the ability to evaluate potential rewarding and punishing consequences of actions in order to select the most appropriate behavioural response (Dayan and Huys, 2008, Huys et al, 2011). Many studies have shown that the 5-HT system may be involved in reward (e.g. Rogers et al, 2003) and punishment processing (Soubriẽ et al, 1986, Cools et al, 2008, Crockett et al, 2009), and it has also been shown that patients diagnosed with depression exhibit maladaptive responses to both rewards and punishments (Eshel and Roiser, 2010). For example, McFarland and Klein (2009) found that depressed participants were significantly less happy than controls when anticipating rewards, despite no differences in mood when anticipating punishments. Furthermore, Pizzagalli (2005) demonstrated that when participants performed a task in which correct responses to one target were three times more likely to be rewarded than those to another target, those with higher scores on the Beck Depression Inventory (BDI) and those with MDD developed a weaker preference for the former target when compared to controls, demonstrating an inability to modulate behaviour according to prior reinforcements. Furthermore, Huys et al (2013) report that this is due to a specific reduction in reward sensitivity, rather than a reduction in sensitivity to prediction errors for reward that determines reward-related learning. With regards to the processing of punishments, Beats et al (1996) demonstrated that on a variety of tasks, depressed patients performed poorly on trials which followed an error (and thus negative feedback) on the previous trial, suggesting a hyper-sensitivity to perceived failure. This tendency also correlates with the severity of depression (Elliott et al, 1996), and Holmes and Pizzagalli (2007) showed that healthy volunteers with high scores on the BDI adjusted their responses significantly less after errors than those with low scores on the Simon (Simon, 1969) and Stroop tasks (Stroop, 1935).

6.1.3 Theory of altered pruning in depression

Dayan and Huys (2008) posit a theory by which serotonin leads to low mood. Here the authors observe that serotonin has been shown to be involved in both the prediction of aversive events (e.g. Deakin, 1983) and behavioural inhibition (Soubrie, 1986), preventing ongoing actions or behaviours in light of these aversive events. As such, the authors argue that serotonin is involved in pruning a tree of potential decisions (specifically, in curtailing the search of such a tree in light of potentially aversive events), and that a decrease in levels of serotonin could result in decreased behavioural inhibition, leading to both decreased pruning and large negative prediction errors. This, in turn, would lead to an increase in negative prediction errors which would occur due to an occurrence of aversive events and more negative consequences, contributing to a pessimistic evaluation of the world, and a consequent decrease in mood. However, Huys et al (2012) found that, contrary to this hypothesis, pruning was associated with *increased* depression scores in a non-clinical sample, and a similar finding was identified in chapter 4 following MDMA administration. Therefore it is important to test the pruning hypothesis of depression in a clinical sample.

6.1.4 Study design and experimental hypotheses

The present study administered the pruning paradigm explained in the methods section (2.4.1), to both control subjects and MDD subjects in order to test Dayan and Huys (2008) hypothesis that the pruning of decision trees is dependent upon normal 5-HT functioning. It was predicted that due to the putative decrease in 5-HT transmission in depression, depressed subjects would exhibit decreased pruning of sub-trees in response to large punishment**.**

6.2 Methods

6.2.1 Participants

Thirty-one control subjects (15 males, mean age 30.4 (10.80) years, range 19-61 years) were recruited via the website advertisements and the University College London Psychology Subject Pool, and thirty MDD patients (15 males, mean age 33.8 (10.44) years, range 19-55 years) were recruited either via the Camden and Islington National Health Service Foundation Trust, Psychological Treatment Services, or via advertisement. This number of participants was chosen due to the fact that a between subjects design with an *N* of 31 controls and 30 patients would give this study 85% to detect a 'large' effect size of 0.8 (Cohen et al, 1988) and 61% power to detect a 'medium effect size (0.5). Patients were initially screened for the below inclusion criteria by telephone, and then in person at the Institute of Cognitive Neuroscience, University College London. The testing session took place at the same location. Inclusion criteria for the control subjects were: no past or present psychiatric disorders (save for a remote (> 6 months) history of substance or alcohol abuse) as assessed by the Mini International Neuropsychiatric Inventory (MINI; Sheehan et al, 1998) and Hamilton Rating Scale for Depression (HAM-D; Hamilton 1960). Inclusion criteria for the depressed participants were: current major depressive episode (assessed as above), no antidepressants within the past month (2 months for fluoxetine), bipolar disorder or psychosis. However, generalized anxiety disorder was allowed, with more than almost two thirds of patients presenting symptoms of anxiety, and the patient group showing significantly increased levels of state and trait anxiety compared to controls (see results section, below). Further, patients were not allowed to have been administered antidepressant medication within the past 6 months, but were permitted to be undertaking a course of behavioural treatment (Cognitive Behavioural Therapy), as was common in those who were recruited via the Camden and Islington Psychological Services. Informed written consent was obtained from all subjects at the beginning of the session, and ethical approval was obtained from the UCL ethics committee. Participants were compensated for their time, which included £10 for participating and up to £20 depending on performance on the task.

6.2.2 Procedure

Participants initially answered questions from the MINI, in order to identify suitability as described above, and then answered questions from the HAM-D, BDI, NEO, STAI and the WTAR Beck Depression Inventory as described in the methods sections 2.1-2.3. Participants were then trained on the behavioural task described in 2.4.1.1 before playing the task for real. The task involved completing 48 episodes of varying length (2-8 moves). Once this was complete, participants were debriefed and paid according to their earnings on the task.

6.2.3 Statistical Analyses and Computational Modelling

As described in the methods section 2.4.1, a set of increasingly complex models was fit to the data using Bayesian model comparison (Huys et al, 2012). After the best fitting model was identified, the parameter estimates from it were extracted for each participant and subjected to classical statistical analyses.

These data were then analysed using Statistical Package for the Social Sciences (SPSS 19, SPSS Inc., Chicago, IL). The main outcome measures of this task were participants' general (γ_G) and specific (γ_S) pruning parameters from the winning model (see methods chapter 2.4.1.1). Both are represented on a scale between 0 and 1, with the former denoting the chance that a participant would continue to search through the tree in general and the latter denoting the chance that a participant would continue to search through the tree specifically when encountering a large punishment. Sensitivities (rhos) to the +140, +20, -20 and -140 transitions were also estimated and compared between the groups (see 2.4.1.1). Independent t tests were performed in order to determine the existence of any differences between the two experimental groups on the task variables, and a set of correlations was conducted in order to determine the existence of any relationship between parameter values and symptoms and personality measures. Due to the large number of correlations there was a need to control for the increased risk of a type 1 error occurring, therefore a threshold of $P<.01$ was adopted.

6.3 Results

Demographic, clinical and mood ratings appear in table 6.1 below.

	Control Subjects	MDD Patients	*t* value	*P* value
N (Male)	31 (15)	30 (15)	-	-
Age (SD)	30.2 (10.7) years	33.3 (10.1) years	1.227	.225
IQ (SD)	108.4 (14.7)	112.2 (10.3)	1.809	.078
BDI	3.00 (4.61)	25.44 (7.48)	13.779	< .001*
HAM-D	1.78 (1.80)	20.28 (4.43)	16.708	< .001*
STAI *state*	10.19 (7.53)	23.44 (11.39)	5.219	< .001*
STAI *trait*	14.71 (8.16)	32.88 (9.74)	7.601	< .001*
NEO *Neuroticism*	17.06 (6.39)	34.80 (7.99)	9.232	< .001*
NEO *Extraversion*	29.81 (6.33)	19.44 (5.72)	6.357	< .001*
NEO *Openness*	29.65 (8.04)	29.63 (5.94)	.119	.906
NEO *Agreeableness*	32.32 (4.99)	29.57 (6.40)	1.524	.133
NEO *Conscientiousness*	33.03 (6.77)	26.68 (7.33)	3.365	.001*
Number of depressive episodes (SD)	-	8.1 (3.82)	-	-
Current episode length (months; SD)	-	3.64 (4.9)	-	-
Age of first episode (years; SD)	-	20.1 (6.3) years	-	-
N of participants attempted suicide	0	7	-	-

Table 6.1. Demographic, clinical and mood data. Asterisk denotes significantly different scores

6.3.1 Computational modeling of decision making task

Eight models were applied to the data in order to explain participants' choices. Figures 6.1 and 6.2 depict the results of these models, each of which were compared using the BIC

method. There was greater evidence (lower BIC) for each of the four models incorporating reward sensitivities to each of the four transitions types (+140, +20, -20, -140) than the corresponding models without such sensitivities. Within these four models the best fitting model (red star in Figure 6.1) included both γ_S and γ_G parameters, suggesting that loss-specific pruning had a robust influence on behaviour. However, including the immediate Pavlovian (state attraction) component decreased the model evidence despite providing a slightly better fit to the data, due to increased model complexity. Therefore parameters were estimated from the "Loss and Pruning" model.

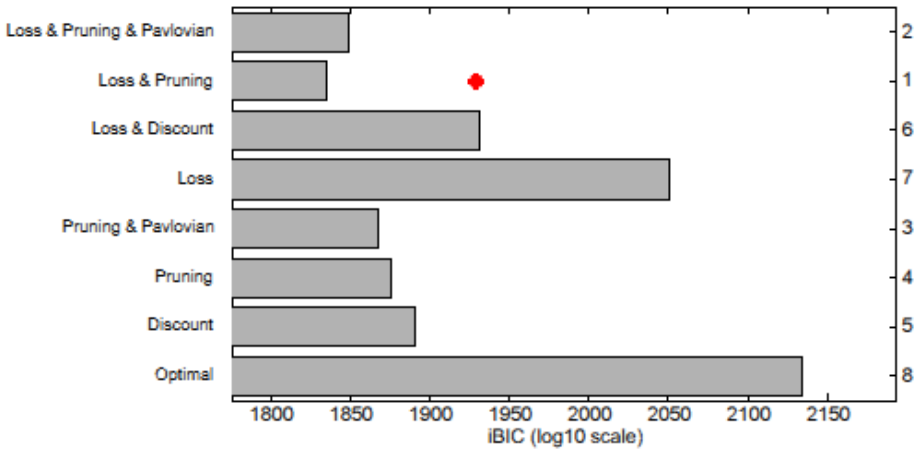


Figure 6.1. Results of BIC model comparison. The top four models are presented in descending order of complexity, as are the bottom four (bottom four without loss aversion (rho) parameters). Model pruning with loss aversion component provided the best fit to the data. Note the decisive evidence in favour of the second most complex model (log10 BIC difference>10)

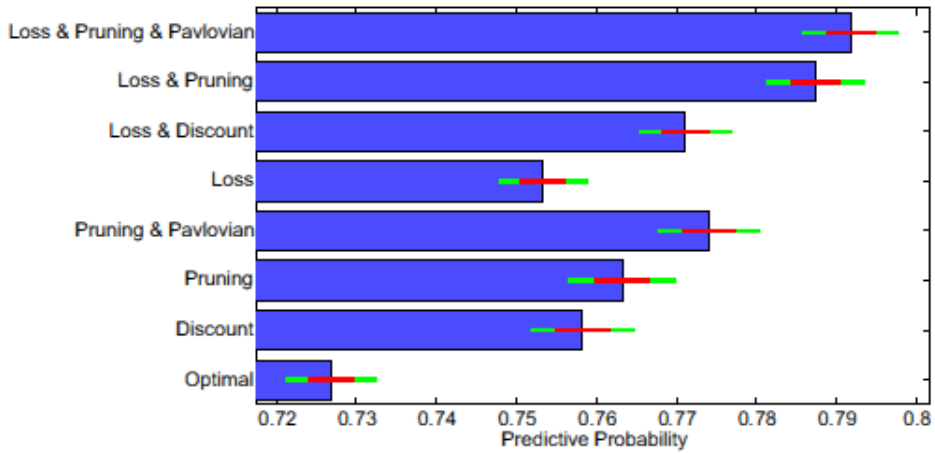


Figure 6.2. Predictive probabilities for all models. All Loss models provide a better prediction of choices than the corresponding models without loss aversion parameters. Although the Loss, Pruning & Pavlovian model has the best predictive probability, the addition of an extra parameter (immediate Pavlovian) is penalized, resulting in a lower BIC

This winning model provided a good fit to the data. Figure 6.3 shows its ability to predict participants' decisions when their next move was the 1st-8th in a sequence. The model performed at a high level across all choices, never predicting less than 80% of choices correctly.

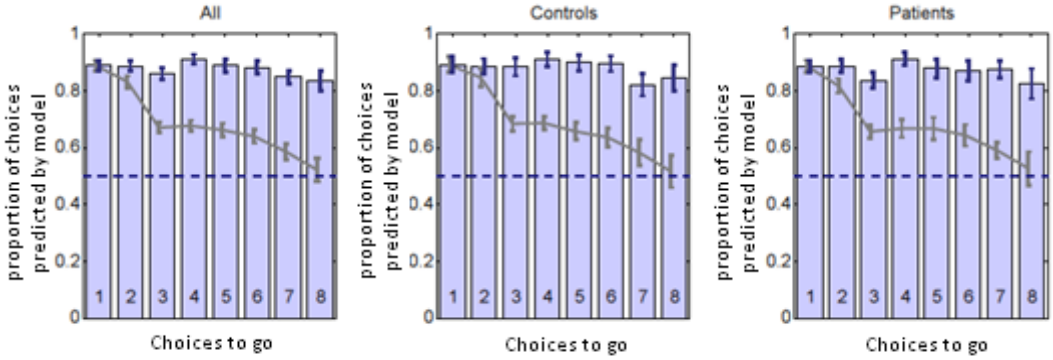


Figure 6.3. Proportion of choices correctly predicted by the winning model 'loss and pruning' across different depths (blue bars). The grey line depicts the simple 'optimal' model (assuming perfect planning), and the blue dashed line denotes chance (50%). Left: patients and controls combined. Middle: controls. Right: patients

Parameters for the winning model are shown in Figure 6.4. It is notable that the reward sensitivity parameters do not provide evidence for loss aversion: the mean (SD) reward

sensitivity across the two groups for the -140 transitions = -2.49 (1.04), the -20 transitions mean = -.008 (.05), the +20 transitions mean = 1.51 (.78) and the +140 transitions mean = 7.31 (3.25), indicating that the +140 transition is more appetitive than the -140 transition is aversive (see Figure 6.4, left). This is also evident in Figure 6.4, middle: participants' estimated valuation of gains vs losses from the parameters of the winning model are depicted. Figure 6.4, right depicts the general and specific pruning for both groups (represented as continuing probabilities). This shows that both groups prune very strongly, as the winning model suggests, with a greater probability of curtailing a tree search in the face of a large negative outcome (γS) relative to other outcomes (γG).

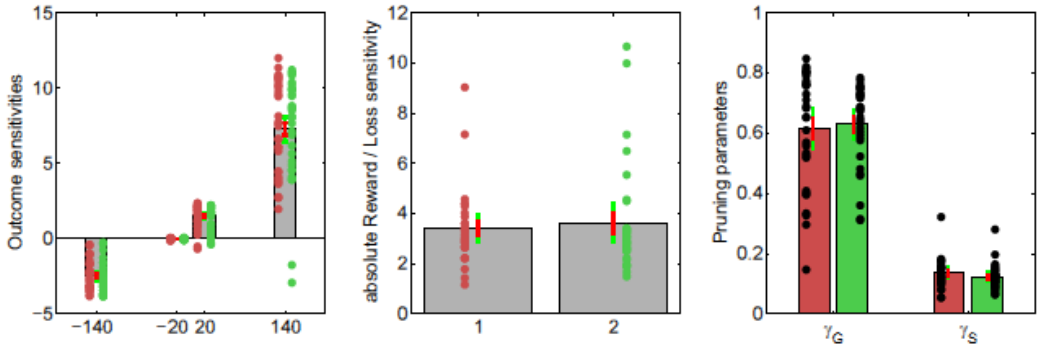


Figure 6.4. Left: Reinforcement sensitivity parameters. Middle: Absolute ratio of large reward (+140) to large loss (-140). Right: Splitting of pruning parameter into general (γ_G) and specific (γ_S) pruning. MDD patients are in red, controls in green. Results here show that both groups prune (as shown by γ_S scores), but that there is no difference between the γ_S scores of each group

6.3.2 Group comparisons

There were no significant differences between groups on either specific (t[59] = 1.123, *P* =.266) or general pruning (t[59] = .322, *P* = .749). There were also no significant differences between groups on reward sensitivities to any of the four transition types: reward sensitivity to -140 transitions- t[59] = .831, *P* = .409; reward sensitivity to -20 transitions - t[59] = .459, *P* = .349; reward sensitivity to +20 transitions - t[59] = -.073, *P* = .942; reward sensitivity to +140 transitions - t[59] = -.105, *P* = .917.

Variable	Control	MDD
γS	.1227 (.042)	.1357 (.049)
γG	.631(.135)	.617(.189)
Reward Sensitivity (-140)	-2.601 (1.115)	-2.378 (.969)
Reward Sensitivity (-20)	-.006 (.047)	-.011 (.046)
Reward Sensitivity (+20)	1.505 (.782)	1.520 (.792)
Reward Sensitivity (+140)	7.252 (3.481)	7.339 (3.046)

Table 6.2. Summary of task variable means and standard deviations in each group. There were no significant differences between groups

6.3.3 Psychometric correlates

MDD patients' pruning parameters (γ 'G' and 'S') and reward sensitivities as defined by the winning model were correlated with their mood and personality scores, as indexed by the NEO, BDI, HAM-D and STAI questionnaires. Results revealed no significant correlations between patients' specific pruning parameter and BDI scores (r=-.083, P=.693), HAM-D scores (r=-.036, P=.853), Neuroticism scores (r=.082, P=.666), Extraversion scores (r=-.066, P=.730), Openness scores (r=.024, P=.902), Agreeableness scores (r=.202, P=.284), Conscientiousness scores (r=-.111, P=.559), state anxiety scores (r=-.120, P=.528) or trait anxiety scores (r=.164, P=.368).

6.4 Discussion

This study used a between subjects design in order to investigate the impact of depression on pruning. Two groups were included in the study: 31 controls and 30 depressed patients, who were all tested on the pruning paradigm described in the methods section 2.4.1.1. The results revealed that the model 'Loss and Pruning' was a good fit to the data, explaining more than 80% of participants' choices. However, there were no significant differences between groups in terms of performance on the pruning task, and no significant correlations between mood or personality scores and pruning. These results will be discussed and considered in light of both previous work by Huys et al (2012) and Dayan and Huys (2008).

6.4.1 Pruning task

Results of the Bayesian model comparison revealed that the model 'Loss and Pruning' best explained participants' choices on the task, which suggested that loss-specific pruning had a robust influence on behaviour. Furthermore, examining general and specific pruning scores from this winning model did not provide evidence for a difference between groups in either their general planning abilities or their pruning behaviours due to a large loss, respectively. Furthermore, patients and controls did not differ in their reward sensitivities to any of the four transition types, with both groups finding the rewarding transitions more appetitive than they found the punishing transitions aversive, meaning that loss aversion was not observed. The fact that no differences were observed in sensitivities to both rewards and punishments was surprising, as many previous studies have shown the opposite effect (see Eshel and Roiser, 2010 for a review). As expected, participants did differ on personality and mood scores, and the results of the NEO are in line with previous reports of increased neuroticism, decreased extraversion and decreased conscientiousness in depression (Klein et al, 2011).

