From the Department of Physiology and Pharmacology
Karolinska Institutet, Stockholm, Sweden

IMBALANCED KYNURENINE PATHWAY IN SCHIZOPHRENIA AND DEPRESSION
- IMMUNOLOGICAL AND GENETIC ASPECTS

Xicong Liu

Karolinska Institutet

Stockholm 2016

All previously published papers were reproduced with permission from the publisher.
Cover: the word cloud of the thesis text in the shape of brain.
Published by Karolinska Institutet.
Printed by E-Print AB 2016
© Xicong Liu, 2016
ISBN 978-91-7676-416-9

Imbalanced Kynurenine Pathway in Schizophrenia and Depression - Immunological and Genetic Aspects
THESIS FOR DOCTORAL DEGREE (Ph.D.)

By

Xicong Liu

Principal Supervisor:
Professor Sophie Erhardt
Karolinska Institutet
Department of Physiology and Pharmacology
Division of Neuropsychoimmunology

Co-supervisor(s):
Dr. Lilly Schwieler
Karolinska Institutet
Department of Physiology and Pharmacology
Division of Neuropsychoimmunology

Associate Professor Simon Cervenka
Karolinska Institutet
Department of Clinical Neuroscience

Opponent:
Professor Lars Oreland
Uppsala University
Department of Neuroscience
Division of Neuropsychopharmacology

Examination Board:
Professor Gilberto Fisone
Karolinska Institutet
Department of Neuroscience

Docent Elisabet Stener-Victorin
Karolinska Institutet
Department of Physiology and Pharmacology
Division of Reproductive endocrinology and metabolism

Docent Mia Ericsson
Göteborgs Universitet
Institute of Neuroscience and Physiology
Section of Psychiatry and Neurochemistry

To my family

ABSTRACT

Kynurenic acid (KYNA), an end metabolite of the kynurenine pathway along tryptophan degradation, has gained increasing interest in the pathophysiology of psychiatric disorders. Elevated levels of KYNA have been found in the cerebrospinal fluid (CSF) and in the postmortem brain of patients with schizophrenia and bipolar disorder with psychotic symptoms. In contrast, reduced levels of KYNA are associated with depressive symptoms. Rapidly emerging evidence points to the involvement of brain immune activation in psychiatric disorders and to the kynurenine pathway as a causal link between brain immune activation and psychiatric disorders. The aim of present thesis was to investigate the role of the kynurenine pathway in the pathophysiology of schizophrenia and depression in experimental settings.

The results show that an infection with neurotropic influenza A virus or a transient elevation of brain KYNA levels in neonatal mice enhanced the sensitivity of d-amphetamine-induced increase in locomotor activity in adulthood. Neonatal elevation of KYNA also impaired prepulse inhibition and working memory in adulthood. These long-lasting behavioral alterations suggest that the kynurenine pathway as a causal link between early-life infection and the development of neuropsychiatric disturbances in adulthood.

Reduced KYNA levels were detected in the prefrontal cortex, but not in the frontal cortex, hippocampus, striatum or cerebellum, of Flinders Sensitive Line (FSL) rats, an animal model of depression, compared with their controls, the Flinders Resistant Line (FRL) rats.

Inhibition of Kynurenine 3-monooxygenase (KMO) shunts the kynurenine pathway towards enhanced synthesis of KYNA. Mice with a targeted deletion of KMO exhibited impairments in contextual memory and social interaction, potentiated horizontal activity following d-amphetamine-induced increase in locomotor activity as well as increased anxiety-like behaviors. In addition, genome-wide differential gene expression analyses identified alterations regarding schizophrenia- and psychosis-related genes in these mice.

Kynurenine aminotransferase (KAT) II is identified as the main enzyme responsible for most of brain KYNA production. Nevertheless, KAT II KO mice receiving kynurenine injection or repeated injections of Lipopolysaccharide (LPS) exhibited increased concentrations of brain KYNA.