A number of studies have examined decision-making in depressed patients (e.g. Beats et al, 1996, Elliot et al, 1996). However, these studies did not examine patients' performances in multi-step decisions, and this is the first study to test the pruning abilities of a depressed population. Huys et al (2012) demonstrated that when faced with a series of actions, healthy volunteers prune away potential actions as soon as they encountered a large loss. The current study was able to replicate these results and provides clear evidence of pruning. However, the winning model from Huys et al (2012) included a 'Pavlovian' parameter, indicating that

participants had a reflexive attraction to certain states (as opposed to transitions), and aversion to others. The Pavlovian parameter did not improve model parsimony in this study however, meaning that considering participants' valuation of states (boxes) rather than considering just their sensitivity to the four transition types did not make for a better model fit. Furthermore, Huys et al (2012) also reported a correlation between sub-clinical mood disturbances and pruning behaviours in their healthy volunteers. The authors explain this latter result in a framework that links mood disturbances with abnormalities in 5-HT functioning (i.e. Everett and Toman, 1959, Coppen 1967) and 5-HT functioning with pruning (based on Dayan and Huys (2008) theoretical work), such that depressed patients (with putative serotonergic abnormalities) should display decreases in the curtailment of potential decision trees in response to a large loss. The present study provided no evidence for this hypothesis.

There are a number of possible explanations as to our failure to observe the existence of differences in the pruning behaviours between healthy volunteers and depressed participants. For example, it may be that whilst 5-HT is involved in the pruning of decision trees, the depressed patients in this study suffer from abnormalities in a range of neurotransmitter systems, rather than just serotonin (Di Chiara et al, 1999). These transmitter systems, such as dopamine, have been shown to influence sensitivity to rewards and punishments (e.g. Cools, Nakamura and Daw, 2011), which may exert an influence upon loss-specific pruning in these subjects. Further, the failure to find effects may simply be due to a lack of suitable power: with only 31% power to detect a 'small' effect size of 0.3 (Cohen et al, 1998) such small group effects may have been missed, resulting in a type 2 error.

6.4.2 Limitations

Due to difficulties with recruitment, only two-thirds of these patients were recruited via the Camden and Islington National Health Service Foundation Trust, Psychological Therapy Services. The remaining patients were recruited via website advertisements, meaning that their psychiatric profile had not been examined by a professional within the National Health Service. However, considering this, all depressed participants included in the study did satisfy criteria for a current major depressive episode, yet without documented anti-depressant use histories for these subjects it is difficult to make conclusions regarding the link between 5-HT and pruning in these subjects.

6.4.3 Further work

In order to assess the evidence for the hypothesis set out in Dayan and Huys (2008) and Huys et al (2012), this pruning task could be administered to (as well as the serotonin-depleted individuals reported in this thesis) participants who have increased levels of serotonin (through acute tryptophan loading and SSRI administration). This would hopefully allow for a better understanding of serotonin's role in pruning. Further, whilst much work indicates that 5-HT may be involved in pruning decision-trees (i.e. Deakin et al, 1983, Soubrie et al, 1986, Dayan and Huys, 2008), other monoamine systems have been shown to be involved in reward and punishment processing (e.g. van der Schaaf, 2012), and observing the role of other neurotransmitter systems may shed light on the neurochemical basis of pruning behaviours. Further, depressed patients have been shown to have altered levels of various receptor subtypes linked to various neurotransmitter systems (i.e. increased 5-HT_2 receptors in the PFC; McKeith et al, 1987 and increased D2/D3 receptors in the amygdaloid complex; Klimek et al, 2002). As such, it may prove fruitful to examine the expression of certain genes linked to various neurotransmitter systems (i.e. HTR1A, HTR2C, DRD2 genes which code for the 5-HT_{1A}, 5-HT_{2C} and dopamine-D2 receptors, respectively) in these groups in order to shed further light on the link between MDD, pruning and neurotransmitter systems. Finally, examination of pruning in different psychiatric groups, such as patients with anxiety, may provide useful insights into whether decision-making is altered in different psychiatric disorders.

6.4.3 Conclusion

We observed no differences between depressed patients and healthy controls on general pruning, specific pruning and reward sensitivities to both small and large rewards and punishments. Therefore, this study failed to provide support for Dayan and Huys (2008) theory that depressed patients would display poorer pruning abilities due to aberrant 5-HT functioning.

7) GENERAL DISCUSSION

This discussion will provide an integrative summary of the experiments presented in chapters 3-6. A brief overview of the main results of each study will be given first, after which a comparison of the effects of 5-HT_{1A} binding, acute tryptophan depletion and subacute MDMA administration upon reward and punishment processing will be presented, along with how these effects relate to those observed in the depressed sample from chapter 6. The results of these studies will be considered in light of how much they may help improve our understanding of the relationship between serotonin, decision-making and mood. Finally, the limitations of these studies and possible directions for future research will be outlined, with the aim of improving further our understanding of the pathophysiology and potential treatments of MDD.

7.1 Summary of experimental investigations

7.1.1 Study 1: Decision-making and the 5-HT_{1A} receptor: a positron emission tomography study

The aim of this study was to build upon studies that have shown a role for general 5-HT transmission in reward and punishment processing (i.e. Rogers et al, 2003, Schweighofer et al, 2008, Tanaka et al, 2007) by observing the relationship between 5-HT_{1A} binding and decision-making behaviour in a group of healthy volunteers. This was done by initially administering the novel ligand CUMI before a placebo in order to observe baseline 5-HT_{1A} availability, and before intravenous administration of 10mg citalopram in order to observe the change in 5-HT_{1A} receptor availability (from which an index of 5-HT release is inferred). The results of the citalopram challenge were surprising in that administration of this SSRI led to an apparent decrease in 5-HT release throughout the cortex. In order to test the relationship between the 5-HT_{1A} receptor and decision-making, performance on 3 behavioural tasks was correlated with the above indices of 5-HT_{1A} receptor availability. Consistent with prior results, participants exhibited robust aversive-based pruning, as indicated by a substantial increase in evidence for the computational models in which the pruning parameter was incorporated. Whilst no significant correlations between 5-HT_{1A} binding or 5-HT release and the pruning parameter were observed, there were significant correlations between sensitivity

to losses and the change in 5-HT_{1A} availability (due to citalopram infusion) in the right nucleus accumbens and between sensitivity to probability and baseline 5-HT_{1A} availability in the right hippocampal complex. Further, a significant correlation between participants' discount factor on the temporal discounting task and baseline 5-HT_{1A} receptor availability in the left hippocampal complex was also observed. The results of this study suggest that a relationship exists between both sensitivity to the probability of winning when gambling and temporal discounting, and baseline 5-HT_{1A} receptor availability in the hippocampus. Further, these results also suggest that there exists a relationship between sensitivity to losses and the change in 5-HT_{1A} receptor availability in the striatum following administration of an SSRI.

7.1.2 Study 2: Decision-making 3 days after administration of 3, 4-methylenedioxymethamphetamine (MDMA)

The aim of this study was to test the theory of Dayan and Huys (2008) that 5-HT is involved in the pruning of decision trees. This was done by administering MDMA on day 1 to a group of healthy volunteers, and then testing them on the pruning paradigm 3 days later in order to assess the effect of putatively decreased levels of 5-HT and low mood upon pruning behaviours. The results suggested that participants' displayed decreased pruning (as shown by significantly decreased scores on the pruning variable of the proportion best remaining and a trend towards a significant difference in the difference estimate) after MDMA compared to after placebo. This MDMA-induced decrease in pruning behaviours was specific, rather than the result of a more general cognitive impairment, as shown by the fact that participants did not significantly differ on their performance on the NLLO trials, from which an index of planning is obtained, between treatments.

Despite the results of Curran and Travill (1997) suggesting that MDMA self-administration leads to decreased mood 3-5 days later, no differences in mood in this study were observed. However, the results did reveal a negative correlation between mood and pruning: those who displayed increased scores on the BDI and the antagonism scale of the POMS (increased negative affect) following MDMA also displayed increased proportion best remaining scores (denoting increased pruning). However, the direction of this relationship is in direct opposition to that predicted by Dayan and Huys (2008), who proposed that decreased 5-HT causes decreased mood by decreasing pruning. Nonetheless, the fact that MDMA still

decreased participants' pruning behaviours provided some support for Dayan and Huys' theory.

7.1.3 Study 3: The influence of acute tryptophan depletion on the decision-making abilities of healthy volunteers

There were 3 main aims of this study. The main aim was to assess the role of 5-HT in pruning by depleting 5-HT by the dietary method of ATD. Secondly, this study aimed to replicate the results of ATD upon win sensitivity during gambling observed in Rogers et al (2003). Thirdly, it aimed to replicate the results of Schweighofer et al (2008) and Tanaka et al (2007), in which it was found that ATD increased discounting of rewards based upon their temporal delay, by administering both the temporal discounting paradigm and model from Pine et al (2009) which permitted the examination of participants' discount factors and utility concavities. Mood and psychometric data were also collected in order to observe any relationships between task performance and mood. Whilst ATD was shown to successfully deplete participants' plasma tryptophan levels, treatment had no effect on the main pruning variables (although there was a trend towards treatment reducing reaction times on NLLO trials and increasing them on LLO trials). Further, on the gambling task there was a trend towards ATD decreasing participants' choosing of the experimental gamble overall, whilst also affecting participants' sensitivity to probability also at trend level. This latter result was driven by a reduction in choices of high probability gambles. The results also revealed no effect of ATD treatment on participants' performance on the temporal discounting paradigm. Finally, ATD was found to increase participants' STAI state anxiety scores, though no relationships between mood change and decision-making behaviour were detected. The results of this study do not support the theory of Dayan and Huys (2008), and nor did they replicate prior reports of decreased win sensitivity and increase delay discounting following ATD (Rogers et al, 2003, Schweighofer et al, 2008). However, they are consistent with one previous report that ATD decreases choices of high probability gambles (Rogers et al, 1999), indicating that 5-HT may indeed play a role in participants' use of information pertaining to probabilities when making choices on a gambling paradigm.

7.1.4 Study 4: Pruning abilities of patients diagnosed with major depressive disorder

Whilst it is important to understand the importance of 5-HT in pruning and decision-making as a whole, Dayan and Huys' original theory attempted to explain how altered decision-making can lead to low mood and ultimately depression. As such, the main aim of this final study was to examine the performance of patients diagnosed with MDD on the pruning paradigm. Using a Bayesian model comparison approach, a set of computational models was applied to the data, and it was found that both groups displayed aversive-based pruning as predicted. Surprisingly, it was also found that participants found the rewarding transitions more appetitive than they found the punishing transitions aversive. However, when comparing the two groups, there was no difference between MDD patients and controls in pruning behaviours. Further, no significant correlations were found between participants' pruning (specific pruning parameter from the most parsimonious computational model) and scores from any of the psychometric questionnaires. As such, these results fail to support the theory of Dayan and Huys (2008) that low mood is driven by low pruning. This may be due to inaccuracies in the theory, or may simply be due to an inability of the task to elucidate differences between groups in pruning behaviours. For example, there may have been discrepancies in the ability of financial rewards and losses to motivate behaviour to the same extent in both groups. If this latter case were to be true, then examining performance on this task with rewards and losses that are not financial in nature could help our understanding of pruning in depression.

7.2 Comparison of the effects of 5-HT_{1A} receptor binding, subacute MDMA administration, acute tryptophan depletion, and major depressive disorder upon decision-making and mood

Each of the four studies in this thesis used a different method in order to assess the effect of altered 5-HT functioning upon decision-making and mood. The below discussion will compare and contrast the results of these studies, considering how they compare to both the previous findings in the depression literature and the final experimental chapter in this thesis which tested pruning in MDD patients. In doing so, it will also consider any similarities or differences between the effects of these 5-HT manipulation techniques and the depressed state.

7.2.1 Pruning

It must be first noted that both the pruning paradigm and the statistical analysis used in chapters 4 and 5 differed from the task and analysis employed in chapters 3 and 6; the task administered in chapters 4 and 5 contained a trials in which participants only had 9 seconds to look at the matrix, and 2.5 seconds to enter their moves (as opposed to the task in chapters in 3 and 6 in which there was no timing element), contained large loss transitions of -70p (as opposed to the -140p in the task in chapters 3 and 6), and was analysed by frequentist statistics only (unlike the task of chapters 3 and 6 which was analysed using Bayesian computational modelling). As such, the former will be considered separately before the latter.

7.2.1.1 Pruning; Chapters 4 and 5 (timed task, without modelling)

The results of chapter 4 were particularly striking: administration of MDMA 3 days prior to performance of the task significantly reduced the proportion best remaining index of pruning. Further, a trend towards a significant decrease in the other index of pruning, the difference estimate, was also observed 3 days following MDMA administration. In contrast, there was no effect of ATD treatment upon either pruning variable in chapter 5 (although there was a trend towards a decrease in reaction times on NLLO trials and an increase in reaction times on LLO trials). One potential explanation for the discrepancy between results on this task in chapters 4 and 5 could be the potency of each method of 5-HT depletion: MDMA has been shown to target the 5-HT system both *in vitro* (e.g. Rudnick and Wall, 1992) and *in vivo (*e.g. Kish et al, 2010), and has been shown to cause long term damage to the axons of 5-HT neurons after repeated exposure (O'Shea et al, 2006). Further, it has been shown *in vivo* to have both acute and subacute effects, increasing 5-HT acutely and decreasing 5-HT subacutely by up to 30%, whilst also reducing tryptophan hydroxylase by up to 75% in the rat brain (e.g. Stone et al, 1986). In contrast, whilst ATD has been shown in humans to decrease plasma tryptophan and levels of 5-HIAA in the CSF (e.g. Carpenter et al, 1998, Moore et al, 2000), these effects last roughly only a few hours (e.g. Robinson et al, 2013, Crockett et al, 2012, Roiser et al, 2006). Further, the potency of these two contrasting techniques can be inferred by their respective effects upon mood: MDMA has been to shown to increase mood at the time of administration but decrease it 3-5 days later (Curran and Travill, 1997), whilst ATD has been shown to neither decrease nor increase mood in healthy volunteers (e.g. Carpenter et al, 1998, Nishizawa et al, 1997, Riedel et al, 1999, Rogers et al, 1999a),

although it has been shown to temporarily reinstate depressive symptoms in those who have recovered from depression via the use of SSRIs (O'Reardon et al, 2004). As such, it may be that the subacute effects of MDMA administration upon pruning are due to the potency of this technique upon the 5-HT system.

The mood effects of both of these studies were also notable: MDMA exposure did not lower mood significantly, which is in direct contradiction to the findings of Curran and Travill (1997) above, whilst ATD did increase participants' state anxiety scores, which is in direct contradiction to the results of previous studies using this technique (e.g. Riedel, 2004). The reason for this former result may be due to the fact that MDMA was both synthesized and administered in a clinical setting, without the participants ingesting any other drug (illicit or otherwise) or being sleep-deprived, unlike the participants from the Curran and Travill (1997) study. Further, a positive correlation between negative affect (increased antagonism and BDI scores) and pruning (as shown by the difference estimate) 3 days after MDMA administration was observed in chapter 4, such that those who displayed increased negative affect also displayed increased pruning. Whilst this result contradicts the original theory put forward by Dayan and Huys (2008) that low mood is associated with decreased pruning, it does replicate the results of Huys et al (2012) in which a positive correlation between greater negative affect (as shown by the BDI) and high pruning was observed. However, no such correlation was observed after ATD, which may be due to the fact that ATD failed to affect pruning behaviours, meaning that analysis of the mood data in chapter 5 cannot be said to support either Dayan and Huys (2008) or Huys et al (2012).

7.2.1.2 Pruning: chapters 3 and 6 (untimed task, incorporating computational modelling

The results of the computational modelling were relatively consistent between these two chapters: modelling participants' choices revealed a reliable influence of loss-specific pruning. However, participants in chapter 3 displayed a further immediate Pavlovian attraction to the state from which a large rewarding transition could be completed (even though simply reaching this state did not lead to said large reward), whereas participants in chapter 6 did not display this behaviour.

Analysis of the pruning parameters in these two chapters however provided little support for Dayan and Huys' (2008) theory for a link between 5-HT, pruning and mood. The results of

chapter 3 did not support a relationship between either baseline 5-HT_{1A} availability or the change in 5-HT_{1A} availability due to citalopram infusion and pruning. Previous research has indicated the 5-HT_{1A} receptor to be an important receptor in both cognition and the aetiology of depression: Deakin and Graeff (1991) for example proposed that one of the two main 5-HT systems in the brain projects from the median raphe nucleus (MRN) to the hippocampus, and mediates responses to life events in which loss is experienced via the 5-HT_{1A} receptor. Further, rodent studies have shown dose-dependent effects of 5-HT_{1A} receptor agonists upon the rewarding effects of drug-self administration (i.e. Parsons et al, 1998, Peltier and Schenk, 1993), indicating that this receptor may be involved in the rewarding consequences experienced during the performance of decision-making tasks. However, whilst the results of this study do not prove that there is no link between pruning and 5-HT in general (especially since the study included only a modest number of participants), they do not support the hypothesis that pruning is related to transmission at the 5-HT_{1A} receptor. Due to the fact that Deakin and Graeff (1991) argue that the 2nd main 5-HT system in the brain projects from the dorsal raphe nucleus to the amygdala, and mediates adaptive responses to potentially dangerous stimuli via the 5-HT_{2C} receptor, examining the availability of 5-HT_2 receptors (and indeed other subtypes) may shed light on the molecular basis of pruning (Shelton et al, 2009).

The results of chapter 6 also failed to support the Dayan and Huys hypothesis, finding no significant differences between depressed and non-depressed in terms of performance on the pruning task. Whilst the results of studies examining reward and punishment processing in depression have found differences between patients and controls (i.e. Eshel and Roiser, 2010), this study was not able to find any differences on our multi-step decision-making task. In fact, no relationships between participants' pruning parameters and mood were found at all. This was unexpected due to the results of Huys et al (2012) who reported a correlation between mood and pruning such that those who displayed increased negative affect also displayed increased pruning. The authors here note that this is contrary to the hypothesis outlined by Dayan and Huys (2008), and explain this relationship was observed because depressed individuals exhibit decision-making that is more dependent upon the 5-HT system, and that those with slightly increased sub-clinical depression scores would also display excessive pruning due to a risk for depression, whilst those who were experiencing a current depressive episode would exhibit decrease pruning. However, the present study, by reporting no effect of depression upon pruning, fails to support this hypothesis.

7.2.2 Gambling task (chapters 3 and 5)

Analysis of the gambling task in chapter 3 produced some interesting results. These results indicate the existence of a relationship between decision-making and 5-HT transmission. These results are consistent with the results of Rogers et al (1999b) that suggest a role for 5-HT in sensitivity to the probability of winning; here the authors showed that participants made more choices of less probable gain outcomes on the CGT after ATD. However, the results of that study were contradicted by those of Talbot et al (2006) who reported, using the same task, an *increased* choosing of more probable gain outcomes after ATD. The authors in the latter study speculate that these conflicting results may be due to factors such as unmeasured intrinsic trait characteristics such as aggression, which Bjork et al (2000) argue can lead to directionally opposite affects after ATD treatment. However, this interpretation remains speculative. Whilst the results of chapter 3 indicate a role for 5-HT_{1A} transmission in reward processing, the task administered here was different to that administered in the above two studies, and more research is needed in order to fully understand the relationship between 5-HT and the processing of rewards and punishments during decision-making.

The results of chapter 3 are also interesting as they indicate the existence of a relationship between the processing of rewards and punishments on a gambling task and activity at a specific 5-HT receptor subtype, with baseline 5-HT_{1A} receptor availability within the right hippocampus correlating with participants' sensitivity to probability, and the change in 5-HT_{1A} receptor availability in the right nucleus accumbens correlating with participants' sensitivity to loss. This supports previous research that has highlighted a role for these structures in reward and punishment processing. For example, Klein et al (2007) report dynamically changing functional connectivity patterns between the hippocampus and ventral striatum on a positive and negative feedback-based learning task, and Cohen et al (2008) showed that the microstructural properties of white matter tracts connecting the amygdala, hippocampus and ventral striatum predicted functional connectivity patterns observed following both positive and negative feedback on a reversal learning task. Whilst the results of these two studies indicate a link between the hippocampus and reward processing, the tasks used in these studies were feedback-based learning tasks, rather than a gambling task that included known probabilities of financial gains and losses, as employed in chapter 3. However, Camara et al (2008) did examine hippocampal responses and performance on such a task. Here, the authors administered a gambling task (designed by Gehring and Willoughby,

2002) in which participants were faced with unexpectedly high gains and losses whilst inside an MRI scanner. The results of the categorical analyses revealed that monetary gains and losses resulted in activation bilaterally in the ventral striatum, while functional connectivity analyses with the seed region in the ventral striatum showed enhanced connectivity with the striatum during the processing of similar responses to gains in the hippocampus bilaterally. Taken together, these prior studies and the results of chapter 3 indicate a potentially important role for 5-HT transmission in the hippocampus in decision-making, and warrant further investigation.

However, it must be noted that due to the results of the citalopram challenge being in the opposite direction to that hypothesised, the relationship between sensitivity to loss and the change in 5-HT_{1A} availability due to citalopram infusion is difficult to interpret. Nevertheless, the positive correlation does support the findings of Schmitz et al (2009) which revealed a role for the 5-HT_{1A} receptor in punishment processing by demonstrating that a 5-HT_{1A} C(-1019)G polymorphism-linked increase in 5-HT_{1A} availability is correlated with a greater sensitivity to punishments.

The results of chapter 5 revealed no significant effect of ATD upon sensitivity to wins or losses, although a trend effect was observed upon sensitivity to probability, which in part supports the results of Rogers et al (1999b) (though not Talbot et al,2006) who showed an effect of ATD upon the choosing of probabilistic gain outcomes. Further, the fact that ATD reduced participant's sensitivity to high probabilities further supports the results of studies showing ATD to have an effect upon reward and punishment processing (e.g. Crocket et al, 2012, Robinson et al, 2012).

However, these results do fail to replicate those of Rogers et al (2003) in which ATD was found to reduce healthy volunteers' sensitivity to wins, although this is to our knowledge the first attempt to replicate the results of Rogers et al (2003). One suggestion to improve this study could be to increase the statistical power to detect smaller effect sizes by increasing the number of participants. However, the effect size of ATD upon sensitivity to wins observed in Rogers et al (2003) was 0.74 (in the region of a 'large' effect size as defined by Cohen et al, 1988), and the current study had 99.9% power to detect such an effect size due to the utilisation of a within subjects design. As such, the failure to replicate the findings of this study is unlikely to lie with the statistical power of the study in chapter 5.

7.2.3 Temporal discounting task (chapters 3 and 5)

The results of the temporal discounting paradigm in chapter 3 were very interesting, with participants' discount factor correlating negatively with their 5-HT_{1A} availability in the left hippocampal complex. These results further suggest that 5-HT is involved in the discounting of future rewards based upon their temporal delay, which supports Schweighofer et al (2008) who reported an increase in participants' discounting after ATD.

These findings support the results of previous research which show a role for the hippocampus in temporal discounting. For example, Mobini et al (2000) reported that rats whose 5-HT systems had been destroyed with 5,7-dihydroxytryptamine became more impulsive and exhibited increased choosing of smaller, sooner rewards in a temporal discounting paradigm, which correlated with a decrease of 5-HT in the hippocampus. Further, studies are beginning to show a role for an involvement of the hippocampus in episodic representations which can affect temporal discounting. For example, Schacter and Addis (2009) have shown that the hippocampus and parahippocampal gyrus play a crucial role in the formation of past, present and future episodic representations, and Peters and Buchel (2010) were able to show that the addition of episodic 'tags' that involved the presentation of relevant future episodes (i.e. vacation in Paris) to a temporal discounting paradigm, led to both a decrease in participants' discount rates and a coupling between the ACC and hippocampus bilaterally. As such the results of chapter 3 further support a role for the hippocampal complex in temporal discounting, indicating that this relationship may be mediated in part by 5-HT_{1A} transmission.

However, the results of this chapter do not support the results of Tanaka et al (2007) who reported a role for the striatum in this relationship. Here the authors administered a temporal discounting paradigm to participants inside an MRI scanner. The results of this study revealed that BOLD responses in the ventral striatum were related to reward prediction at shorter time scales which was stronger after ATD, and that such responses within the dorsal striatum were related to reward prediction at longer time scales, which was stronger after acute tryptophan loading. Whilst the results of chapter 3 do not support a role for 5-HT_{1A} receptor availability within the striatum in temporal discounting, this may be due in part to the very low binding values within the caudate nucleus (which is why the striatal mask

applied to correct for multiple comparisons in chapter 3 did not contain the caudate; for binding values see Selvaraj et al, 2012).

Contrary to chapter 3, the results of the temporal discounting paradigm in chapter 5 do not support a role for 5-HT in temporal discounting, thus failing to support studies in the animal literature (e.g. Soubrie, 1986) and the results of Schweighofer et al (2008) and Tanaka et al (2007). Once again one suggestion to improve this study could be to increase the statistical power by increasing the number of participants. The effect size seen in Schweighofer et al (2008) was 0.73, and the present study had 92% power to detect such an effect size. As such, there is a small possibility that the failure to replicate the results seen in Schweighofer et al (2008) here represents a false negative.

It should be noted that the results of this task are supported by those of Crean et al (2002) who reported no effect of ATD upon temporal discounting in both males with and without a family history of alcoholism. Further, this study is the first examination of the role of 5-HT in temporal discounting using the model of Pine et al (2009), which includes the discount factor from Schweighofer et al, in addition to an extra parameter, 'utility concavity'. This latter parameter examines the extent to which participants differ on how much value they place upon an amount (i.e. £1) in a total (i.e. £100), with the concavity meaning £1 is worth more in a £2 total than it is in a £100 total. Including this parameter could have led this model to capture participants' choices differently compared to that of Schweighofer et al (2008), Crean et al (2002) and Tanaka et al (2007), all of which used models that included discount factors without the addition of a utility concavity parameter in order to examine participants' discounting behaviours.

Further, whilst there are many animal studies that have examined the variable of the number of sooner choices chosen in a temporal discounting paradigm (e.g. Soubrie et al, 1986), there are no reported results of an examination of this measure in humans who have undergone ATD treatment. Whilst it could be argued that this measure is a 'rawer' measurement of impulsivity, unconfounded by model deficiencies, no treatment effects were observed, once again failing to support a role for 5-HT in this form of decision-making. This is supported by human work performed by Rogers et al (1999b) and Talbot et al (2006) who failed to observe an effect of ATD upon impulsive responding (assessed by the difference between the 'ascending' and 'descending conditions' on the CGT. As such, whilst the results of this task

in chapter 5 do not provide support for those of chapter 3 and other published studies, perhaps more consistent models of impulsive responding on this task need to be developed in order to better understand the role of 5-HT in this form of decision-making.

7.3 Limitations of the studies within this thesis and directions for future research

Whilst the above studies suggest that there may be some role for 5-HT in disrupted reward and punishment processing in depression, there exist certain limitations with respect to the conclusions that may be drawn. These limitations will be discussed below, as will directions for future research that aim to answer the questions arising from these studies.

7.3.1 Chapter 3 limitations and aims for future research

This study used a novel ligand (^{11}C-CUMI-101) in order to assess the relationship between 5-HT release and reward and punishment processing. However, a particular difficulty in interpreting the meaning of these data exists due to the fact that whilst CUMI is a competitive agonist of the 5-HT_{1A} receptor (thus allowing for the observing of 5-HT release), both receptor density and levels of extracellular 5-HT contribute to the measured CUMI signal, with more receptors increasing the signal, and more extracellular 5-HT decreasing it. As such, whilst this research does allow us to make predictions about the effects of increasing or decreasing the former or the latter upon decision making, it is difficult to know the extent to which sensitivity to probability and participants' discount factors were correlated with receptor density *per se* or levels of extracellular 5-HT at baseline. This question could be addressed using a non-competitive ligand such as [^{11}C]WAY-100635 which would allow for a more definitive conclusion to be made regarding the nature of the PET signal.

An obvious extension to this study would be to have participants perform the above decision-making tasks whilst under the influence of citalopram. This would allow for an observation of the effects of baseline, and in particular, the change in 5-HT_{1A} receptor availability upon task performance following 5-HT manipulation. The autoreceptor hypothesis argues that SSRIs exert their antidepressant effects by initially inhibiting the 5-HT system via agonism of the 5-HT_{1A} autoreceptors in the dorsal raphe nucleus, with these autoreceptors in turn becoming desensitized over a number of weeks until they cease their inhibiting effects upon downstream 5-HT release, which allows for an increase in cortical 5-HT and decrease in

depressive symptoms (Blier, 2003). Further, due to the fact that it is difficult to confirm that such correlations are causative, it would be informative to test this theory using the CUMI ligand again whilst both depressed patients and healthy volunteers performed the above decision-making tasks in the PET scanner so that any changes in performance of these tasks could be linked to changes in 5-HT_{1A} availability due to SSRI treatment. Such an approach might help shed light on the decision-making differences in depression and the effects SSRI treatment.

Further, perhaps correcting the number of correlations for multiple comparisons may improve confidence in the significance of such results. However, such corrections were not performed due to the exploratory nature of this study in order to ensure that any significant correlations with 5-HT_{1A} availability were observed.

Finally, in the study in chapter 3 participants with greater 5-HT_{1A} availability in the hippocampus were less likely to discount the value of rewards that were available further in the future. Although the hippocampus is typically associated with episodic memory processing and contextual learning, as described above Peters and Buchel (2010) were able to show that the addition of episodic tags to a temporal discounting paradigm decreased participants' discount rates which was associated with increased coupling between the ACC and the hippocampus bilaterally. As such, it could be very interesting to extend the current study using a similar paradigm in order to understand whether such episodic tags mediate this relationship between temporal discounting and the 5-HT_{1A} receptor availability.

7.3.2 Chapter 4 limitations and directions for future research

Whilst the results of chapter 4 were interesting in that they indicated that MDMA administration affected participants' pruning of decision trees, there are also some limitations to this study. For example, the interpretation of these results is that participants who were administered MDMA 3 days prior to testing had depleted levels of 5-HT, which affected their performance on the task. Whilst this assumption is based upon previous research in both animals (i.e. Stone et al, 1986) and humans (i.e. Kish et al, 2000, 2010), there was no index of 5-HT functioning in the present sample. As such, obtaining markers of 5-HT function, such as cerebrospinal fluid 5-HT metabolite levels, or indeed levels of extracellular 5-HT in the

brain (as shown by CUMI) would increase confidence in the interpretation that these results reflect a link between 5-HT and pruning.

A further limitation to this study is that it only examined the subacute effects of MDMA administration, and an examination of the effect of acute MDMA-induced increase in 5-HT transmission upon pruning behaviours would be informative. However, this would bring with it its own difficulties, including the fact that participants may find it difficult to concentrate on and adequately perform the complex pruning paradigm while under the influence of MDMA; in all the experiments presented in this thesis there have been participants who have failed to adhere to task demands and have simply found this paradigm too complex to successfully complete even when not under the influence of such a powerful psychoactive stimulant.

Furthermore, the reason the subacute effects of MDMA were examined precisely 3 days after administration was based upon the findings of Curran and Travill (1997), who reported progressively decreasing mood 2-5 days after MDMA self-administration. However, the results of chapter 4 revealed no difference in mood post MDMA compared to post-placebo. As mentioned above, this could be due to a variety of reasons, including a lack of multi-drug ingestion and sleep deprivation in the current study. Further, it may be that the time of administration of MDMA (9am) affected participants' subacute mood in this study: participants may have experienced low subacute mood, but at an earlier time point than the actual subacute session due to the earlier time point on the administration day compared to Curran and Travill (1997), in which participants self-administered on a Saturday night. As such, it would be interesting to test participants on each of the 3 subacute days, as was done in the above naturalistic study. However, this may also be problematic, as one of the measures of pruning, the proportion best remaining, exhibited practice effects, which would presumably be exacerbated over 3 consecutive testing sessions so close together.

Whilst the results of the chapter 4 indicate a role for 5-HT in pruning, which supports Dayan and Huys (2008) theory of altered pruning in depression, it must be noted that activation of certain 5-HT receptors has been shown to affect the transmission of other neurotransmitter systems that have been implicated in reward and punishment processing. For example, Daw et al (2002) discuss the opponency between 5-HT and dopamine, and Di Matteo et al, 2008 show that agonism of 5-HT_{2C} receptors decreases the output of dopamine neurons *in vitro*,

whilst other 5-HT receptor subtypes have facilitative effects upon the dopamine system. Whilst no results exist examining the subacute effects upon the dopamine system due to MDMA-induced increased binding at any 5-HT sub-receptors, it is possible that dopamine function was also disrupted in these participants during the subacute period, which led to the observed differences in pruning behaviours. Further, the fact that dopamine has also been hypothesised to be dysfunctional in depression (e.g. Di Chiara et al, 1999) and the link between mood and pruning discovered in chapter 4, along with that reported in Huys et al (2012) further indicates that it may be fruitful to examine the role of dopamine in the pruning of decision trees.

Finally, it will prove useful to better understand participants' pruning behaviours by computationally modelling their choices on the pruning paradigm: doing so in chapters 3 and 6 provided evidence that participants' displayed aversive-based pruning behaviours very reliably. Whilst such computational models were built in order to better understand participants' choices in chapters 4 and 5, these models did not adequately capture such choices or performance on the task, particularly the effect of MDMA upon pruning behaviours. In part this reflects the difficulty in modelling within-subjects data using computational approaches. As such, further models are currently being developed, but could not be included in this thesis due to time constraints.

7.3.3 Chapter 5 limitations and directions for future research

The results of chapter 5 were intriguing, in that whilst they did provide some support for a role of 5-HT in decision-making, they did not support the theory of Dayan and Huys (2008) or replicate the findings of Rogers et al (2003) and Schweighofer et al (2008). The fact that administration of this dietary technique has led to conflicting results may indicate a lack of consistency in the study design. However, participants within this study were of a similar age (mean of 30.82 years) compared to other studies using this technique (i.e. Crockett et al, 2012) which had a mean age of 25.6 years. Further, participants were tested 5 hours after ingestion of the drink and had also not eaten throughout the testing session as in the above studies (although Crockett et al (2012) administered their behavioural tasks 5.5 hours after participants had ingested the amino acid mixture). However, Robinson et al (2013) allowed participants a small, tryptophan-free meal at lunchtime and detected ATD effects on a forced-choice emotion identification task. As such, it may be interesting to replicate this study whilst

allowing participants such small amounts of food midway through the testing day in order to reduce possible fatigue effects.

One explanation for why ATD produced no effect in the current sample, but MDMA did in chapter 4, is that there may be several alternative underlying mechanisms of this dietary technique that were not considered in this study. For instance, a recent review by Van Donkelaar et al (2011) argues that direct evidence that ATD decreases extracellular 5-HT concentrations is lacking. They posit possible alternative mechanisms by which ATD could affect performance on various tasks, some of them working indirectly via the 5-HT system. For instance, they argue that decreasing 5-HT (which is a powerful vasoconstrictor; Earley et al, 2013) may lead to decreased vasoconstrictor tone, which may contribute to the ATD-induced behavioural effects observed in other studies. Furthermore, they argue that decreasing 5-HT may decrease nitric oxide synthase (NOS) activity (Blokland et al, 1998) as well as decreasing both brain-derived neurotrophic factor (BDNF) and kynurenine (KYN) metabolites such as tryptophan 2, 3-dioxygenase, the latter of which converts the majority of the body's tryptophan into KYN, leading to a decrease in the availability of tryptophan in organs such as the liver. As such, the authors here argue that changes in neuronal 5-HT functioning may not be the sole, or even main contributor to ATD-induced behavioural effects observed in other studies. If this were to be the case, this may help explain why ATD had little effect upon pruning behaviours while large effects of subacute MDMA administration were observed in chapter 4: pruning is hypothesized to be affected solely by levels of 5-HT. However, it must be noted that a counter-argument to Van Donkelaar's was put forward by Crockett et al (2012), who show that there is strong evidence that ATD reduces brain 5-HT and disrupts stimulated 5-HT release in rats (Stancampiano et al, 1997, Moja et al, 1989), and that converging translational findings support a central role for brain 5-HT in ATD's effects upon performance on various cognitive tasks (e.g. Cools et al, 2008, Bari et al, 2010). However, no research findings exist that show the extent to which ATD leads to a decrease in 5-HT *release* in the human brain, and further research using the PET ligand used in chapter 3 (^{11}C-CUMI-101) could be performed in order to shed light on the ability of ATD to affect 5-HT release in the brain.

7.3.4 Chapter 6 limitations and directions for future research

The results of chapter 6 were counter to the hypothesis of Dayan and Huys (2008), revealing no differences between groups in terms of pruning behaviour. These results are interesting due to the fact that there have been no published results to date comparing the multi-step decisions made by a depressed and non-depressed population, and this study was the first to test Dayan and Huys' theory of altered pruning in depression. However, whilst it did not find any support for this theory, there are some limitations to this study.

Firstly, not every depressed patient in this study was recruited via a health care professional, with the remaining patients being recruited via advertisement. This means that the antidepressant histories of the latter patients could not be verified by the health care professional. If any of these patients were not forthcoming about their current use of antidepressants in order to be recruited for this study, it would make drawing conclusions as to a link between 5-HT and pruning difficult due to the fact that most antidepressants (e.g. the SSRIs) affect the 5-HT system (e.g. Anderson, 2000).

Secondly, the fact that those patients recruited from psychological services were undergoing cognitive behavioural therapy (CBT), and those recruited via advertisement were not, may have affected the results: for example CBT includes behavioural sessions that improve patients' ability to plan, which is an aspect of general pruning, as defined by the above computational models. As such, repeating this study including groups who both were and were not undergoing such therapy may improve the ability of this study to understand pruning in depression.

Thirdly, in order to better understand participants' pruning behaviours, developing further, more complex computational models of participants' choices could be fruitful. Whilst this would be more important for the studies included in chapters 4 and 5 (as no such models have yet been successfully applied to the data), adding further parameters to the model that attempt to understand participants' choices based upon their previous choices (indicating the formation of habits), for example, may help model parsimony. Whilst these are planned for future analyses, new and more complex computational models were unfortunately not formed in time for this thesis. Future research should examine this avenue of analysis.

Finally, testing both depressed patients who are, and who are not undergoing treatment with a range of antidepressants against control subjects would allow for a more complete understanding of how 5-HT, and potentially other neurotransmitter systems, are linked to pruning and mood. Further, this would also allow for the observation of how improvements in mood due to antidepressant treatment may be linked to improvements in decision-making.

7.4 Conclusion

This thesis has examined the relationship between 5-HT, decision-making and mood. It has provided an overview of the literature on the cognitive and monoamine deficits in depression, before describing the results of studies examining the relationship between 5-HT and reward and punishment processing. It then explained the experimental techniques used in each of the four studies presented, before presenting the results of these four experiments.

The first experiment utilised PET in order to examine a relationship between the availability of the 5-HT_{1A} receptor and performance on 3 decision-making tasks. The results of this study showed positive correlations between both 5-HT_{1A} availability in the hippocampus and the amount participants use information pertaining to probability on a gambling task; and between 5-HT_{1A} availability after citalopram infusion in the nucleus accumbens and the amount participants use information pertaining to losses on the same gambling task; and a negative relationship between 5-HT_{1A} availability and temporal discounting. The second study tested Dayan and Huys' (2008) of altered 5-HT transmission and pruning by examining the subacute effects of MDMA administration upon pruning. The results of this study revealed that MDMA subacutely decreased pruning behaviours but not mood. However, a negative correlation between negative affect and pruning was observed, such that those participants who displayed increased negative affect also displayed increased pruning, which contradicts that hypothesised by Dayan and Huys (2008). The third experiment examined the effect of ATD upon decision-making. The results of this study showed no effect of ATD upon participants' pruning behaviours (thus providing no support for Dayan and Huys' theory), but reduced participants' choices to high probability gambles, supporting previous research. No effect of ATD treatment upon the rate at which participants discount the value of a future reward were observed. Finally, the fourth study tested Dayan and Huys' (2008) theory that low levels of 5-HT leads to low mood by decreasing pruning. Using Bayesian model comparison, it was revealed that both groups displayed aversive-based pruning, but that there were no significant differences between groups in the performance on this task. As

such, the results of this study failed to provide support for Dayan and Huys (2008) above theory.