Taken together, these results give strong experimental support for the connection between immune activation and KYNA in schizophrenia and suggest that low brain KYNA could be of importance for the depressive-like behaviors observed in FSL rats

LIST OF SCIENTIFIC PAPERS

I. **Xicong Liu***, Maria Holtze*, Susan B. Powell, Niccolò Terrando, Markus K. Larsson, Anna Persson, Sara K. Olsson, Funda Orhan, Magdalena Kegel, Linnea Asp, Michel Goiny, Lilly Schwieler, Göran Engberg, Håkan Karlsson, Sophie Erhardt. Behavioral disturbances in adult mice following neonatal virus infection or kynurenine treatment – Role of brain kynurenic acid. *Brain Behavior and Immunity* 2014; 36: 80–89

II. **Xicong Liu**, Sophie Erhardt, Michel Goiny, Göran Engberg, Aleksander A. Mathé. Decreased levels of kynurenic acid in prefrontal cortex in a genetic animal model of depression. *Acta Neuropsychiatrica* 2016; Jul 13:1-5. Epub ahead of print. DOI: 10.1017/neu.2016.31

III. Sophie Erhardt*, Ana Pocivavsek*, Mariaelena Repici, **Xicong Liu**, Sophie Imbeault, Daniel C Maddison, Marian AR Thomas, Joshua L Smalley, Markus K Larsson, Paul J Muchowski, Flaviano Giorgini, Robert Schwarcz. Adaptive changes and behavioral abnormalities in mice deficient in kynurenine 3-monooxygenase: Relevance to psychotic disorders. *Submitted manuscript.*

IV. **Xicong Liu***, Anthi Faka*, Lilly Schwieler, Sophie Erhardt. Repeated administration of LPS induces changes in the kynurenine pathway in KAT II deficient mice. *Manuscript.*

*Contributed equally

CONTENTS

LIST OF ABBREVIATIONS

3-HK	3-hydroxykynurenine
AADAT	Aminoadipate aminotransferase
AMPA	α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid
ANOVA	Analysis of variance
ASAT	Mitochondrial aspartate aminotransferase
BBB	Blood-brain barrier
CCBL	Cysteine conjugate beta-lyas
CNS	Central nerves system
COX	Cyclo-oxygenase
CSF	Cerebrospinal fluid
DEGs	Differentially expressed genes
DFP	Diisopropyl fluorophosphates
DISC	Disrupted-in-schizophrenia
FRL	Flinders Resistant Line
FSL	Flinders Sensitive Line
GOT	Glutamic-oxaloacetic transaminase
GTK	Glutamine transaminase K
GWAS	Genome-wide association studies
HPA	Hypothalamic-pituitary-adrenal
HPLC	High performance liquid chromatography
HSV-2	Herpes simplex virus type 2
i.e.	That is (*id est* lat.)
i.p.	Intraperitonatally
IC_{50}	Half maximal inhibitory concentration
IDO	Indoleamine 2,3-dioxygenase
IFN	Interferon
IL	Interleukin
KAT	Kynurenine aminotransferases
Km	Michaelis-Menten constant
KMO	Kynurenine 3-monooxygenase

KO	Knockout
KYNA	Kynurenic acid
KYNU	Kynureninase
LPS	Lipopolysaccharide
MDD	Major depressive disorder
NAD^+	Nicotinamide adenine dinucleotide
NMDA	N-methyl-D-aspartate
PAP	Passive avoidance paradigm
PBS	Phosphate buffered saline
PCP	Phencyclidine
PET	Positron Emission Tomography
PIC	Picolinic acid
PND	Postnatal day
PPI	Prepulse inhibition
QUIN	Quinolinic acid
RIN	RNA Integrity Number
RM	Repeated measure
TDO	Tryptophan 2,3-dioxygenase
TLR	Toll-like receptors
TNF	Tumor necrosis factor
α7nACh	α7 nicotinic acetylcholine

1 INTRODUCTION

1.1 Schizophrenia

The history of schizophrenia can be retrospected as early as 1550BC as the oldest documentation about an illness like schizophrenia is found in Ebers Papyrus from ancient Egypt (Burton, 2012). Symptoms similar to those manifested in schizophrenia are also described in the Bible. In the late 19th century, the German psychiatrist Emil Kraepelin integrated the pieces of various clinical observations of mental disorders into a single category termed "dementia praecox" (Kraepelin, 1896). Later, the Swiss psychiatrist Eugen Bleuler developed Kraepelin's theory and replaced the term with "schizophrenia" in 1911 (Bleuler, 1911).