In conclusion, this thesis has provided some evidence of a relationship between 5-HT, decision-making and mood. For example a relationship between reward and punishment processing during gambling behaviours and temporal discounting with the availability of a specific 5-HT receptor subtype was observed, as was a relationship between pruning behaviours and a putative decrease in 5-HT following MDMA administration. However, with the focus of this thesis being on the relationship between 5-HT, pruning and mood, this thesis has largely failed to provide support for the theory of Dayan and Huys (2008); no relationship between pruning and low levels of tryptophan after ATD or depression were observed, and the only correlation between pruning behaviours and mood (from chapter 4) was in the opposite direction to that predicted by the above theory. As such, the experiments presented in this thesis have yielded some potentially important results, consistent with a role for 5-HT in reward and punishment processing, particularly in the pruning of decision trees and probabilistic choice. However, whilst depression has been associated with dysfunctional 5-HT transmission, the results of this chapter have indicated that attempting to understand depressed patients' pruning behaviours in order to improve treatments for the disorder may not be the most fruitful avenue of research.

8) REFERENCES

Abler, B., Grön, G., Hartmann, A., Metzger, C., Walter, M., 2012. Modulation of frontostriatal interaction aligns with reduced primary reward processing under serotonergic drugs. J. Neurosci. 32, 1329–1335.

Adell, A., Casanovas, J.M., Artigas, F., 1997. Comparative study in the rat of the actions of different types of stress on the release of 5-HT in raphe nuclei and forebrain areas. Neuropharmacology 36, 735–741.

Ahn, K.-C., Pazderka-Robinson, H., Clements, R., Ashcroft, R., Ali, T., Morse, C., Greenshaw, A.J., 2005. Differential effects of intra-midbrain raphe and systemic 8-OH-DPAT on VTA self-stimulation thresholds in rats. Psychopharmacology (Berl.) 178, 381–388.

Allen, P.P., Cleare, A.J., Lee, F., Fusar-Poli, P., Tunstall, N., Fu, C.H.Y., Brammer, M.J., McGuire, P.K., 2006. Effect of acute tryptophan depletion on pre-frontal engagement. Psychopharmacology (Berl.) 187, 486–497.

Altman, H.J., Normile, H.J., 1988. What is the nature of the role of the serotonergic nervous system in learning and memory: prospects for development of an effective treatment strategy for senile dementia. Neurobiol. Aging 9, 627–638.

Altman, S.E., Shankman, S.A., Spring, B., 2010. Effect of acute tryptophan depletion on emotions in individuals with personal and family history of depression following a mood induction. Neuropsychobiology 62, 171–176.

Amat, J., Matus-Amat, P., Watkins, L.R., Maier, S.F., 1998. Escapable and inescapable stress differentially and selectively alter extracellular levels of 5-HT in the ventral hippocampus and dorsal periaqueductal gray of the rat. Brain Res. 797, 12–22.

Amin, Z., Gueorguieva, R., Cappiello, A., Czarkowski, K.A., Stiklus, S., Anderson, G.M., Naftolin, F., Epperson, C.N., 2006. Estradiol and tryptophan depletion interact to modulate cognition in menopausal women. Neuropsychopharmacology 31, 2489–2497.

Anderson, I.M., 2000. Selective serotonin reuptake inhibitors versus tricyclic antidepressants: a meta-analysis of efficacy and tolerability. J Affect Disord 58, 19–36.

Anderson, I.M., Richell, R.A., Bradshaw, C.M., 2003. The effect of acute tryptophan depletion on probabilistic choice. J. Psychopharmacol. (Oxford) 17, 3–7.

Asberg, M., Träskman, L., Thorén, P., 1976. 5-HIAA in the cerebrospinal fluid. A biochemical suicide predictor? Arch. Gen. Psychiatry 33, 1193–1197.

Attar-Lévy, D., Martinot, J.L., Blin, J., Dao-Castellana, M.H., Crouzel, C., Mazoyer, B., Poirier, M.F., Bourdel, M.C., Aymard, N., Syrota, A., Féline, A., 1999. The cortical serotonin2 receptors studied with positron-emission tomography and [18F]-setoperone during depressive illness and antidepressant treatment with clomipramine. Biol. Psychiatry 45, 180–186.

Attenburrow, M.J., Mitter, P.R., Whale, R., Terao, T., Cowen, P.J., 2001. Low-dose citalopram as a 5-HT neuroendocrine probe. Psychopharmacology (Berl.) 155, 323–326.

Austin, M.P., Ross, M., Murray, C., O'Carroll, R.E., Ebmeier, K.P., Goodwin, G.M., 1992. Cognitive function in major depression. J Affect Disord 25, 21–29.

Bäckman, J., Alling, C., Alsén, M., Regnéll, G., Träskman-Bendz, L., 2000. Changes of cerebrospinal fluid monoamine metabolites during long-term antidepressant treatment. Eur Neuropsychopharmacol 10, 341–349.

Bari, A., Eagle, D.M., Mar, A.C., Robinson, E.S.J., Robbins, T.W., 2009. Dissociable effects of noradrenaline, dopamine, and serotonin uptake blockade on stop task performance in rats. Psychopharmacology (Berl.) 205, 273–283.

Bari, A., Theobald, D.E., Caprioli, D., Mar, A.C., Aidoo-Micah, A., Dalley, J.W., Robbins, T.W., 2010. Serotonin modulates sensitivity to reward and negative feedback in a probabilistic reversal learning task in rats. Neuropsychopharmacology 35, 1290–1301.

Barnes, N.M., Sharp, T., 1999. A review of central 5-HT receptors and their function. Neuropharmacology 38, 1083–1152.

Barrientos, R.M., O'Reilly, R.C., Rudy, J.W., 2002. Memory for context is impaired by injecting anisomycin into dorsal hippocampus following context exploration. Behav. Brain Res. 134, 299–306.

Baumeister, A.A., Hawkins, M.F., Uzelac, S.M., 2003. The myth of reserpine-induced depression: role in the historical development of the monoamine hypothesis. J Hist Neurosci 12, 207–220.

Beats, B.C., Sahakian, B.J., Levy, R., 1996. Cognitive performance in tests sensitive to frontal lobe dysfunction in the elderly depressed. Psychol Med 26, 591–603.

Bechara, A., Damasio, A.R., Damasio, H., Anderson, S.W., 1994. Insensitivity to future consequences following damage to human prefrontal cortex. Cognition 50, 7–15.

Beck, A.T., Ward, C.H., Mendelson, M., Mock, J., Erbaugh, J., 1961. An inventory for measuring depression. Arch. Gen. Psychiatry 4, 561–571.

Beck, A.T., Weissman, A., Kovacs, M., 1976. Alcoholism, hopelessness and suicidal behavior. J. Stud. Alcohol 37, 66–77.

Beck, D., 1967. [Indication for psychoanalytic short time therapy]. Z Psychosom Med Psychoanal 13, 257–265.

Benko, A., Lazary, J., Molnar, E., Gonda, X., Tothfalusi, L., Pap, D., Mirnics, Z., Kurimay, T., Chase, D., Juhasz, G., Anderson, I.M., Deakin, J.F.W., Bagdy, G., 2010. Significant association between the C(-1019)G functional polymorphism of the HTR1A gene and impulsivity. Am. J. Med. Genet. B Neuropsychiatr. Genet. 153B, 592–599.

Bhagwagar, Z., Rabiner, E.A., Sargent, P.A., Grasby, P.M., Cowen, P.J., 2004. Persistent reduction in brain serotonin1A receptor binding in recovered depressed men measured by positron emission tomography with [11C]WAY-100635. Mol. Psychiatry 9, 386–392.

Bickel, W.K., Marsch, L.A., 2001. Toward a behavioral economic understanding of drug dependence: delay discounting processes. Addiction 96, 73–86.

Bickel, W.K., Pitcock, J.A., Yi, R., Angtuaco, E.J.C., 2009. Congruence of BOLD response across intertemporal choice conditions: fictive and real money gains and losses. J. Neurosci. 29, 8839–8846.

Bizot, J.C., Thiébot, M.H., Le Bihan, C., Soubrié, P., Simon, P., 1988. Effects of imipramine-like drugs and serotonin uptake blockers on delay of reward in rats. Possible implication in the behavioral mechanism of action of antidepressants. J. Pharmacol. Exp. Ther. 246, 1144–1151.

Bjork, J.M., Dougherty, D.M., Moeller, F.G., Swann, A.C., 2000. Differential behavioral effects of plasma tryptophan depletion and loading in aggressive and nonaggressive men. Neuropsychopharmacology 22, 357–369.

Blier, P., 2003. The pharmacology of putative early-onset antidepressant strategies. Eur Neuropsychopharmacol 13, 57–66.

Blier, P., Piñeyro, G., el Mansari, M., Bergeron, R., de Montigny, C., 1998. Role of somatodendritic 5-HT autoreceptors in modulating 5-HT neurotransmission. Ann. N. Y. Acad. Sci. 861, 204–216.

Blier, P., Ward, N.M., 2003. Is there a role for 5-HT1A agonists in the treatment of depression? Biol. Psychiatry 53, 193–203.

Blokland, A., Prickaerts, J., Honig, W., de Vente, J., 1998. State-dependent impairment in object recognition after hippocampal NOS inhibition. Neuroreport 9, 4205–4208.

Blokland, A., Sik, A., Lieben, C., 2005. Evaluation of DOI, 8-OH-DPAT, eticlopride and amphetamine on impulsive responding in a reaction time task in rats. Behav Pharmacol 16, 93–100.

Booij, L., Van der Does, A.J.W., Riedel, W.J., 2003. Monoamine depletion in psychiatric and healthy populations: review. Mol. Psychiatry 8, 951–973.

Booij, L., Van der Does, W., Benkelfat, C., Bremner, J.D., Cowen, P.J., Fava, M., Gillin, C., Leyton, M., Moore, P., Smith, K.A., Van der Kloot, W.A., 2002. Predictors of mood response to acute tryptophan depletion. A reanalysis. Neuropsychopharmacology 27, 852–861.

Bostwick, J.M., Pankratz, V.S., 2000. Affective disorders and suicide risk: a reexamination. Am J Psychiatry 157, 1925–1932.

Bremner, J.D., Innis, R.B., Salomon, R.M., Staib, L.H., Ng, C.K., Miller, H.L., Bronen, R.A., Krystal, J.H., Duncan, J., Rich, D., Price, L.H., Malison, R., Dey, H., Soufer, R., Charney, D.S., 1997. Positron emission tomography measurement of cerebral metabolic correlates of tryptophan depletion-induced depressive relapse. Arch. Gen. Psychiatry 54, 364–374.

Breslow, R., Kocsis, J., Belkin, B., 1980. Memory deficits in depression: evidence utilizing the Wechsler Memory Scale. Percept Mot Skills 51, 541–542.

Brown, R.G., Scott, L.C., Bench, C.J., Dolan, R.J., 1994. Cognitive function in depression: its relationship to the presence and severity of intellectual decline. Psychol Med 24, 829–847.

Budygin, E.A., Brodie, M.S., Sotnikova, T.D., Mateo, Y., John, C.E., Cyr, M., Gainetdinov, R.R., Jones, S.R., 2004. Dissociation of rewarding and dopamine transporter-mediated properties of amphetamine. Proc. Natl. Acad. Sci. U.S.A. 101, 7781–7786.

Camara, E., Rodriguez-Fornells, A., Münte, T.F., 2008. Functional connectivity of reward processing in the brain. Front Hum Neurosci 2, 19.

Cardinal, R.N., Winstanley, C.A., Robbins, T.W., Everitt, B.J., 2004. Limbic corticostriatal systems and delayed reinforcement. Ann. N. Y. Acad. Sci. 1021, 33–50.

Carli, M., Samanin, R., 2000. The 5-HT(1A) receptor agonist 8-OH-DPAT reduces rats' accuracy of attentional performance and enhances impulsive responding in a five-choice serial reaction time task: role of presynaptic 5-HT(1A) receptors. Psychopharmacology (Berl.) 149, 259–268.

Carpenter, L.L., Anderson, G.M., Pelton, G.H., Gudin, J.A., Kirwin, P.D., Price, L.H., Heninger, G.R., McDougle, C.J., 1998. Tryptophan depletion during continuous CSF sampling in healthy human subjects. Neuropsychopharmacology 19, 26–35.

Castaneda, A.E., Tuulio-Henriksson, A., Marttunen, M., Suvisaari, J., Lönnqvist, J., 2008. A review on cognitive impairments in depressive and anxiety disorders with a focus on young adults. J Affect Disord 106, 1–27.

Cervantes, M.C., Delville, Y., 2009. Serotonin 5-HT1A and 5-HT3 receptors in an impulsive-aggressive phenotype. Behav. Neurosci. 123, 589–598.

Chamberlain, S.R., Del Campo, N., Dowson, J., Müller, U., Clark, L., Robbins, T.W., Sahakian, B.J., 2007. Atomoxetine improved response inhibition in adults with attention deficit/hyperactivity disorder. Biol. Psychiatry 62, 977–984.

Chambers, C.D., Johnson, K.A., Dick, L.M., Felix, R.J., Jones, K.L., 1996. Birth outcomes in pregnant women taking fluoxetine. N. Engl. J. Med. 335, 1010–1015.

Chase, H.W., Frank, M.J., Michael, A., Bullmore, E.T., Sahakian, B.J., Robbins, T.W., 2010. Approach and avoidance learning in patients with major depression and healthy controls: relation to anhedonia. Psychol Med 40, 433–440.

Cheung, T.H.C., Cardinal, R.N., 2005. Hippocampal lesions facilitate instrumental learning with delayed reinforcement but induce impulsive choice in rats. BMC Neurosci 6, 36.

Clark, L., Dombrovski, A.Y., Siegle, G.J., Butters, M.A., Shollenberger, C.L., Sahakian, B.J., Szanto, K., 2011. Impairment in risk-sensitive decision-making in older suicide attempters with depression. Psychol Aging 26, 321–330.

Clark, L., Roiser, J.P., Cools, R., Rubinsztein, D.C., Sahakian, B.J., Robbins, T.W., 2005. Stop signal response inhibition is not modulated by tryptophan depletion or the serotonin transporter polymorphism in healthy volunteers: implications for the 5-HT theory of impulsivity. Psychopharmacology (Berl.) 182, 570–578.

Clark, L., Roiser, J.P., Robbins, T.W., Sahakian, B.J., 2009. Disrupted "reflection" impulsivity in cannabis users but not current or former ecstasy users. J. Psychopharmacol. (Oxford) 23, 14–22.

Clifford, P.I., Hemsley, D.R., 1987. The influence of depression on the processing of personal attributes. Br J Psychiatry 150, 98–103.

Cohen, M.X., Elger, C.E., Weber, B., 2008. Amygdala tractography predicts functional connectivity and learning during feedback-guided decision-making. Neuroimage 39, 1396–1407.

Cole, J.C., Bailey, M., Sumnall, H.R., Wagstaff, G.F., King, L.A., 2002. The content of ecstasy tablets: implications for the study of their long-term effects. Addiction 97, 1531–1536.

Cools, R., Blackwell, A., Clark, L., Menzies, L., Cox, S., Robbins, T.W., 2005. Tryptophan depletion disrupts the motivational guidance of goal-directed behavior as a function of trait impulsivity. Neuropsychopharmacology 30, 1362–1373.

Cools, R., Nakamura, K., Daw, N.D., 2011. Serotonin and dopamine: unifying affective, activational, and decision functions. Neuropsychopharmacology 36, 98–113.

Cools, R., Robinson, O.J., Sahakian, B., 2008. Acute tryptophan depletion in healthy volunteers enhances punishment prediction but does not affect reward prediction. Neuropsychopharmacology 33, 2291–2299.

Cools, R., Sheridan, M., Jacobs, E., D'Esposito, M., 2007. Impulsive personality predicts dopamine-dependent changes in frontostriatal activity during component processes of working memory. J. Neurosci. 27, 5506–5514.

Coppen, A., 1967. The biochemistry of affective disorders. Br J Psychiatry 113, 1237–1264.

Corbetta, M., Shulman, G.L., 2002. Control of goal-directed and stimulus-driven attention in the brain. Nat. Rev. Neurosci. 3, 201–215.

Corruble, E., Hatem, N., Damy, C., Falissard, B., Guelfi, J.-D., Reynaud, M., Hardy, P., 2003. Defense styles, impulsivity and suicide attempts in major depression. Psychopathology 36, 279–284.

Cowan, R.L., 2007. Neuroimaging research in human MDMA users: a review. Psychopharmacology (Berl.) 189, 539–556.

Cowen, P.J., 2008. Serotonin and depression: pathophysiological mechanism or marketing myth? Trends Pharmacol. Sci. 29, 433–436.

Crean, J., Richards, J.B., de Wit, H., 2002. Effect of tryptophan depletion on impulsive behavior in men with or without a family history of alcoholism. Behav. Brain Res. 136, 349–357.

Cremniter, D., Jamain, S., Kollenbach, K., Alvarez, J.C., Lecrubier, Y., Gilton, A., Jullien, P., Lesieur, P., Bonnet, F., Spreux-Varoquaux, O., 1999. CSF 5-HIAA levels are lower in impulsive as compared to nonimpulsive violent suicide attempters and control subjects. Biol. Psychiatry 45, 1572–1579.

Crockett, M.J., Clark, L., Robbins, T.W., 2009. Reconciling the role of serotonin in behavioral inhibition and aversion: acute tryptophan depletion abolishes punishment-induced inhibition in humans. J. Neurosci. 29, 11993–11999.

Crockett, M.J., Clark, L., Roiser, J.P., Robinson, O.J., Cools, R., Chase, H.W., Ouden, H. den, Apergis-Schoute, A., Campbell-Meiklejohn, D., Campbell-Meikeljohn, D., Seymour, B., Sahakian, B.J., Rogers, R.D., Robbins, T.W., 2012a. Converging evidence for central 5-HT effects in acute tryptophan depletion. Mol. Psychiatry 17, 121–123.

Crockett, M.J., Clark, L., Smillie, L.D., Robbins, T.W., 2012b. The effects of acute tryptophan depletion on costly information sampling: impulsivity or aversive processing? Psychopharmacology (Berl.) 219, 587–597.

Croft, R.J., Mackay, A.J., Mills, A.T., Gruzelier, J.G., 2001. The relative contributions of ecstasy and cannabis to cognitive impairment. Psychopharmacology (Berl.) 153, 373–379.

Curran, H.V., Travill, R.A., 1997. Mood and cognitive effects of +/-3,4-methylenedioxymethamphetamine (MDMA, "ecstasy"): week-end "high" followed by mid-week low. Addiction 92, 821–831.

Cuyàs, E., Verdejo-García, A., Fagundo, A.B., Khymenets, O., Rodríguez, J., Cuenca, A., de Sola Llopis, S., Langohr, K., Peña-Casanova, J., Torrens, M., Martín-Santos, R., Farré, M., de la Torre, R., 2011. The influence of genetic and environmental factors among MDMA users in cognitive performance. PLoS ONE 6, e27206.

Czesak, M., Le François, B., Millar, A.M., Deria, M., Daigle, M., Visvader, J.E., Anisman, H., Albert, P.R., 2012. Increased serotonin-1A (5-HT1A) autoreceptor expression and reduced raphe serotonin levels in deformed epidermal autoregulatory factor-1 (Deaf-1) gene knock-out mice. J. Biol. Chem. 287, 6615–6627.

Dafters, R.I., Hoshi, R., Talbot, A.C., 2004. Contribution of cannabis and MDMA ("ecstasy") to cognitive changes in long-term polydrug users. Psychopharmacology (Berl.) 173, 405–410.

Dahlström, M., Ahonen, A., Ebeling, H., Torniainen, P., Heikkilä, J., Moilanen, I., 2000. Elevated hypothalamic/midbrain serotonin (monoamine) transporter availability in depressive drug-naive children and adolescents. Mol. Psychiatry 5, 514–522.