1.1.1 Symptoms of schizophrenia

Patients with schizophrenia often have distorted perception of reality caused by a variety of symptoms. The symptoms are generally classified into three distinct categories (Andreasen and Olsen, 1982):

- *Positive symptoms*, also known as psychotic symptoms, refer to features not seen in healthy population but present in patients, such as hallucinations, delusions as well as disorganized thought processes.

- *Negative symptoms* are characterized by withdrawal or lack of function that is normally present in healthy people, such as anhedonia, social withdrawal, blunted affect and loss of motivation.

- *Cognitive dysfunctions* refer to a wide range of features regarding difficulties in problem solving, impairments in working memory and attention.

1.1.2 Prevalence and onset of schizophrenia

Schizophrenia has a global lifetime prevalence of approximately 0.40-0.87% (McGrath et al., 2008; Perälä et al., 2007; Saha et al., 2005). Schizophrenia is often accompanied with high suicide rates, approximately 5-10% higher than general population (Black and Fisher, 1992; Meltzer, 2002; Palmer et al., 2005). The average life expectancy in woman and man with

schizophrenia is 12 and 15 years shorter, respectively, than the rest of the population (Crump et al., 2013).

The onset of schizophrenia generally emerges in late adolescence or early adulthood. Since adolescence is a critical period for the development of the brain, the pathogenesis of schizophrenia may be embedded during early development (Paus, 2005; Paus et al., 2008; Welham et al., 2009). The neurodevelopmental theory of schizophrenia was suggested in the 1980s (Lewis, 1989; Lyon et al., 1989; Murray and Lewis, 1988; Weinberger, 1987) and proposed that early brain insults, caused by environmental or genetic risk factors, affect brain development and lead to pathological processes of schizophrenia (Rapoport et al., 2005).

1.1.3 Risk factors of schizophrenia

1.1.3.1 Environmental risk factors

Environmental risk factors related to prenatal, neonatal and early life have been highlighted by several studies pointing to the etiology of schizophrenia. In particular, increased risk of schizophrenia was found in subjects born in the winter or in the beginning of spring (Davies et al., 2003; Torrey et al., 1997) as well as in individuals born in urban area (Marcelis et al., 1999; Mortensen et al., 1999). Moreover, prenatal stress or early life trauma can also be risk factors for the development of the disease (Huttunen and Niskanen, 1978; Khashan et al., 2008; Morgan and Fisher, 2007; van Os and Selten, 1998). Furthermore, prenatal malnutrition regarding folate, iron and vitamin D are correlated with the development of the disease (Brown et al., 1996; Insel et al., 2008; McGrath, 1999; Rosso, 1990). In addition, studies have provided evidences for the association between maternal and postnatal infections, induced for example by influenza, *Toxoplasma gondii* and herpes simplex virus type 2 (HSV-2), and schizophrenia (Brown and Derkits, 2010; Dalman et al., 2008; Meyer, 2013). Therefore, it seems that this is not a pathogen-specific association; instead it sheds light upon the mediation of upregulated expression of inflammatory cytokines (Brown et al., 2004; Buka et al., 2001), suggesting an involvement of immunological factors in schizophrenia.

1.1.3.2 Genetic risk factors

For decades, family history of schizophrenia has been identified as the most predominant risk factor for the disease. When both parents are affected by schizophrenia, there is a relative risk of 89% for their child to develop the disease (Lichtenstein et al., 2006). Furthermore, twins studies have provided evidence showing that genetic factors account for more than 80% of the risk for the development of schizophrenia (Cannon et al., 1998; Cardno et al., 1999;

Sullivan et al., 2003). However, the complexity of the disease makes the search for a causal gene a hard and tortuous journey. In all probability, due to cooperative effects of multiple vulnerable genes that interact with environmental risk factors (Harrison and Owen, 2003).

1.1.4 Hypotheses of schizophrenia

1.1.4.1 The dopamine hypothesis of schizophrenia

The dopamine hypothesis has been one of the main theories of the pathophysiology of schizophrenia, proposing that dopaminergic hyperactivity is responsible for the positive symptoms observed in the disorder (Carlsson and Lindqvist, 1963). The hypothesis originates from findings that antipsychotic drugs have the potency to ameliorate the symptoms of schizophrenia by blocking dopamine D_2 receptors (Creese et al., 1976; Seeman and Lee, 1975; Seeman et al., 1976). In addition, dopamine enhancing drugs, such as amphetamine, can deteriorate the symptoms in patients with schizophrenia (Angrist et al., 1974) and induce a state of psychosis in healthy people (Snyder, 1973). Nevertheless, negative symptoms and cognitive impairments of schizophrenia have drawn more attentions for many years, and their treatment resistance to antipsychotic drugs led to a revision of the dopamine hypothesis (Breier, 1999; King, 1998). The revised dopamine hypothesis proposes an imbalance of dopaminergic transmission in schizophrenia involving hyperactivation of the subcortical mesolimbic dopamine pathway, which leads to positive symptoms, and a hypoactivation of the mesocortical dopamine projection to the prefrontal cortex resulting in negative symptoms and cognitive dysfunctions (Abi-Dargham and Laruelle, 2005; Davis et al., 1991; Goldman-Rakic et al., 2000).

1.1.4.2 The glutamate hypothesis of schizophrenia

The glutamate deficiency theory of schizophrenia was initially generated from a report suggesting low glutamate levels in the cerebrospinal fluid (CSF) of patients with schizophrenia (Kim et al., 1980). Around the same time, phencyclidine (PCP), known as an anesthetic, was found to induce schizophrenia-like symptoms in healthy volunteers and to exacerbate existing symptoms in patients with schizophrenia (Allen and Young, 1978; Itil et al., 1967; Luby et al., 1959). Later, PCP was characterized as a non-competitive N-methyl-D-aspartate (NMDA) receptor antagonist and found to reduce glutamate neurotransmission (Anis et al., 1983; Lodge and Anis, 1982). Moreover, ketamine, another non-competitive NMDA receptor antagonist, is shown to aggravate symptoms in patients with schizophrenia (Lahti et al., 2001; Malhotra et al., 1997) and mimic all spectra of symptoms in healthy individuals (Krystal et al., 1994). Furthermore, several studies have implied that competitive

NMDA receptor antagonists as well as compounds that block the glycine site of the NMDA receptor could evoke the symptoms of schizophrenia in human (Albers et al., 1999; Grotta et al., 1995; Kristensen et al., 1992; Yenari et al., 1998), which further give support to the glutamate deficiency theory.

The dopamine hypothesis and the glutamate deficiency theory are not totally irrelevant hypotheses. Several studies have confirmed that blockade of NMDA receptor leads to an increased dopamine activity in the midbrain of rodents (Erhardt and Engberg, 2002; French, 1994; French et al., 1993; Linderholm et al., 2007; Schwieler et al., 2004). Besides, administration of ketamine in healthy volunteers induces an increase of dopamine release and potentiates the amphetamine-induced dopamine response in striatum (Breier et al., 1998; Kegeles et al., 2000; Vollenweider et al., 2000). Altogether, these two theories support each other by implicating that NMDA receptor hypofunction induces an imbalance of dopamine neurotransmission in schizophrenia (Svensson, 2000).

1.1.4.3 The Kynurenic acid hypothesis of schizophrenia

In recent years, the kynurenic acid (KYNA) hypothesis of schizophrenia has drawn more and more attention. The hypothesis is constructed on the fact that KYNA is an endogenous antagonist of the NMDA receptor as well as of the α7 nicotinic acetylcholine (α7nACh) receptor in the brain (Hilmas et al., 2001; Stone, 1993). Several studies have shown that patients with schizophrenia display elevated levels of KYNA in the CSF (Erhardt et al., 2001a; Linderholm et al., 2012; Nilsson et al., 2005) as well as in the prefrontal cortex of postmortem brain (Sathyasaikumar et al., 2011; Schwarcz et al., 2001). In line with these studies, it has also been shown that kynurenine, the precursor of KYNA, is increased in the CSF and postmortem brain of patients (Linderholm et al., 2012; Miller et al., 2006).