Dalley, J.W., Roiser, J.P., 2012. Dopamine, serotonin and impulsivity. Neuroscience 215, 42–58.

Daw, N.D., Kakade, S., Dayan, P., 2002. Opponent interactions between serotonin and dopamine. Neural Netw 15, 603–616.

Dayan, P., Huys, Q.J.M., 2008. Serotonin, inhibition, and negative mood. PLoS Comput. Biol. 4, e4.

De Kwaasteniet, B., Ruhe, E., Caan, M., Rive, M., Olabarriaga, S., Groefsema, M., Heesink, L., van Wingen, G., Denys, D., 2013. Relation Between Structural and Functional Connectivity in Major Depressive Disorder. Biological Psychiatry 74, 40–47.

Deakin, J.F., Graeff, F.G., 1991. 5-HT and mechanisms of defence. J. Psychopharmacol. (Oxford) 5, 305–315.

Dearing, K.F., Gotlib, I.H., 2009. Interpretation of ambiguous information in girls at risk for depression. J Abnorm Child Psychol 37, 79–91.

Del-Ben, C.M., Deakin, J.F.W., Mckie, S., Delvai, N.A., Williams, S.R., Elliott, R., Dolan, M., Anderson, I.M., 2005. The Effect of Citalopram Pretreatment on Neuronal Responses to Neuropsychological Tasks in Normal Volunteers: An fMRI Study. Neuropsychopharmacology 30, 1724–1734.

Delgado, M.R., Nystrom, L.E., Fissell, C., Noll, D.C., Fiez, J.A., 2000. Tracking the hemodynamic responses to reward and punishment in the striatum. J. Neurophysiol. 84, 3072–3077.

Delgado, P.L., Charney, D.S., Price, L.H., Aghajanian, G.K., Landis, H., Heninger, G.R., 1990. Serotonin function and the mechanism of antidepressant action. Reversal of antidepressant-induced remission by rapid depletion of plasma tryptophan. Arch. Gen. Psychiatry 47, 411–418.

Demoto, Y., Okada, G., Okamoto, Y., Kunisato, Y., Aoyama, S., Onoda, K., Munakata, A., Nomura, M., Tanaka, S.C., Schweighofer, N., Doya, K., Yamawaki, S., 2012. Neural and personality correlates of individual differences related to the effects of acute tryptophan depletion on future reward evaluation. Neuropsychobiology 65, 55–64.

Di Chiara, G., Loddo, P., Tanda, G., 1999. Reciprocal changes in prefrontal and limbic dopamine responsiveness to aversive and rewarding stimuli after chronic mild stress: implications for the psychobiology of depression. Biol. Psychiatry 46, 1624–1633.

Di Matteo, V., Di Giovanni, G., Pierucci, M., Esposito, E., 2008. Serotonin control of central dopaminergic function: focus on in vivo microdialysis studies. Prog. Brain Res. 172, 7–44.

Dias, R., Robbins, T.W., Roberts, A.C., 1996. Dissociation in prefrontal cortex of affective and attentional shifts. Nature 380, 69–72.

Dombrovski, A.Y., Szanto, K., Clark, L., Reynolds, C.F., Siegle, G.J., 2013. Reward Signals, Attempted Suicide, and Impulsivity in Late-Life Depression. JAMA Psychiatry.

Drevets, W.C., 2000. Neuroimaging studies of mood disorders. Biol. Psychiatry 48, 813–829.

Drevets, W.C., 2001. Neuroimaging and neuropathological studies of depression: implications for the cognitive-emotional features of mood disorders. Curr. Opin. Neurobiol. 11, 240–249.

Drevets, W.C., 2003. Neuroimaging abnormalities in the amygdala in mood disorders. Ann. N. Y. Acad. Sci. 985, 420–444.

Drevets, W.C., Frank, E., Price, J.C., Kupfer, D.J., Holt, D., Greer, P.J., Huang, Y., Gautier, C., Mathis, C., 1999. PET imaging of serotonin 1A receptor binding in depression. Biol. Psychiatry 46, 1375–1387.

Drevets, W.C., Ongür, D., Price, J.L., 1998a. Neuroimaging abnormalities in the subgenual prefrontal cortex: implications for the pathophysiology of familial mood disorders. Mol. Psychiatry 3, 220–226, 190–191.

Drevets, W.C., Ongür, D., Price, J.L., 1998b. Reduced glucose metabolism in the subgenual prefrontal cortex in unipolar depression. Mol. Psychiatry 3, 190–191.

Drevets, W.C., Price, J.L., Bardgett, M.E., Reich, T., Todd, R.D., Raichle, M.E., 2002. Glucose metabolism in the amygdala in depression: relationship to diagnostic subtype and plasma cortisol levels. Pharmacol. Biochem. Behav. 71, 431–447.

Drevets, W.C., Price, J.L., Simpson, J.R., Jr, Todd, R.D., Reich, T., Vannier, M., Raichle, M.E., 1997. Subgenual prefrontal cortex abnormalities in mood disorders. Nature 386, 824–827.

Drevets, W.C., Thase, M.E., Moses-Kolko, E.L., Price, J., Frank, E., Kupfer, D.J., Mathis, C., 2007. Serotonin-1A receptor imaging in recurrent depression: replication and literature review. Nucl. Med. Biol. 34, 865–877.

Duman, R.S., Nakagawa, S., Malberg, J., 2001. Regulation of adult neurogenesis by antidepressant treatment. Neuropsychopharmacology 25, 836–844.

Eagle, D.M., Lehmann, O., Theobald, D.E.H., Pena, Y., Zakaria, R., Ghosh, R., Dalley, J.W., Robbins, T.W., 2009. Serotonin depletion impairs waiting but not stop-signal reaction time in rats: implications for theories of the role of 5-HT in behavioral inhibition. Neuropsychopharmacology 34, 1311–1321.

Earley, S., Leblanc, N., 2013. Serotonin receptors take the TRiPV4 highway in chronic hypoxic pulmonary hypertension. Focus on "TRPV4 channel contributes to serotonin-induced pulmonary vasoconstriction and the enhanced vascular reactivity in chronic hypoxic pulmonary hypertension". Am. J. Physiol., Cell Physiol. 305, C690–692.

Ellenbogen, M.A., Young, S.N., Dean, P., Palmour, R.M., Benkelfat, C., 1999. Acute tryptophan depletion in healthy young women with a family history of major affective disorder. Psychol Med 29, 35–46.

Elliott, R., Friston, K.J., Dolan, R.J., 2000. Dissociable Neural Responses in Human Reward Systems. J. Neurosci. 20, 6159–6165.

Elliott, R., Newman, J.L., Longe, O.A., Deakin, J.F.W., 2003. Differential Response Patterns in the Striatum and Orbitofrontal Cortex to Financial Reward in Humans: A Parametric Functional Magnetic Resonance Imaging Study. J. Neurosci. 23, 303–307.

Elliott, R., Newman, J.L., Longe, O.A., William Deakin, J.., 2004. Instrumental responding for rewards is associated with enhanced neuronal response in subcortical reward systems. NeuroImage 21, 984–990.

Elliott, R., Sahakian, B.J., McKay, A.P., Herrod, J.J., Robbins, T.W., Paykel, E.S., 1996. Neuropsychological impairments in unipolar depression: the influence of perceived failure on subsequent performance. Psychol Med 26, 975–989.

Eshel, N., Roiser, J.P., 2010. Reward and punishment processing in depression. Biol. Psychiatry 68, 118–124.

Evenden, J.L., Ryan, C.N., 1999. The pharmacology of impulsive behaviour in rats VI: the effects of ethanol and selective serotonergic drugs on response choice with varying delays of reinforcement. Psychopharmacology (Berl.) 146, 413–421.

Everitt, B.J., Parkinson, J.A., Olmstead, M.C., Arroyo, M., Robledo, P., Robbins, T.W., 1999. Associative processes in addiction and reward. The role of amygdala-ventral striatal subsystems. Ann. N. Y. Acad. Sci. 877, 412–438.

Evers, E.A.T., van der Veen, F.M., van Deursen, J.A., Schmitt, J.A.J., Deutz, N.E.P., Jolles, J., 2006. The effect of acute tryptophan depletion on the BOLD response during performance monitoring and response inhibition in healthy male volunteers. Psychopharmacology (Berl.) 187, 200–208.

Fanselow, M.S., Dong, H.-W., 2010. Are the dorsal and ventral hippocampus functionally distinct structures? Neuron 65, 7–19.

Fantegrossi, W.E., Ullrich, T., Rice, K.C., Woods, J.H., Winger, G., 2002. 3,4-Methylenedioxymethamphetamine (MDMA, "ecstasy") and its stereoisomers as reinforcers in rhesus monkeys: serotonergic involvement. Psychopharmacology (Berl.) 161, 356–364.

Feder, A., Skipper, J., Blair, J.R., Buchholz, K., Mathew, S.J., Schwarz, M., Doucette, J.T., Alonso, A., Collins, K.A., Neumeister, A., Charney, D.S., 2011. Tryptophan depletion and emotional processing in healthy volunteers at high risk for depression. Biol. Psychiatry 69, 804–807.

Fitzgerald, J.L., Reid, J.J., 1990. Effects of methylenedioxymethamphetamine on the release of monoamines from rat brain slices. Eur. J. Pharmacol. 191, 217–220.

Fleckenstein, A.E., Volz, T.J., Riddle, E.L., Gibb, J.W., Hanson, G.R., 2007. New insights into the mechanism of action of amphetamines. Annu. Rev. Pharmacol. Toxicol. 47, 681–698.

Fletcher, P.J., Korth, K.M., Robinson, S.R., Baker, G.B., 2002. Multiple 5-HT receptors are involved in the effects of acute MDMA treatment: studies on locomotor activity and responding for conditioned reinforcement. Psychopharmacology (Berl.) 162, 282–291.

Fossati, P., Guillaume, L.B., Ergis, A.-M., Allilaire, J.-F., 2003. Qualitative analysis of verbal fluency in depression. Psychiatry Res 117, 17–24.

Fossati, P., Harvey, P.-O., Le Bastard, G., Ergis, A.-M., Jouvent, R., Allilaire, J.-F., 2004. Verbal memory performance of patients with a first depressive episode and patients with unipolar and bipolar recurrent depression. J Psychiatr Res 38, 137–144.

Foster, D.J., Morris, R.G., Dayan, P., 2000. A model of hippocampally dependent navigation, using the temporal difference learning rule. Hippocampus 10, 1–16.

Frankle, W.G., Lombardo, I., New, A.S., Goodman, M., Talbot, P.S., Huang, Y., Hwang, D.-R., Slifstein, M., Curry, S., Abi-Dargham, A., Laruelle, M., Siever, L.J., 2005. Brain serotonin transporter distribution in subjects with impulsive aggressivity: a positron emission study with [11C]McN 5652. Am J Psychiatry 162, 915–923.

FREIS, E.D., 1954. Mental depression in hypertensive patients treated for long periods with large doses of reserpine. N. Engl. J. Med. 251, 1006–1008.

Frodl, T., Bokde, A.L.W., Scheuerecker, J., Lisiecka, D., Schoepf, V., Hampel, H., Möller, H.-J., Brückmann, H., Wiesmann, M., Meisenzahl, E., 2010. Functional Connectivity Bias of the Orbitofrontal Cortex in Drug-Free Patients with Major Depression. Biological Psychiatry 67, 161–167.

Furst, S., 1990. Brain monoamines are involved in the sedative effects of opiates and neuroleptics. Prog. Clin. Biol. Res. 328, 311–314.

Gallagher, P., Massey, A.E., Young, A.H., McAllister-Williams, R.H., 2003. Effects of acute tryptophan depletion on executive function in healthy male volunteers. BMC Psychiatry 3, 10.

Gamma, A., Buck, A., Berthold, T., Liechti, M.E., Vollenweider, F.X., Hell, D., 2000. 3,4-Methylenedioxymethamphetamine (MDMA) modulates cortical and limbic brain activity as measured by [H(2)(15)O]-PET in healthy humans. Neuropsychopharmacology 23, 388–395.

Gehring, W.J., Willoughby, A.R., 2002. The medial frontal cortex and the rapid processing of monetary gains and losses. Science 295, 2279–2282.

George, M.S., Ketter, T.A., Parekh, P.I., Horwitz, B., Herscovitch, P., Post, R.M., 1995. Brain activity during transient sadness and happiness in healthy women. Am J Psychiatry 152, 341–351.

Gerra, G., Zaimovic, A., Giucastro, G., Maestri, D., Monica, C., Sartori, R., Caccavari, R., Delsignore, R., 1998. Serotonergic function after (+/-)3,4-methylene-dioxymethamphetamine (’Ecstasy’) in humans. Int Clin Psychopharmacol 13, 1–9.

Gevins, A., Cutillo, B., 1993. Spatiotemporal dynamics of component processes in human working memory. Electroencephalogr Clin Neurophysiol 87, 128–143.

Giovacchini, G., Lang, L., Ma, Y., Herscovitch, P., Eckelman, W.C., Carson, R.E., 2005. Differential effects of paroxetine on raphe and cortical 5-HT1A binding: a PET study in monkeys. Neuroimage 28, 238–248.

Gotlib, I.H., Joormann, J., 2010. Cognition and depression: current status and future directions. Annu Rev Clin Psychol 6, 285–312.

Graeff, F.G., Viana, M.B., Mora, P.O., 1996. Opposed regulation by dorsal raphe nucleus 5-HT pathways of two types of fear in the elevated T-maze. Pharmacol. Biochem. Behav. 53, 171–177.

Greenberg, P.E., Birnbaum, H.G., 2005. The economic burden of depression in the US: societal and patient perspectives. Expert Opinion on Pharmacotherapy 6, 369–376.

Gu, H., Liu, C., Liu, C., Chen, M., Zhang, Q., Zhai, J., Wang, K., Ji, F., Xu, Z., Shen, Q., Bao, X., Chen, X., Li, J., Dong, Q., Chen, C., 2013. The combined effects of the 5- HTTLPR and HTR1A rs6295 polymorphisms modulate decision making in schizophrenia patients. Genes Brain Behav. 12, 133–139.

Guitart-Masip, M., Bunzeck, N., Stephan, K.E., Dolan, R.J., Düzel, E., 2010. Contextual novelty changes reward representations in the striatum. J. Neurosci. 30, 1721–1726.

Gunn, R.N., Lammertsma, A.A., Hume, S.P., Cunningham, V.J., 1997. Parametric imaging of ligand-receptor binding in PET using a simplified reference region model. Neuroimage 6, 279–287.

Gurden, H., Takita, M., Jay, T.M., 2000. Essential role of D1 but not D2 receptors in the NMDA receptor-dependent long-term potentiation at hippocampal-prefrontal cortex synapses in vivo. J. Neurosci. 20, RC106.

Halpern, J.H., Pope, H.G., Jr, Sherwood, A.R., Barry, S., Hudson, J.I., Yurgelun-Todd, D., 2004. Residual neuropsychological effects of illicit 3,4-methylenedioxymethamphetamine (MDMA) in individuals with minimal exposure to other drugs. Drug Alcohol Depend 75, 135–147.

Hamilton, J.P., Etkin, A., Furman, D.J., Lemus, M.G., Johnson, R.F., Gotlib, I.H., 2012. Functional neuroimaging of major depressive disorder: a meta-analysis and new integration of base line activation and neural response data. Am J Psychiatry 169, 693–703.

HAMILTON, M., 1960. A rating scale for depression. J. Neurol. Neurosurg. Psychiatr. 23, 56–62.

Hammers, A., Chen, C.-H., Lemieux, L., Allom, R., Vossos, S., Free, S.L., Myers, R., Brooks, D.J., Duncan, J.S., Koepp, M.J., 2007. Statistical neuroanatomy of the human inferior frontal gyrus and probabilistic atlas in a standard stereotaxic space. Hum Brain Mapp 28, 34–48.

Hannon, J., Hoyer, D., 2008. Molecular biology of 5-HT receptors. Behav. Brain Res. 195, 198–213.

Harmer, C.J., Cowen, P.J., 2013. "It"s the way that you look at it'--a cognitive neuropsychological account of SSRI action in depression. Philos. Trans. R. Soc. Lond., B, Biol. Sci. 368, 20120407.

Harmer, C.J., Goodwin, G.M., Cowen, P.J., 2009. Why do antidepressants take so long to work? A cognitive neuropsychological model of antidepressant drug action. Br J Psychiatry 195, 102–108.

Harrison, A.A., Everitt, B.J., Robbins, T.W., 1997. Doubly dissociable effects of median- and dorsal-raphé lesions on the performance of the five-choice serial reaction time test of attention in rats. Behav. Brain Res. 89, 135–149.

Harrison, A.A., Markou, A., 2001. Serotonergic manipulations both potentiate and reduce brain stimulation reward in rats: involvement of serotonin-1A receptors. J. Pharmacol. Exp. Ther. 297, 316–325.

Harrison, A.A., Parsons, L.H., Koob, G.F., Markou, A., 1999. RU 24969, a 5-HT1A/1B agonist, elevates brain stimulation reward thresholds: an effect reversed by GR 127935, a 5-HT1B/1D antagonist. Psychopharmacology (Berl.) 141, 242–250.

Harrison, B.J., Olver, J.S., Norman, T.R., Burrows, G.D., Wesnes, K.A., Nathan, P.J., 2004. Selective effects of acute serotonin and catecholamine depletion on memory in healthy women. J. Psychopharmacol. (Oxford) 18, 32–40.

Hartlage, S., Alloy, L.B., Vázquez, C., Dykman, B., 1993. Automatic and effortful processing in depression. Psychol Bull 113, 247–278.

Harvey, P.O., Le Bastard, G., Pochon, J.B., Levy, R., Allilaire, J.F., Dubois, B., Fossati, P., 2004. Executive functions and updating of the contents of working memory in unipolar depression. J Psychiatr Res 38, 567–576.

Hayes, D.J., Graham, D.A., Greenshaw, A.J., 2009. Effects of systemic 5-HT(1B) receptor compounds on ventral tegmental area intracranial self-stimulation thresholds in rats. Eur. J. Pharmacol. 604, 74–78.

Hayes, D.J., Greenshaw, A.J., 2011. 5-HT receptors and reward-related behaviour: a review. Neurosci Biobehav Rev 35, 1419–1449.

Hertel, P.T., Hardin, T.S., 1990. Remembering with and without awareness in a depressed mood: evidence of deficits in initiative. J Exp Psychol Gen 119, 45–59.

Hertel, P.T., Rude, S.S., 1991. Depressive deficits in memory: focusing attention improves subsequent recall. J Exp Psychol Gen 120, 301–309.

Hierholzer, R., 2006. Remission rates for depression in STAR*D study. Am J Psychiatry 163, 1293; author reply 1293–1294.

Hinz, R., Selvaraj, S., Murthy, N.V., Bhagwagar, Z., Taylor, M., Cowen, P.J., Grasby, P.M., 2008. Effects of citalopram infusion on the serotonin transporter binding of [11C]DASB in healthy controls. J. Cereb. Blood Flow Metab. 28, 1478–1490.

Hogg, S., Andrews, N., File, S.E., 1994. Contrasting behavioural effects of 8-OH DPAT in the dorsal raphé nucleus and ventral hippocampus. Neuropharmacology 33, 343–348.

Holmes, A.J., Pizzagalli, D.A., 2007. Task feedback effects on conflict monitoring and executive control: relationship to subclinical measures of depression. Emotion 7, 68–76.

Hood, S.D., Hince, D.A., Davies, S.J.C., Argyropoulos, S., Robinson, H., Potokar, J., Nutt, D.J., 2010. Effects of acute tryptophan depletion in serotonin reuptake inhibitor-remitted patients with generalized anxiety disorder. Psychopharmacology (Berl.) 208, 223–232.

Hoplight, B.J., Sandygren, N.A., Neumaier, J.F., 2006. Increased expression of 5-HT1B receptors in rat nucleus accumbens via virally mediated gene transfer increases voluntary alcohol consumption. Alcohol 38, 73–79.