Fluctuations in endogenous KYNA levels mediate alterations of dopamine neurotransmission. Thus, it has been shown that pharmacological elevation of KYNA increases firing of rat midbrain dopamine neurons (Erhardt and Engberg, 2002; Erhardt et al., 2001b; Linderholm et al., 2007; Nilsson et al., 2006), and reduced concentration of KYNA decreases the activity (Linderholm et al., 2016; Schwieler et al., 2006; Schwieler et al., 2008). Furthermore, subchronical elevation of KYNA distinctly enhanced the dopaminergic response to an amphetamine challenge (Olsson et al., 2009). This finding is in consonance with clinical observations of patients with schizophrenia showing hyper-dopaminergic response to amphetamine compared to healthy volunteers (Abi-Dargham et al., 1998; Laruelle, 1998; Laruelle and Abi-Dargham, 1999). Indeed, increased midbrain dopamine firing contributes to

the excess of subcortical dopamine, which is involved in the generation of positive symptoms in schizophrenia (Davis et al., 1991; Jentsch and Roth, 1999).

The KYNA hypothesis is further supported by a large amount of studies showing the association of NMDA and α7nACh receptors with cognitive functions (Timofeeva and Levin, 2011). Cognitive deficits, such as impairments in working memory, speed of processing, attention, learning and memory, problem solving are considered as core features in schizophrenia (Nuechterlein et al., 2004). The hypothesis has been tested in several behavioral experimental settings to understand how fluctuations in KYNA levels affect cognitive function. It has been shown that elevated levels of KYNA induce impairments in contextual processing (Alexander et al., 2012; Chess and Bucci, 2006), spatial working memory (Chess et al., 2007; Pocivavsek et al., 2011) as well as contextual memory (Chess et al., 2009), while lower levels of KYNA improve cognitive performance (Kozak et al., 2014; Potter et al., 2010). These results are in line with a recent study showing that elevated levels of KYNA are associated with cognitive impairments in patients with bipolar disorder (Sellgren et al., 2015). In addition, increased levels of KYNA disrupt prepulse inhibition (PPI) in the rat (Erhardt et al., 2004). PPI is associated with sensorimotor gating and reflects the ability of the brain to filter out abundant information. Indeed, deficits in PPI and sensorimotor gating are a core deficit of patients with schizophrenia (Braff and Geyer, 1990).

Taken together, the kynurenic acid hypothesis of schizophrenia covers the whole spectra of symptoms of schizophrenia and links the dopamine and glutamate hypotheses to explain the possible underling pathophysiology mechanisms of the disease.

1.2 Depression

Depression is a complex, multifactorial, heterogeneous and recurrent mental disorder with estimated lifetime prevalence from 11.1% to 14.6% among different counties (Bromet et al., 2011). It was ranked as the fourth leading cause of disability worldwide (Murray and Lopez, 1996) by World Health Organization and is projected as the second by 2020 (Murray and Lopez, 1997). Patients suffering from major depressive disorder (MDD) often experience multiple recurrences of the disease. Approximate two-thirds of the patients developed at least one recurrence (Solomon et al., 2000). Patients with depression often present persistent low mood and anhedonia accompanied by physical and mental alterations causing impairments in social, occupational and other capacity of functioning.

1.3 The kynurenine pathway

KYNA, when discovered in dog urine by the German chemist Justus von Liebig in 1853 (Liebig, 1853), was the first identified member of the kynurenine pathway. Half a century later, KYNA was recognized as a metabolite of tryptophan (Alexander, 1904), an essential amino acid required for the biosynthesis of proteins. Subsequently, with the identification of other metabolites of tryptophan degradation towards kynurenine, this pathway was termed the "kynurenine pathway" (Beadle et al., 1947). The majority of tryptophan is metabolized through the kynurenine pathway. Only less than 5% of tryptophan goes into the serotonin pathway (Gal and Sherman, 1980). Therefore, the kynurenine pathway is the main route of tryptophan degradations, accounting for approximate 90-95% of tryptophan in most mammalian tissues (Gal and Sherman, 1980; Leklem, 1971).

1.3.1 Kynurenic acid

KYNA is an endogenous antagonist acting on a wide range of glutamate receptors. At low concentrations, it blocks the strychnine-insensitive glycine site of the NMDA receptor [$IC_{50} \approx$ 8-15 μM] (Birch et al., 1988; Kessler et al., 1989; Parsons et al., 1997) and the α7nACh receptor [$IC_{50} \approx 7$ μM] (Hilmas et al., 2001; Stone, 2007). At higher concentrations, it also blocks the glutamate recognition site of the NMDA receptor [$IC_{50} \approx$ 200-500μM] (Kessler et al., 1989). At millimolar concentrations, KYNA competitively antagonizes on the α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA) receptor as well as the kainate receptor (Kessler et al., 1989). Additional targeting receptors of KYNA have been identified during the last couple of years. Thus, studies have suggested that KYNA acts as an agonist of the former orphan G protein-coupled receptor GPR35 (Berlinguer-Palmini et al., 2013; Resta et al., 2016) and as an efficient agonist of the aryl hydrocarbon receptor. Stimulation of the aryl hydrocarbon receptor with KYNA in the presence of interleukin (IL)-1β synergistically induces IL-6 expression. (DiNatale et al., 2010).

1.3.2 Enzymatic steps

The initial and rate-limiting step of the kynurenine pathway (Figure 1) is the oxidative opening of the indole ring of tryptophan to produce N-formyl kynurenine. Two enzymes are responsible for this process; indoleamine 2,3-dioxygenase (IDO1) and/or tryptophan 2,3-dioxygenase (TDO2). Subsequently, N-formyl kynurenine is rapidly hydrolyzed to kynurenine, the pivotal metabolite, by kynurenine formamidase. Kynurenine is able to pass

the blood-brain barrier (BBB) and is further catabolized to three different molecules: 1) Kynurenine aminotransferases (KAT I, KAT II, KAT III and KAT IV), which catabolize kynurenine towards KYNA 2) kynurenine 3-monooxygenase (KMO), which converts the degradation of kynurenine to 3-hydroxykynurenine (3-HK) and 3) kynureninase (KYNU), which forms anthranilic acid. KYNA is an end product, whereas 3-HK and anthranilic acid are subsequently metabolized by KYNU or a nonspecific oxidation to form 3-hydroxyanthranilic acid and then further converted to quinolinic acid (QUIN), which, as opposed to KYNA, is an agonist of the NMDA receptor (Stone and Perkins, 1981). The final metabolism along the QUIN branch leads to the production of NAD^+, which is an oxidized form of nicotinamide adenine dinucleotide (Moroni, 1999; Stone, 1993).