Hrdina, P.D., Demeter, E., Vu, T.B., Sótónyi, P., Palkovits, M., 1993. 5-HT uptake sites and 5-HT2 receptors in brain of antidepressant-free suicide victims/depressives: increase in 5-HT2 sites in cortex and amygdala. Brain Res. 614, 37–44.

Huys, Q.J., Pizzagalli, D.A., Bogdan, R., Dayan, P., 2013. Mapping anhedonia onto reinforcement learning: a behavioural meta-analysis. Biol Mood Anxiety Disord 3, 12.

Huys, Q.J.M., Cools, R., Gölzer, M., Friedel, E., Heinz, A., Dolan, R.J., Dayan, P., 2011. Disentangling the roles of approach, activation and valence in instrumental and pavlovian responding. PLoS Comput. Biol. 7, e1002028.

Huys, Q.J.M., Dayan, P., 2009. A Bayesian formulation of behavioral control. Cognition 113, 314–328.

Huys, Q.J.M., Eshel, N., O'Nions, E., Sheridan, L., Dayan, P., Roiser, J.P., 2012. Bonsai trees in your head: how the pavlovian system sculpts goal-directed choices by pruning decision trees. PLoS Comput. Biol. 8, e1002410.

Insel, T.R., 2006. Beyond efficacy: the STAR*D trial. Am J Psychiatry 163, 5–7.

Jay, T.M., Burette, F., Laroche, S., 1995. NMDA Receptor-dependent Long-term Potentiation in the Hippocampal Afferent Fibre System to the Prefrontal Cortex in the Rat. European Journal of Neuroscience 7, 247–250.

Johnson, A., van der Meer, M.A.A., Redish, A.D., 2007. Integrating hippocampus and striatum in decision-making. Curr. Opin. Neurobiol. 17, 692–697.

Jokinen, J., Nordström, A.-L., Nordström, P., 2007. The relationship between CSF HVA/5-HIAA ratio and suicide intent in suicide attempters. Arch Suicide Res 11, 187–192.

Joyce, J.N., Shane, A., Lexow, N., Winokur, A., Casanova, M.F., Kleinman, J.E., 1993. Serotonin uptake sites and serotonin receptors are altered in the limbic system of schizophrenics. Neuropsychopharmacology 8, 315–336.

Kable, J.W., Glimcher, P.W., 2007. The neural correlates of subjective value during intertemporal choice. Nat. Neurosci. 10, 1625–1633.

Kagan, J., 1966. Reflection--impulsivity: the generality and dynamics of conceptual tempo. J Abnorm Psychol 71, 17–24.

Kalechstein, A.D., De La Garza, R., 2nd, Mahoney, J.J., 3rd, Fantegrossi, W.E., Newton, T.F., 2007. MDMA use and neurocognition: a meta-analytic review. Psychopharmacology (Berl.) 189, 531–537.

Kempton, M.J., Salvador, Z., Munafò, M.R., Geddes, J.R., Simmons, A., Frangou, S., Williams, S.C.R., 2011. Structural neuroimaging studies in major depressive disorder. Meta-analysis and comparison with bipolar disorder. Arch. Gen. Psychiatry 68, 675–690.

Kessler, R.C., 1997. The effects of stressful life events on depression. Annu Rev Psychol 48, 191–214.

Kish, S.J., Furukawa, Y., Ang, L., Vorce, S.P., Kalasinsky, K.S., 2000. Striatal serotonin is depleted in brain of a human MDMA (Ecstasy) user. Neurology 55, 294–296.

Kish, S.J., Lerch, J., Furukawa, Y., Tong, J., McCluskey, T., Wilkins, D., Houle, S., Meyer, J., Mundo, E., Wilson, A.A., Rusjan, P.M., Saint-Cyr, J.A., Guttman, M., Collins, D.L., Shapiro, C., Warsh, J.J., Boileau, I., 2010. Decreased cerebral cortical serotonin transporter binding in ecstasy users: a positron emission tomography/[(11)C]DASB and structural brain imaging study. Brain 133, 1779–1797.

Klaassen, T., Riedel, W.J., Deutz, N.E.P., Van Praag, H.M., 2002. Mood congruent memory bias induced by tryptophan depletion. Psychol Med 32, 167–172.

Klein, D.N., Kotov, R., Bufferd, S.J., 2011. Personality and depression: explanatory models and review of the evidence. Annu Rev Clin Psychol 7, 269–295.

Klein, T.A., Neumann, J., Reuter, M., Hennig, J., von Cramon, D.Y., Ullsperger, M., 2007. Genetically determined differences in learning from errors. Science 318, 1642–1645.

Kleven, M.S., Anthony, E.W., Goldberg, L.I., Woolverton, W.L., 1988. Blockade of the discriminative stimulus effects of cocaine in rhesus monkeys with the D1 dopamine antagonist SCH 23390. Psychopharmacology (Berl.) 95, 427–429.

Klimek, V., Schenck, J.E., Han, H., Stockmeier, C.A., Ordway, G.A., 2002. Dopaminergic abnormalities in amygdaloid nuclei in major depression: a postmortem study. Biol. Psychiatry 52, 740–748.

Knutson, B., Bhanji, J.P., Cooney, R.E., Atlas, L.Y., Gotlib, I.H., 2008. Neural responses to monetary incentives in major depression. Biol. Psychiatry 63, 686–692.

Kong, L., Chen, K., Tang, Y., Wu, F., Driesen, N., Womer, F., Fan, G., Ren, L., Jiang, W., Cao, Y., Blumberg, H.P., Xu, K., Wang, F., 2013. Functional connectivity between the amygdala and prefrontal cortex in medication-naive individuals with major depressive disorder. J Psychiatry Neurosci 38, 120117.

Kotwicki, R., Harvey, P.D., 2013. Systematic Study of Structured Diagnostic Procedures in Outpatient Psychiatric Rehabilitation: A Three-year, Three-cohort Study of the Stability of Psychiatric Diagnoses. Innov Clin Neurosci 10, 14–19.

Kringelbach, M.L., Rolls, E.T., 2004. The functional neuroanatomy of the human orbitofrontal cortex: evidence from neuroimaging and neuropsychology. Progress in Neurobiology 72, 341–372.

Krishnan, V., Nestler, E.J., 2011. Animal Models of Depression: Molecular Perspectives. Curr Top Behav Neurosci 7, 121–147.

Kulin, N.A., Pastuszak, A., Sage, S.R., Schick-Boschetto, B., Spivey, G., Feldkamp, M., Ormond, K., Matsui, D., Stein-Schechman, A.K., Cook, L., Brochu, J., Rieder, M., Koren, G., 1998. Pregnancy outcome following maternal use of the new selective serotonin reuptake inhibitors: a prospective controlled multicenter study. JAMA 279, 609–610.

Laasonen-Balk, T., Viinamäki, H., Kuikka, J.T., Husso-Saastamoinen, M., Lehtonen, J., Tiihonen, J., 2004. 123I-beta-CIT binding and recovery from depression. A six-month follow-up study. Eur Arch Psychiatry Clin Neurosci 254, 152–155.

Lammertsma, A.A., Hume, S.P., 1996. Simplified reference tissue model for PET receptor studies. Neuroimage 4, 153–158.

Larisch, R., Klimke, A., Mayoral, F., Hamacher, K., Herzog, H.R., Vosberg, H., Tosch, M., Gaebel, W., Rivas, F., Coenen, H.H., Müller-Gärtner, H.W., 2001. Disturbance of serotonin

5HT2 receptors in remitted patients suffering from hereditary depressive disorder. Nuklearmedizin 40, 129–134.

Lawson, C., MacLeod, C., 1999. Depression and the interpretation of ambiguity. Behav Res Ther 37, 463–474.

Lawson, C., MacLeod, C., Hammond, G., 2002. Interpretation revealed in the blink of an eye: depressive bias in the resolution of ambiguity. J Abnorm Psychol 111, 321–328.

LeMarquand, D.G., Benkelfat, C., Pihl, R.O., Palmour, R.M., Young, S.N., 1999. Behavioral disinhibition induced by tryptophan depletion in nonalcoholic young men with multigenerational family histories of paternal alcoholism. Am J Psychiatry 156, 1771–1779.

LeMarquand, D.G., Pihl, R.O., Young, S.N., Tremblay, R.E., Séguin, J.R., Palmour, R.M., Benkelfat, C., 1998. Tryptophan depletion, executive functions, and disinhibition in aggressive, adolescent males. Neuropsychopharmacology 19, 333–341.

Leykin, Y., Roberts, C.S., DeRubeis, R.J., 2011. Decision-Making and Depressive Symptomatology. Cognit Ther Res 35, 333–341.

Lindström, M.B., Ryding, E., Bosson, P., Ahnlide, J.-A., Rosén, I., Träskman-Bendz, L., 2004. Impulsivity related to brain serotonin transporter binding capacity in suicide attempters. Eur Neuropsychopharmacol 14, 295–300.

Lisman, J.E., Grace, A.A., 2005. The Hippocampal-VTA Loop: Controlling the Entry of Information into Long-Term Memory. Neuron 46, 703–713.

Liu, Y.P., Wilkinson, L.S., Robbins, T.W., 2004. Effects of acute and chronic buspirone on impulsive choice and efflux of 5-HT and dopamine in hippocampus, nucleus accumbens and prefrontal cortex. Psychopharmacology (Berl.) 173, 175–185.

Lloyd, G.G., Lishman, W.A., 1975. Effect of depression on the speed of recall of pleasant and unpleasant experiences. Psychol Med 5, 173–180.

Lockwood, K.A., Alexopoulos, G.S., van Gorp, W.G., 2002. Executive dysfunction in geriatric depression. Am J Psychiatry 159, 1119–1126.

Logan, G.D., Cowan, W.B., Davis, K.A., 1984. On the ability to inhibit simple and choice reaction time responses: a model and a method. J Exp Psychol Hum Percept Perform 10, 276–291.

Luciana, M., Burgund, E.D., Berman, M., Hanson, K.L., 2001. Effects of tryptophan loading on verbal, spatial and affective working memory functions in healthy adults. J. Psychopharmacol. (Oxford) 15, 219–230.

Macoveanu, J., Rowe, J.B., Hornboll, B., Elliott, R., Paulson, O.B., Knudsen, G.M., Siebner, H.R., 2013. Serotonin 2A receptors contribute to the regulation of risk-averse decisions. NeuroImage 83, 35–44.

MacQueen, G., Frodl, T., 2011. The hippocampus in major depression: evidence for the convergence of the bench and bedside in psychiatric research? Mol Psychiatry 16, 252–264.

Malison, R.T., Price, L.H., Berman, R., van Dyck, C.H., Pelton, G.H., Carpenter, L., Sanacora, G., Owens, M.J., Nemeroff, C.B., Rajeevan, N., Baldwin, R.M., Seibyl, J.P., Innis, R.B., Charney, D.S., 1998. Reduced brain serotonin transporter availability in major depression as measured by [123I]-2 beta-carbomethoxy-3 beta-(4-iodophenyl)tropane and single photon emission computed tomography. Biol. Psychiatry 44, 1090–1098.

Mann, J.J., Waternaux, C., Haas, G.L., Malone, K.M., 1999. Toward a clinical model of suicidal behavior in psychiatric patients. Am J Psychiatry 156, 181–189.

Mar, A.C., Robbins, T.W., 2007. Delay discounting and impulsive choice in the rat. Curr Protoc Neurosci Chapter 8, Unit 8.22.

Mariano, T.Y., Bannerman, D.M., McHugh, S.B., Preston, T.J., Rudebeck, P.H., Rudebeck, S.R., Rawlins, J.N.P., Walton, M.E., Rushworth, M.F.S., Baxter, M.G., Campbell, T.G., 2009. Impulsive choice in hippocampal but not orbitofrontal cortex-lesioned rats on a nonspatial decision-making maze task. Eur. J. Neurosci. 30, 472–484.

Markou, A., Harrison, A.A., Chevrette, J., Hoyer, D., 2005. Paroxetine combined with a 5-HT(1A) receptor antagonist reversed reward deficits observed during amphetamine withdrawal in rats. Psychopharmacology (Berl.) 178, 133–142.

Marteau, T.M., Bekker, H., 1992. The development of a six-item short-form of the state scale of the Spielberger State—Trait Anxiety Inventory (STAI). British Journal of Clinical Psychology 31, 301–306.

Masaki, D., Yokoyama, C., Kinoshita, S., Tsuchida, H., Nakatomi, Y., Yoshimoto, K., Fukui, K., 2006. Relationship between limbic and cortical 5-HT neurotransmission and acquisition and reversal learning in a go/no-go task in rats. Psychopharmacology (Berl.) 189, 249–258.

Massou, J.M., Trichard, C., Attar-Levy, D., Feline, A., Corruble, E., Beaufils, B., Martinot, J.L., 1997. Frontal 5-HT2A receptors studied in depressive patients during chronic treatment by selective serotonin reuptake inhibitors. Psychopharmacology (Berl.) 133, 99–101.

Mathews, A., MacLeod, C., 2005. Cognitive vulnerability to emotional disorders. Annu Rev Clin Psychol 1, 167–195.

Mayberg, H.S., Brannan, S.K., Mahurin, R.K., Jerabek, P.A., Brickman, J.S., Tekell, J.L., Silva, J.A., McGinnis, S., Glass, T.G., Martin, C.C., Fox, P.T., 1997. Cingulate function in depression: a potential predictor of treatment response. Neuroreport 8, 1057–1061.

Mayberg, H.S., Liotti, M., Brannan, S.K., McGinnis, S., Mahurin, R.K., Jerabek, P.A., Silva, J.A., Tekell, J.L., Martin, C.C., Lancaster, J.L., Fox, P.T., 1999. Reciprocal limbic-cortical function and negative mood: converging PET findings in depression and normal sadness. Am J Psychiatry 156, 675–682.

McAllister-Williams, R.H., Massey, A.E., Rugg, M.D., 2002. Effects of tryptophan depletion on brain potential correlates of episodic memory retrieval. Psychopharmacology (Berl.) 160, 434–442.

McCabe, C., Cowen, P.J., Harmer, C.J., 2009. Neural representation of reward in recovered depressed patients. Psychopharmacology (Berl.) 205, 667–677.

McCabe, C., Mishor, Z., Cowen, P.J., Harmer, C.J., 2010. Diminished neural processing of aversive and rewarding stimuli during selective serotonin reuptake inhibitor treatment. Biol. Psychiatry 67, 439–445.

McCann, U.D., Szabo, Z., Scheffel, U., Dannals, R.F., Ricaurte, G.A., 1998. Positron emission tomographic evidence of toxic effect of MDMA (“Ecstasy”) on brain serotonin neurons in human beings. Lancet 352, 1433–1437.

McCrae, R.R., Kurtz, J.E., Yamagata, S., Terracciano, A., 2011. Internal Consistency, Retest Reliability, and their Implications For Personality Scale Validity. Pers Soc Psychol Rev 15, 28–50.

McFarland, B.R., Klein, D.N., 2009. Emotional reactivity in depression: diminished responsiveness to anticipated reward but not to anticipated punishment or to nonreward or avoidance. Depress Anxiety 26, 117–122.

McKeith, I.G., Marshall, E.F., Ferrier, I.N., Armstrong, M.M., Kennedy, W.N., Perry, R.H., Perry, E.K., Eccleston, D., 1987. 5-HT receptor binding in post-mortem brain from patients with affective disorder. J Affect Disord 13, 67–74.

McLean, A., Rubinsztein, J.S., Robbins, T.W., Sahakian, B.J., 2004. The effects of tyrosine depletion in normal healthy volunteers: implications for unipolar depression. Psychopharmacology (Berl.) 171, 286–297.

Meeter, M., Talamini, L., Schmitt, J.A.J., Riedel, W.J., 2006. Effects of 5-HT on memory and the hippocampus: model and data. Neuropsychopharmacology 31, 712–720.

Meltzer, C.C., Price, J.C., Mathis, C.A., Greer, P.J., Cantwell, M.N., Houck, P.R., Mulsant, B.H., Ben-Eliezer, D., Lopresti, B., DeKosky, S.T., Reynolds, C.F., 3rd, 1999. PET imaging of serotonin type 2A receptors in late-life neuropsychiatric disorders. Am J Psychiatry 156, 1871–1878.

Mendelsohn, D., Riedel, W.J., Sambeth, A., 2009. Effects of acute tryptophan depletion on memory, attention and executive functions: a systematic review. Neurosci Biobehav Rev 33, 926–952.

Merriam, E.P., Thase, M.E., Haas, G.L., Keshavan, M.S., Sweeney, J.A., 1999. Prefrontal cortical dysfunction in depression determined by Wisconsin Card Sorting Test performance. Am J Psychiatry 156, 780–782.

Messa, C., Colombo, C., Moresco, R.M., Gobbo, C., Galli, L., Lucignani, G., Gilardi, M.C., Rizzo, G., Smeraldi, E., Zanardi, R., Artigas, F., Fazio, F., 2003. 5-HT(2A) receptor binding is reduced in drug-naive and unchanged in SSRI-responder depressed patients compared to healthy controls: a PET study. Psychopharmacology (Berl.) 167, 72–78.

Meyer, J.H., Kapur, S., Eisfeld, B., Brown, G.M., Houle, S., DaSilva, J., Wilson, A.A., Rafi-Tari, S., Mayberg, H.S., Kennedy, S.H., 2001. The effect of paroxetine on 5-HT(2A) receptors in depression: an [(18)F]setoperone PET imaging study. Am J Psychiatry 158, 78–85.

Meyer, J.H., Kapur, S., Houle, S., DaSilva, J., Owczarek, B., Brown, G.M., Wilson, A.A., Kennedy, S.H., 1999. Prefrontal cortex 5-HT2 receptors in depression: an [18F]setoperone PET imaging study. Am J Psychiatry 156, 1029–1034.

Meyer, J.H., McMain, S., Kennedy, S.H., Korman, L., Brown, G.M., DaSilva, J.N., Wilson, A.A., Blak, T., Eynan-Harvey, R., Goulding, V.S., Houle, S., Links, P., 2003. Dysfunctional attitudes and 5-HT2 receptors during depression and self-harm. Am J Psychiatry 160, 90–99.

Milak, M.S., Severance, A.J., Prabhakaran, J., Kumar, J.S.D., Majo, V.J., Ogden, R.T., Mann, J.J., Parsey, R.V., 2011. In vivo serotonin-sensitive binding of [11C]CUMI-101: a serotonin 1A receptor agonist positron emission tomography radiotracer. J. Cereb. Blood Flow Metab. 31, 243–249.

Miller, E., Lewis, P., 1977. Recognition memory in elderly patients with depression and dementia: a signal detection analysis. J Abnorm Psychol 86, 84–86.

Miyazaki, K.W., Miyazaki, K., Doya, K., 2012. Activation of dorsal raphe serotonin neurons is necessary for waiting for delayed rewards. J. Neurosci. 32, 10451–10457.

Mobini, S., Chiang, T.J., Ho, M.Y., Bradshaw, C.M., Szabadi, E., 2000. Effects of central 5-hydroxytryptamine depletion on sensitivity to delayed and probabilistic reinforcement. Psychopharmacology (Berl.) 152, 390–397.

Moja, E.A., Cipolla, P., Castoldi, D., Tofanetti, O., 1989. Dose-response decrease in plasma tryptophan and in brain tryptophan and serotonin after tryptophan-free amino acid mixtures in rats. Life Sci. 44, 971–976.

Mondelli, V., Gianotti, L., Picu, A., Abbate Daga, G., Giordano, R., Berardelli, R., Pariante, C.M., Fassino, S., Ghigo, E., Arvat, E., 2006. Neuroendocrine effects of citalopram infusion in anorexia nervosa. Psychoneuroendocrinology 31, 1139–1148.

Montague, P.R., Dayan, P., Person, C., Sejnowski, T.J., 1995. Bee foraging in uncertain environments using predictive hebbian learning. Nature 377, 725–728.

Montgomery, A.J., McTavish, S.F.B., Cowen, P.J., Grasby, P.M., 2003. Reduction of brain dopamine concentration with dietary tyrosine plus phenylalanine depletion: an [11C]raclopride PET study. Am J Psychiatry 160, 1887–1889.

Moore, P., Landolt, H.P., Seifritz, E., Clark, C., Bhatti, T., Kelsoe, J., Rapaport, M., Gillin, J.C., 2000. Clinical and physiological consequences of rapid tryptophan depletion. Neuropsychopharmacology 23, 601–622.

Morgan, M.J., McFie, L., Fleetwood, H., Robinson, J.A., 2002. Ecstasy (MDMA): are the psychological problems associated with its use reversed by prolonged abstinence? Psychopharmacology (Berl.) 159, 294–303.

Moritz, S., Birkner, C., Kloss, M., Jahn, H., Hand, I., Haasen, C., Krausz, M., 2002. Executive functioning in obsessive-compulsive disorder, unipolar depression, and schizophrenia. Arch Clin Neuropsychol 17, 477–483.

Morris, J.S., Smith, K.A., Cowen, P.J., Friston, K.J., Dolan, R.J., 1999. Covariation of activity in habenula and dorsal raphé nuclei following tryptophan depletion. Neuroimage 10, 163–172.

Moser, P.C., Moran, P.M., Frank, R.A., Kehne, J.H., 1996. Reversal of amphetamine-induced behaviours by MDL 100,907, a selective 5-HT2A antagonist. Behav. Brain Res. 73, 163–167.

Moses-Kolko, E.L., Berga, S.L., Greer, P.J., Smith, G., Cidis Meltzer, C., Drevets, W.C., 2003. Widespread increases of cortical serotonin type 2A receptor availability after hormone therapy in euthymic postmenopausal women. Fertil. Steril. 80, 554–559.

Moses-Kolko, E.L., Wisner, K.L., Price, J.C., Berga, S.L., Drevets, W.C., Hanusa, B.H., Loucks, T.L., Meltzer, C.C., 2008. Serotonin 1A receptor reductions in postpartum depression: a positron emission tomography study. Fertil. Steril. 89, 685–692.

Mosienko, V., Bert, B., Beis, D., Matthes, S., Fink, H., Bader, M., Alenina, N., 2012. Exaggerated aggression and decreased anxiety in mice deficient in brain serotonin. Transl Psychiatry 2, e122.

Murphy, F.C., Michael, A., Robbins, T.W., Sahakian, B.J., 2003. Neuropsychological impairment in patients with major depressive disorder: the effects of feedback on task performance. Psychol Med 33, 455–467.

Murphy, F.C., Rubinsztein, J.S., Michael, A., Rogers, R.D., Robbins, T.W., Paykel, E.S., Sahakian, B.J., 2001. Decision-making cognition in mania and depression. Psychol Med 31, 679–693.

Murphy, F.C., Smith, K.A., Cowen, P.J., Robbins, T.W., Sahakian, B.J., 2002. The effects of tryptophan depletion on cognitive and affective processing in healthy volunteers. Psychopharmacology (Berl.) 163, 42–53.

Nelson, H.E., 1976. A modified card sorting test sensitive to frontal lobe defects. Cortex 12, 313–324.

Nelson, J.C., 2006. The STAR*D study: a four-course meal that leaves us wanting more. Am J Psychiatry 163, 1864–1866.

Neumeister, A., Nugent, A.C., Waldeck, T., Geraci, M., Schwarz, M., Bonne, O., Bain, E.E., Luckenbaugh, D.A., Herscovitch, P., Charney, D.S., Drevets, W.C., 2004. Neural and behavioral responses to tryptophan depletion in unmedicated patients with remitted major depressive disorder and controls. Arch. Gen. Psychiatry 61, 765–773.

Neumeister, A., Pirker, W., Willeit, M., Praschak-Rieder, N., Asenbaum, S., Brücke, T., Kasper, S., 2000. Seasonal variation of availability of serotonin transporter binding sites in healthy female subjects as measured by [123I]-2 beta-carbomethoxy-3 beta-(4-iodophenyl)tropane and single photon emission computed tomography. Biol. Psychiatry 47, 158–160.

Nezu, A.M., Perri, M.G., 1989. Social problem-solving therapy for unipolar depression: an initial dismantling investigation. J Consult Clin Psychol 57, 408–413.

Nishizawa, S., Benkelfat, C., Young, S.N., Leyton, M., Mzengeza, S., de Montigny, C., Blier, P., Diksic, M., 1997. Differences between males and females in rates of serotonin synthesis in human brain. Proc. Natl. Acad. Sci. U.S.A. 94, 5308–5313.

Nordström, P., Samuelsson, M., Asberg, M., Träskman-Bendz, L., Aberg-Wistedt, A., Nordin, C., Bertilsson, L., 1994. CSF 5-HIAA predicts suicide risk after attempted suicide. Suicide Life Threat Behav 24, 1–9.

O'Reardon, J.P., Chopra, M.P., Bergan, A., Gallop, R., DeRubeis, R.J., Crits-Christoph, P., 2004. Response to tryptophan depletion in major depression treated with either cognitive

therapy or selective serotonin reuptake inhibitor antidepressants. Biol. Psychiatry 55, 957–959.

O'shea, E., Orio, L., Escobedo, I., Sanchez, V., Camarero, J., Green, A.R., Colado, M.I., 2006. MDMA-induced neurotoxicity: long-term effects on 5-HT biosynthesis and the influence of ambient temperature. Br J Pharmacol 148, 778–785.

Owen, A.M., Downes, J.J., Sahakian, B.J., Polkey, C.E., Robbins, T.W., 1990. Planning and spatial working memory following frontal lobe lesions in man. Neuropsychologia 28, 1021–1034.

Owens, M.J., Nemeroff, C.B., 1994. Role of serotonin in the pathophysiology of depression: focus on the serotonin transporter. Clin. Chem. 40, 288–295.

Palacios, J.M., Waeber, C., Hoyer, D., Mengod, G., 1990. Distribution of serotonin receptors. Ann. N. Y. Acad. Sci. 600, 36–52.

Park, S.B., Coull, J.T., McShane, R.H., Young, A.H., Sahakian, B.J., Robbins, T.W., Cowen, P.J., 1994. Tryptophan depletion in normal volunteers produces selective impairments in learning and memory. Neuropharmacology 33, 575–588.

Parrott, A.C., 2000. Human research on MDMA (3,4-methylene- dioxymethamphetamine) neurotoxicity: cognitive and behavioural indices of change. Neuropsychobiology 42, 17–24.

Parsey, R.V., Ogden, R.T., Miller, J.M., Tin, A., Hesselgrave, N., Goldstein, E., Mikhno, A., Milak, M., Zanderigo, F., Sullivan, G.M., Oquendo, M.A., Mann, J.J., 2010. Higher serotonin 1A binding in a second major depression cohort: modeling and reference region considerations. Biol. Psychiatry 68, 170–178.

Parsey, R.V., Oquendo, M.A., Ogden, R.T., Olvet, D.M., Simpson, N., Huang, Y.-Y., Van Heertum, R.L., Arango, V., Mann, J.J., 2006. Altered serotonin 1A binding in major depression: a [carbonyl-C-11]WAY100635 positron emission tomography study. Biol. Psychiatry 59, 106–113.

Parsons, L.H., Weiss, F., Koob, G.F., 1998. Serotonin1B receptor stimulation enhances cocaine reinforcement. J. Neurosci. 18, 10078–10089.

Pazos, A., Probst, A., Palacios, J.M., 1987. Serotonin receptors in the human brain--III. Autoradiographic mapping of serotonin-1 receptors. Neuroscience 21, 97–122.

Peltier, R., Schenk, S., 1993. Effects of serotonergic manipulations on cocaine self-administration in rats. Psychopharmacology (Berl.) 110, 390–394.

Perona, M.T.G., Waters, S., Hall, F.S., Sora, I., Lesch, K.-P., Murphy, D.L., Caron, M., Uhl, G.R., 2008. Animal models of depression in dopamine, serotonin, and norepinephrine transporter knockout mice: prominent effects of dopamine transporter deletions. Behav Pharmacol 19, 566–574.

Peters, J., Büchel, C., 2010. Episodic future thinking reduces reward delay discounting through an enhancement of prefrontal-mediotemporal interactions. Neuron 66, 138–148.

Petty, F., Davis, L.L., Kabel, D., Kramer, G.L., 1996. Serotonin dysfunction disorders: a behavioral neurochemistry perspective. J Clin Psychiatry 57 Suppl 8, 11–16.

Petty, F., Kramer, G., Wilson, L., Jordan, S., 1994. In vivo serotonin release and learned helplessness. Psychiatry Res 52, 285–293.

Pine, A., Seymour, B., Roiser, J.P., Bossaerts, P., Friston, K.J., Curran, H.V., Dolan, R.J., 2009. Encoding of marginal utility across time in the human brain. J. Neurosci. 29, 9575–9581.

Piñeyro, G., Blier, P., 1999. Autoregulation of Serotonin Neurons: Role in Antidepressant Drug Action. Pharmacol Rev 51, 533–591.

Pizzagalli, D.A., Holmes, A.J., Dillon, D.G., Goetz, E.L., Birk, J.L., Bogdan, R., Dougherty, D.D., Iosifescu, D.V., Rauch, S.L., Fava, M., 2009. Reduced caudate and nucleus accumbens response to rewards in unmedicated individuals with major depressive disorder. Am J Psychiatry 166, 702–710.

Pizzagalli, D.A., Jahn, A.L., O'Shea, J.P., 2005. Toward an objective characterization of an anhedonic phenotype: a signal-detection approach. Biol. Psychiatry 57, 319–327.

Porter, R.J., Lunn, B.S., Walker, L.L., Gray, J.M., Ballard, C.G., O'Brien, J.T., 2000. Cognitive deficit induced by acute tryptophan depletion in patients with Alzheimer's disease. Am J Psychiatry 157, 638–640.

Porter, R.J., Phipps, A.J., Gallagher, P., Scott, A., Stevenson, P.S., O'Brien, J.T., 2005. Effects of acute tryptophan depletion on mood and cognitive functioning in older recovered depressed subjects. Am J Geriatr Psychiatry 13, 607–615.

Purcell, R., Maruff, P., Kyrios, M., Pantelis, C., 1997. Neuropsychological function in young patients with unipolar major depression. Psychol Med 27, 1277–1285.

Quednow, B.B., Jessen, F., Kuhn, K.-U., Maier, W., Daum, I., Wagner, M., 2006. Memory deficits in abstinent MDMA (ecstasy) users: neuropsychological evidence of frontal dysfunction. J. Psychopharmacol. (Oxford) 20, 373–384.

Quednow, B.B., Kühn, K.-U., Hoppe, C., Westheide, J., Maier, W., Daum, I., Wagner, M., 2007. Elevated impulsivity and impaired decision-making cognition in heavy users of MDMA ("Ecstasy"). Psychopharmacology (Berl.) 189, 517–530.

R, J., 1935. Studies of interference in serial verbal reactions. Journal of Experimental Psychology 18, 643–662.

Raichle, M.E., MacLeod, A.M., Snyder, A.Z., Powers, W.J., Gusnard, D.A., Shulman, G.L., 2001. A default mode of brain function. Proc. Natl. Acad. Sci. U.S.A. 98, 676–682.

Raj, V., Liang, H.C., Woodward, N.D., Bauernfeind, A.L., Lee, J., Dietrich, M.S., Park, S., Cowan, R.L., 2010. MDMA (ecstasy) use is associated with reduced BOLD signal change during semantic recognition in abstinent human polydrug users: a preliminary fMRI study. J. Psychopharmacol. (Oxford) 24, 187–201.

Rawal, A., Collishaw, S., Thapar, A., Rice, F., 2013. "The risks of playing it safe": a prospective longitudinal study of response to reward in the adolescent offspring of depressed parents. Psychol Med 43, 27–38.

Rawlins, J.N., Feldon, J., Butt, S., 1985. The effects of delaying reward on choice preference in rats with hippocampal or selective septal lesions. Behav. Brain Res. 15, 191–203.

Remy, S.M., Schreiber, R., Dalmus, M., De Vry, J., 1996. Somatodendritic 5-HT1A receptors are critically involved in the anxiolytic effects of 8-OH-DPAT. Psychopharmacology (Berl.) 125, 89–91.

Reneman, L., Lavalaye, J., Schmand, B., de Wolff, F.A., van den Brink, W., den Heeten, G.J., Booij, J., 2001. Cortical serotonin transporter density and verbal memory in individuals who stopped using 3,4-methylenedioxymethamphetamine (MDMA or "ecstasy"): preliminary findings. Arch. Gen. Psychiatry 58, 901–906.

Ricaurte, G.A., Finnegan, K.T., Irwin, I., Langston, J.W., 1990. Aminergic metabolites in cerebrospinal fluid of humans previously exposed to MDMA: preliminary observations. Ann. N. Y. Acad. Sci. 600, 699–708; discussion 708–710.

Rice, F., Harold, G., Thapar, A., 2002. The genetic aetiology of childhood depression: a review. J Child Psychol Psychiatry 43, 65–79.

Richard, J., 1969. Reactions toward the source of stimulation. Journal of Experimental Psychology 81, 174–176.

Riedel, G., Micheau, J., Lam, A.G., Roloff, E.L., Martin, S.J., Bridge, H., de Hoz, L., Poeschel, B., McCulloch, J., Morris, R.G., 1999. Reversible neural inactivation reveals hippocampal participation in several memory processes. Nat. Neurosci. 2, 898–905.

Riedel, W.J., 2004. Cognitive changes after acute tryptophan depletion: what can they tell us? Psychol Med 34, 3–8.

Riedel, W.J., Klaassen, T., Deutz, N.E., van Someren, A., van Praag, H.M., 1999. Tryptophan depletion in normal volunteers produces selective impairment in memory consolidation. Psychopharmacology (Berl.) 141, 362–369.

Riedel, W.J., Klaassen, T., Griez, E., Honig, A., Menheere, P.P.C.A., van Praag, H.M., 2002. Dissociable hormonal, cognitive and mood responses to neuroendocrine challenge: evidence for receptor-specific serotonergic dysregulation in depressed mood. Neuropsychopharmacology 26, 358–367.

Robbins, T.W., Everitt, B.J., 1996. Neurobehavioural mechanisms of reward and motivation. Curr. Opin. Neurobiol. 6, 228–236.

Robinson, O.J., Cools, R., Sahakian, B.J., 2012. Tryptophan depletion disinhibits punishment but not reward prediction: implications for resilience. Psychopharmacology (Berl.) 219, 599–605.

Robinson, Oliver J, Frank, M.J., Sahakian, B.J., Cools, R., 2010. Dissociable responses to punishment in distinct striatal regions during reversal learning. Neuroimage 51, 1459–1467.

Robinson, Oliver J., Frank, M.J., Sahakian, B.J., Cools, R., 2010. Dissociable responses to punishment in distinct striatal regions during reversal learning. Neuroimage 51, 1459–1467.

Robinson, Oliver J, Overstreet, C., Allen, P.S., Letkiewicz, A., Vytal, K., Pine, D.S., Grillon, C., 2013. The role of serotonin in the neurocircuitry of negative affective bias: serotonergic modulation of the dorsal medial prefrontal-amygdala "aversive amplification" circuit. Neuroimage 78, 217–223.

Robinson, Oliver J., Overstreet, C., Charney, D.R., Vytal, K., Grillon, C., 2013. Stress increases aversive prediction error signal in the ventral striatum. PNAS 110, 4129–4133.

Roediger, H.L., 3rd, McDermott, K.B., 1992. Depression and implicit memory: a commentary. J Abnorm Psychol 101, 587–591.

Rogers, R.D., Everitt, B.J., Baldacchino, A., Blackshaw, A.J., Swainson, R., Wynne, K., Baker, N.B., Hunter, J., Carthy, T., Booker, E., London, M., Deakin, J.F., Sahakian, B.J., Robbins, T.W., 1999a. Dissociable deficits in the decision-making cognition of chronic amphetamine abusers, opiate abusers, patients with focal damage to prefrontal cortex, and tryptophan-depleted normal volunteers: evidence for monoaminergic mechanisms. Neuropsychopharmacology 20, 322–339.

Rogers, R.D., Owen, A.M., Middleton, H.C., Williams, E.J., Pickard, J.D., Sahakian, B.J., Robbins, T.W., 1999b. Choosing between small, likely rewards and large, unlikely rewards activates inferior and orbital prefrontal cortex. J. Neurosci. 19, 9029–9038.

Rogers, R.D., Tunbridge, E.M., Bhagwagar, Z., Drevets, W.C., Sahakian, B.J., Carter, C.S., 2003. Tryptophan depletion alters the decision-making of healthy volunteers through altered processing of reward cues. Neuropsychopharmacology 28, 153–162.

Roggenbach, J., Müller-Oerlinghausen, B., Franke, L., 2002. Suicidality, impulsivity and aggression--is there a link to 5HIAA concentration in the cerebrospinal fluid? Psychiatry Res 113, 193–206.

Roiser, Jonathan P, Blackwell, A.D., Cools, R., Clark, L., Rubinsztein, D.C., Robbins, T.W., Sahakian, B.J., 2006a. Serotonin transporter polymorphism mediates vulnerability to loss of incentive motivation following acute tryptophan depletion. Neuropsychopharmacology 31, 2264–2272.

Roiser, Jonathan P., Blackwell, A.D., Cools, R., Clark, L., Rubinsztein, D.C., Robbins, T.W., Sahakian, B.J., 2006. Serotonin Transporter Polymorphism Mediates Vulnerability to Loss of

Incentive Motivation Following Acute Tryptophan Depletion. Neuropsychopharmacology 31, 2264–2272.

Roiser, J.P., Levy, J., Fromm, S.J., Nugent, A.C., Talagala, S.L., Hasler, G., Henn, F.A., Sahakian, B.J., Drevets, W.C., 2009. The effects of tryptophan depletion on neural responses to emotional words in remitted depression. Biol. Psychiatry 66, 441–450.

Roiser, J.P., Levy, J., Fromm, S.J., Wang, H., Hasler, G., Sahakian, B.J., Drevets, W.C., 2008. The effect of acute tryptophan depletion on the neural correlates of emotional processing in healthy volunteers. Neuropsychopharmacology 33, 1992–2006.

Roiser, J.P., Müller, U., Clark, L., Sahakian, B.J., 2007. The effects of acute tryptophan depletion and serotonin transporter polymorphism on emotional processing in memory and attention. The International Journal of Neuropsychopharmacology 10, 449–461.

Roiser, Jonathan P, Rogers, R.D., Cook, L.J., Sahakian, B.J., 2006b. The effect of polymorphism at the serotonin transporter gene on decision-making, memory and executive function in ecstasy users and controls. Psychopharmacology (Berl.) 188, 213–227.

Rose, E.J., Ebmeier, K.P., 2006. Pattern of impaired working memory during major depression. J Affect Disord 90, 149–161.

Rubia, K., Lee, F., Cleare, A.J., Tunstall, N., Fu, C.H.Y., Brammer, M., McGuire, P., 2005. Tryptophan depletion reduces right inferior prefrontal activation during response inhibition in fast, event-related fMRI. Psychopharmacology (Berl.) 179, 791–803.

Rudnick, G., Wall, S.C., 1992. The molecular mechanism of “ecstasy” [3,4-methylenedioxy-methamphetamine (MDMA)]: serotonin transporters are targets for MDMA-induced serotonin release. Proc. Natl. Acad. Sci. U.S.A. 89, 1817–1821.

Ruhé, H.G., Booij, J., Reitsma, J.B., Schene, A.H., 2009. Serotonin transporter binding with [123I]beta-CIT SPECT in major depressive disorder versus controls: effect of season and gender. Eur. J. Nucl. Med. Mol. Imaging 36, 841–849.

Ruhé, H.G., Mason, N.S., Schene, A.H., 2007. Mood is indirectly related to serotonin, norepinephrine and dopamine levels in humans: a meta-analysis of monoamine depletion studies. Mol. Psychiatry 12, 331–359.

Ryding, E., Ahnlide, J.-A., Lindström, M., Rosén, I., Träskman-Bendz, L., 2006. Regional brain serotonin and dopamine transporter binding capacity in suicide attempters relate to impulsiveness and mental energy. Psychiatry Res 148, 195–203.

Sahakian, B.J., Morris, R.G., Evenden, J.L., Heald, A., Levy, R., Philpot, M., Robbins, T.W., 1988. A comparative study of visuospatial memory and learning in Alzheimer-type dementia and Parkinson's disease. Brain 111 (Pt 3), 695–718.

Salomon, R.M., Karageorgiou, J., Dietrich, M.S., McLellan, J.Y., Charboneau, E.J., Blackford, J.U., Cowan, R.L., 2012. MDMA (Ecstasy) association with impaired fMRI BOLD thalamic coherence and functional connectivity. Drug Alcohol Depend 120, 41–47.

Sambeth, A., Riedel, W.J., Tillie, D.E., Blokland, A., Postma, A., Schmitt, J.A.J., 2009. Memory impairments in humans after acute tryptophan depletion using a novel gelatin-based protein drink. J. Psychopharmacol. (Oxford) 23, 56–64.

Samuelsson, M., Jokinen, J., Nordström, A.-L., Nordström, P., 2006. CSF 5-HIAA, suicide intent and hopelessness in the prediction of early suicide in male high-risk suicide attempters. Acta Psychiatr Scand 113, 44–47.

Santarelli, L., Saxe, M., Gross, C., Surget, A., Battaglia, F., Dulawa, S., Weisstaub, N., Lee, J., Duman, R., Arancio, O., Belzung, C., Hen, R., 2003. Requirement of Hippocampal Neurogenesis for the Behavioral Effects of Antidepressants. Science 301, 805–809.

Sargent, P.A., Kjaer, K.H., Bench, C.J., Rabiner, E.A., Messa, C., Meyer, J., Gunn, R.N., Grasby, P.M., Cowen, P.J., 2000. Brain serotonin1A receptor binding measured by positron emission tomography with [11C]WAY-100635: effects of depression and antidepressant treatment. Arch. Gen. Psychiatry 57, 174–180.

Scerbo, A., Raine, A., O'Brien, M., Chan, C.J., Rhee, C., Smiley, N., 1990. Reward dominance and passive avoidance learning in adolescent psychopaths. J Abnorm Child Psychol 18, 451–463.

Schacter, D.L., Addis, D.R., 2009. On the nature of medial temporal lobe contributions to the constructive simulation of future events. Philos. Trans. R. Soc. Lond., B, Biol. Sci. 364, 1245–1253.

Scheurich, A., Fellgiebel, A., Schermuly, I., Bauer, S., Wölfges, R., Müller, M.J., 2008. Experimental evidence for a motivational origin of cognitive impairment in major depression. Psychol Med 38, 237–246.

Schmitt, J.A., Jorissen, B.L., Sobczak, S., van Boxtel, M.P., Hogervorst, E., Deutz, N.E., Riedel, W.J., 2000. Tryptophan depletion impairs memory consolidation but improves focussed attention in healthy young volunteers. J. Psychopharmacol. (Oxford) 14, 21–29.

Schmitz, A., Kirsch, P., Reuter, M., Alexander, N., Kozyra, E., Kuepper, Y., Osinsky, R., Hennig, J., 2009. The 5-HT1A C(-1019)G polymorphism, personality and electrodermal reactivity in a reward/punishment paradigm. Int. J. Neuropsychopharmacol. 12, 383–392.

Scholes, K.E., Harrison, B.J., O'Neill, B.V., Leung, S., Croft, R.J., Pipingas, A., Phan, K.L., Nathan, P.J., 2007. Acute serotonin and dopamine depletion improves attentional control: findings from the stroop task. Neuropsychopharmacology 32, 1600–1610.

Schultz, W., Dickinson, A., 2000. Neuronal coding of prediction errors. Annu. Rev. Neurosci. 23, 473–500.

Schultz, W., Tremblay, L., Hollerman, J.R., 2000. Reward processing in primate orbitofrontal cortex and basal ganglia. Cereb. Cortex 10, 272–284.

Schweighofer, N., Bertin, M., Shishida, K., Okamoto, Y., Tanaka, S.C., Yamawaki, S., Doya, K., 2008. Low-serotonin levels increase delayed reward discounting in humans. J. Neurosci. 28, 4528–4532.

Selvaraj, S., Turkheimer, F., Rosso, L., Faulkner, P., Mouchlianitis, E., Roiser, J.P., McGuire, P., Cowen, P.J., Howes, O., 2012. Measuring endogenous changes in serotonergic neurotransmission in humans: a [11C]CUMI-101 PET challenge study. Mol. Psychiatry 17, 1254–1260.

Seymour, B., Daw, N.D., Roiser, J.P., Dayan, P., Dolan, R., 2012. Serotonin selectively modulates reward value in human decision-making. J. Neurosci. 32, 5833–5842.

Shah, P.J., O'Carroll, R.E., Rogers, A., Moffoot, A.P., Ebmeier, K.P., 1999. Abnormal response to negative feedback in depression. Psychol Med 29, 63–72.

Shallice, T., 1982. Specific impairments of planning. Philos. Trans. R. Soc. Lond., B, Biol. Sci. 298, 199–209.

Sheehan, D.V., Lecrubier, Y., Sheehan, K.H., Amorim, P., Janavs, J., Weiller, E., Hergueta, T., Baker, R., Dunbar, G.C., 1998. The Mini-International Neuropsychiatric Interview (M.I.N.I.): the development and validation of a structured diagnostic psychiatric interview for DSM-IV and ICD-10. J Clin Psychiatry 59 Suppl 20, 22–33;quiz 34–57.

Sheline, Y.I., Price, J.L., Yan, Z., Mintun, M.A., 2010. Resting-state functional MRI in depression unmasks increased connectivity between networks via the dorsal nexus. Proc. Natl. Acad. Sci. U.S.A. 107, 11020–11025.

Shelton, R.C., Sanders-Bush, E., Manier, D.H., Lewis, D.A., 2009. Elevated 5-HT 2A receptors in postmortem prefrontal cortex in major depression is associated with reduced activity of protein kinase A. Neuroscience 158, 1406–1415.

Shrestha, S., Hirvonen, J., Hines, C.S., Henter, I.D., Svenningsson, P., Pike, V.W., Innis, R.B., 2012. Serotonin-1A receptors in major depression quantified using PET: controversies, confounds, and recommendations. Neuroimage 59, 3243–3251.

Simon, J.R., 1969. Reactions toward the source of stimulation. J Exp Psychol 81, 174–176.

Smith, K.A., Morris, J.S., Friston, K.J., Cowen, P.J., Dolan, R.J., 1999. Brain mechanisms associated with depressive relapse and associated cognitive impairment following acute tryptophan depletion. Br J Psychiatry 174, 525–529.

Smoski, M.J., Lynch, T.R., Rosenthal, M.Z., Cheavens, J.S., Chapman, A.L., Krishnan, R.R., 2008. Decision-making and risk aversion among depressive adults. J Behav Ther Exp Psychiatry 39, 567–576.

Sobczak, S., Honig, A., van Duinen, M.A., Maes, M., Riedel, W.J., 2002. Mood, prolactin and cortisol responses following intravenous L-tryptophan challenge: evidence for serotonergic vulnerability in first-degree relatives of bipolar patients. Int. J. Neuropsychopharmacol. 5, 249–254.

Soubrié, P., 1986. Reconciling the role of central serotonin neurons in human and animal behavior. Behavioral and Brain Sciences 9, 319–335.

Stancampiano, R., Melis, F., Sarais, L., Cocco, S., Cugusi, C., Fadda, F., 1997. Acute administration of a tryptophan-free amino acid mixture decreases 5-HT release in rat hippocampus in vivo. Am. J. Physiol. 272, R991–994.

Stanley, B., Molcho, A., Stanley, M., Winchel, R., Gameroff, M.J., Parsons, B., Mann, J.J., 2000. Association of aggressive behavior with altered serotonergic function in patients who are not suicidal. Am J Psychiatry 157, 609–614.

Steele, J.D., Kumar, P., Ebmeier, K.P., 2007. Blunted response to feedback information in depressive illness. Brain 130, 2367–2374.

Steele, J.D., Meyer, M., Ebmeier, K.P., 2004. Neural predictive error signal correlates with depressive illness severity in a game paradigm. Neuroimage 23, 269–280.

Steffens, D.C., Krishnan, K.R., Helms, M.J., 1997. Are SSRIs better than TCAs? Comparison of SSRIs and TCAs: a meta-analysis. Depress Anxiety 6, 10–18.

Steffens, D.C., Wagner, H.R., Levy, R.M., Horn, K.A., Krishnan, K.R., 2001. Performance feedback deficit in geriatric depression. Biol. Psychiatry 50, 358–363.

Steinfels, G.F., Heym, J., Strecker, R.E., Jacobs, B.L., 1983. Response of dopaminergic neurons in cat to auditory stimuli presented across the sleep-waking cycle. Brain Res. 277, 150–154.

Stewart, M.E., Deary, I.J., Ebmeier, K.P., 2002. Neuroticism as a predictor of mood change: the effects of tryptophan depletion. Br J Psychiatry 181, 242–247.

Stockmeier, C.A., 2003. Involvement of serotonin in depression: evidence from postmortem and imaging studies of serotonin receptors and the serotonin transporter. J Psychiatr Res 37, 357–373.

Stockmeier, C.A., Shapiro, L.A., Dilley, G.E., Kolli, T.N., Friedman, L., Rajkowska, G., 1998. Increase in serotonin-1A autoreceptors in the midbrain of suicide victims with major depression-postmortem evidence for decreased serotonin activity. J. Neurosci. 18, 7394–7401.

Stone, D.M., Stahl, D.C., Hanson, G.R., Gibb, J.W., 1986. The effects of 3,4-methylenedioxymethamphetamine (MDMA) and 3,4-methylenedioxyamphetamine (MDA) on monoaminergic systems in the rat brain. Eur. J. Pharmacol. 128, 41–48.

Storch, E.A., Roberti, J.W., Roth, D.A., 2004. Factor structure, concurrent validity, and internal consistency of the Beck Depression Inventory-Second Edition in a sample of college students. Depress Anxiety 19, 187–189.

Studholme, C., Hill, D.L., Hawkes, D.J., 1997. Automated three-dimensional registration of magnetic resonance and positron emission tomography brain images by multiresolution optimization of voxel similarity measures. Med Phys 24, 25–35.

Suri, R., Altshuler, L.A., Mintz, J., 2004. Depression and the decision to abort. Am J Psychiatry 161, 1502.

Takahashi, H., 2012. Monoamines and assessment of risks. Curr. Opin. Neurobiol. 22, 1062–1067.

Takahashi, H., Matsui, H., Camerer, C., Takano, H., Kodaka, F., Ideno, T., Okubo, S., Takemura, K., Arakawa, R., Eguchi, Y., Murai, T., Okubo, Y., Kato, M., Ito, H., Suhara, T., 2010. Dopamine D_1 receptors and nonlinear probability weighting in risky choice. J. Neurosci. 30, 16567–16572.

Takahashi, T., Oono, H., Inoue, T., Boku, S., Kako, Y., Kitaichi, Y., Kusumi, I., Masui, T., Nakagawa, S., Suzuki, K., Tanaka, T., Koyama, T., Radford, M.H.B., 2008. Depressive patients are more impulsive and inconsistent in intertemporal choice behavior for monetary gain and loss than healthy subjects--an analysis based on Tsallis' statistics. Neuro Endocrinol. Lett. 29, 351–358.

Talbot, P.S., Cooper, S.J., 2006. Anterior cingulate and subgenual prefrontal blood flow changes following tryptophan depletion in healthy males. Neuropsychopharmacology 31, 1757–1767.

Talbot, P.S., Watson, D.R., Barrett, S.L., Cooper, S.J., 2006. Rapid tryptophan depletion improves decision-making cognition in healthy humans without affecting reversal learning or set shifting. Neuropsychopharmacology 31, 1519–1525.

Talpos, J.C., Wilkinson, L.S., Robbins, T.W., 2006. A comparison of multiple 5-HT receptors in two tasks measuring impulsivity. J. Psychopharmacol. (Oxford) 20, 47–58.

Tanaka, S.C., Doya, K., Okada, G., Ueda, K., Okamoto, Y., Yamawaki, S., 2004. Prediction of immediate and future rewards differentially recruits cortico-basal ganglia loops. Nat. Neurosci. 7, 887–893.

Tanaka, S.C., Schweighofer, N., Asahi, S., Shishida, K., Okamoto, Y., Yamawaki, S., Doya, K., 2007. Serotonin differentially regulates short- and long-term prediction of rewards in the ventral and dorsal striatum. PLoS ONE 2, e1333.

Thiébot, M.H., Hamon, M., Soubríe, P., 1982. Attenuation of induced-anxiety in rats by chlordiazepoxide: role of raphe dorsalis benzodiazepine binding sites and serotoninergic neurons. Neuroscience 7, 2287–2294.

Thomas, R.M., Hotsenpiller, G., Peterson, D.A., 2007. Acute psychosocial stress reduces cell survival in adult hippocampal neurogenesis without altering proliferation. J. Neurosci. 27, 2734–2743.

Trivedi, M.H., 2004. The Link Between Depression and Physical Symptoms. Prim Care Companion J Clin Psychiatry 6, 12–16.

Turkheimer, F.E., Brett, M., Visvikis, D., Cunningham, V.J., 1999. Multiresolution analysis of emission tomography images in the wavelet domain. J. Cereb. Blood Flow Metab. 19, 1189–1208.

Turkheimer, F.E., Edison, P., Pavese, N., Roncaroli, F., Anderson, A.N., Hammers, A., Gerhard, A., Hinz, R., Tai, Y.F., Brooks, D.J., 2007. Reference and target region modeling of [11C]-(R)-PK11195 brain studies. J. Nucl. Med. 48, 158–167.

Tye, N.C., Everitt, B.J., Iversen, S.D., 1977. 5-Hydroxytryptamine and punishment. Nature 268, 741–743.

Tye, N.C., Iversen, S.D., Green, A.R., 1979. The effects of benzodiazepines and serotonergic manipulations on punished responding. Neuropharmacology 18, 689–695.

Uchida, S., Umeeda, H., Kitamoto, A., Masushige, S., Kida, S., 2007. Chronic reduction in dietary tryptophan leads to a selective impairment of contextual fear memory in mice. Brain Res. 1149, 149–156.

Valdes, I.H., Steinberg, J.L., Narayana, P.A., Kramer, L.A., Dougherty, D.M., Swann, A.C., Barratt, E.S., Moeller, F.G., 2006. Impulsivity and BOLD fMRI activation in MDMA users and healthy control subjects. Psychiatry Res 147, 239–242.

Van der Schaaf, M.E., van Schouwenburg, M.R., Geurts, D.E.M., Schellekens, A.F.A., Buitelaar, J.K., Verkes, R.J., Cools, R., 2012. Establishing the Dopamine Dependency of Human Striatal Signals During Reward and Punishment Reversal Learning. Cereb. Cortex.

Van der Veen, F.M., Evers, E.A.T., van Deursen, J.A., Deutz, N.E.P., Backes, W.H., Schmitt, J.A.J., 2006. Acute tryptophan depletion reduces activation in the right hippocampus during encoding in an episodic memory task. Neuroimage 31, 1188–1196.

Van Donkelaar, E.L., Blokland, A., Ferrington, L., Kelly, P. a. T., Steinbusch, H.W.M., Prickaerts, J., 2011. Mechanism of acute tryptophan depletion: is it only serotonin? Mol Psychiatry 16, 695–713.

Varnäs, K., Halldin, C., Hall, H., 2004. Autoradiographic distribution of serotonin transporters and receptor subtypes in human brain. Hum Brain Mapp 22, 246–260.

Verheyden, S.L., Henry, J.A., Curran, H.V., 2003. Acute, sub-acute and long-term subjective consequences of "ecstasy" (MDMA) consumption in 430 regular users. Hum Psychopharmacol 18, 507–517.

Viard, A., Doeller, C.F., Hartley, T., Bird, C.M., Burgess, N., 2011. Anterior hippocampus and goal-directed spatial decision making. J. Neurosci. 31, 4613–4621.

Videbech, P., Ravnkilde, B., 2004. Hippocampal Volume and Depression: A Meta-Analysis of MRI Studies. Am J Psychiatry 161, 1957–1966.

Völlm, B., Richardson, P., McKie, S., Elliott, R., Deakin, J.F.W., Anderson, I.M., 2006. Serotonergic modulation of neuronal responses to behavioural inhibition and reinforcing stimuli: an fMRI study in healthy volunteers. Eur. J. Neurosci. 23, 552–560.

Wagenmakers, E.-J., Wetzels, R., Borsboom, D., van der Maas, H.L.J., 2011. Why psychologists must change the way they analyze their data: the case of psi: comment on Bem (2011). J Pers Soc Psychol 100, 426–432.

Walderhaug, E., Lunde, H., Nordvik, J.E., Landrø, N.I., Refsum, H., Magnusson, A., 2002. Lowering of serotonin by rapid tryptophan depletion increases impulsiveness in normal individuals. Psychopharmacology (Berl.) 164, 385–391.

Watts, F.N., MacLeod, A.K., Morris, L., 1988. Associations between phenomenal and objective aspects of concentration problems in depressed patients. Br J Psychol 79 (Pt 2), 241–250.

Willeit, M., Praschak-Rieder, N., Neumeister, A., Pirker, W., Asenbaum, S., Vitouch, O., Tauscher, J., Hilger, E., Stastny, J., Brücke, T., Kasper, S., 2000. [123I]-beta-CIT SPECT imaging shows reduced brain serotonin transporter availability in drug-free depressed patients with seasonal affective disorder. Biol. Psychiatry 47, 482–489.

Winstanley, C.A., Dalley, J.W., Theobald, D.E.H., Robbins, T.W., 2003. Global 5-HT depletion attenuates the ability of amphetamine to decrease impulsive choice on a delay-discounting task in rats. Psychopharmacology (Berl.) 170, 320–331.

Winstanley, C.A., Dalley, J.W., Theobald, D.E.H., Robbins, T.W., 2004. Fractionating impulsivity: contrasting effects of central 5-HT depletion on different measures of impulsive behavior. Neuropsychopharmacology 29, 1331–1343.

Winstanley, C.A., Theobald, D.E.H., Dalley, J.W., Cardinal, R.N., Robbins, T.W., 2006. Double dissociation between serotonergic and dopaminergic modulation of medial prefrontal and orbitofrontal cortex during a test of impulsive choice. Cereb. Cortex 16, 106–114.

Wise, C.D., Berger, B.D., Stein, L., 1970. Serotonin: a possible mediator of behavioral suppression induced by anxiety. Dis Nerv Syst 31, Suppl:34–37.

Wise, C.D., Berger, B.D., Stein, L., 1973. Evidence of -noradrenergic reward receptors and serotonergic punishment receptors in the rat brain. Biol. Psychiatry 6, 3–21.

Wyrwich, K.W., Yu, H., 2011. Validation of POMS questionnaire in postmenopausal women. Qual Life Res 20, 1111–1121.

Yoshioka, M., Matsumoto, M., Togashi, H., Saito, H., 1995. Effects of conditioned fear stress on 5-HT release in the rat prefrontal cortex. Pharmacol. Biochem. Behav. 51, 515–519.

Zanardi, R., Artigas, F., Moresco, R., Colombo, C., Messa, C., Gobbo, C., Smeraldi, E., Fazio, F., 2001. Increased 5-hydroxytryptamine-2 receptor binding in the frontal cortex of depressed patients responding to paroxetine treatment: a positron emission tomography scan study. J Clin Psychopharmacol 21, 53–58.