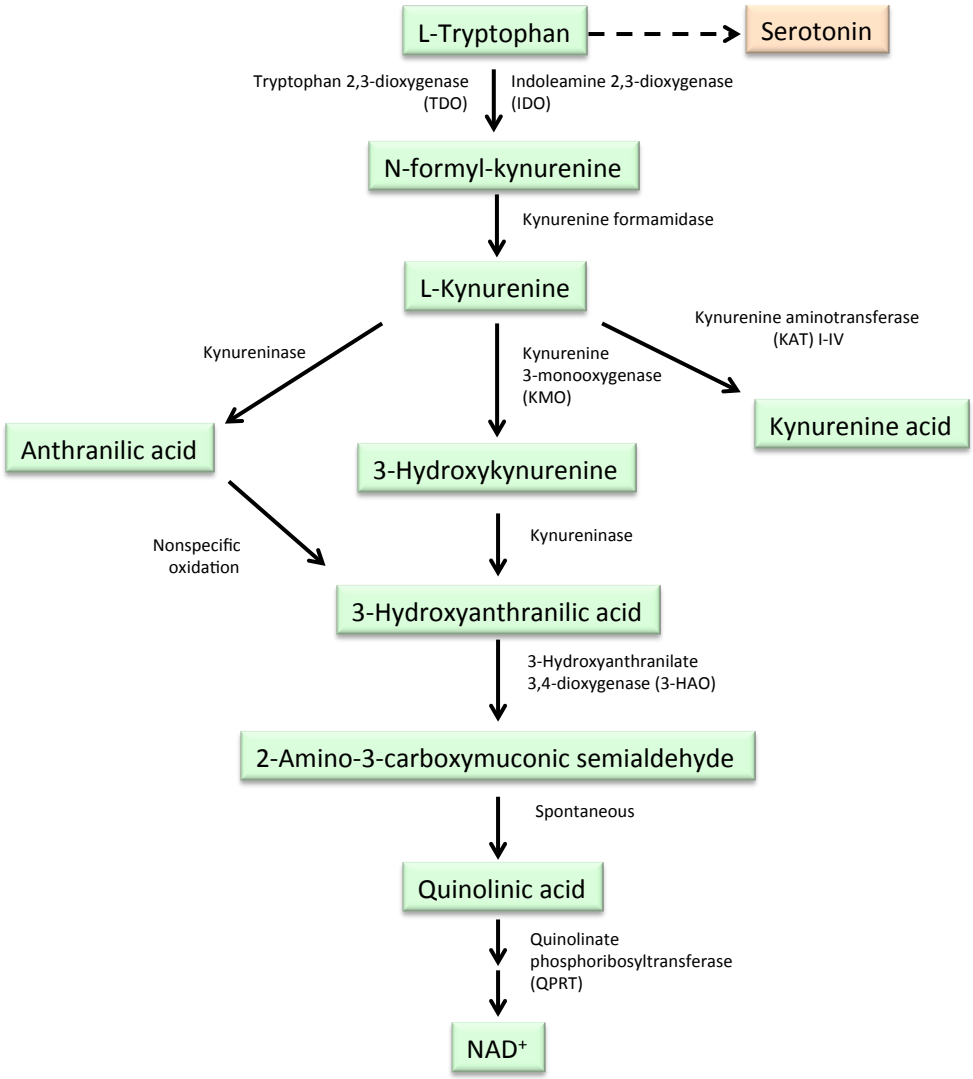

Figure 1. The kynurenine pathway

Enzymes involved in the kynurenine pathway have been found both in the central nerves system (CNS) and in the periphery. After the transportation of kynurenine from circulation to the CNS, it is taken up by glial cells (Speciale and Schwarcz, 1990). Notably, KAT enzymes, which convert kynurenine to KYNA, are primarily expressed in astrocytes (Guillemin et al., 2001b), however, KMO and KYNU enzymes, which participate in the 3-HK synthesis and further downstream production of QUIN, are mainly expressed in microglia (Guillemin et al., 2003). Thus, the two different branches of the kynurenine pathway are spatially segregated in the brain.

KAT enzymes have been identified with four homologues (Han et al., 2010a): KAT I/glutamine transaminase K (GTK)/cysteine conjugate beta-lyas (CCBL)1, KAT II/aminoadipate aminotransferase (AADAT), KAT III/CCBL2, KAT IV/glutamic-oxaloacetic transaminase (GOT)2/mitochondrial aspartate aminotransferase (ASAT). All these four enzymes are considered to be involved in the formation of KYNA in the brain (Guidetti et al., 2007; Guidetti et al., 1997; Okuno et al., 1991; Yu et al., 2006), however, due to different characteristics of the four isoforms such as substrate specificity, optimum pH and temperature, KAT II enzyme stands out to be the main biosynthesis enzyme for KYNA under physiological conditions (Han et al., 2010b; Schmidt et al., 1993).

1.3.3 Regulation of the kynurenine pathway

The KAT II enzyme has a Michaelis Menten constant (*Km*) in millimolar range, suggesting that the enzyme is far from saturated under normal physiological conditions (Okuno et al., 1991). Thus, the production of KYNA is largely dependent on the availability of its precursor, kynurenine, and indeed, systematic administration of kynurenine increases the levels of KYNA in brains in both rats and primates (Jauch et al., 1993; Swartz et al., 1990; Wu et al., 1992).

The activity of the kynurenine pathway is regulated by several distinct mechanisms. Intracellular concentration of amino acid such as glutamine and phenylalanine, which are the competitive substrates of KAT I and KAT II, have been demonstrated to control the formation of KYNA in the brain (Chang et al., 1997). KMO is the enzyme converting the degradation of kynurenine towards QUIN branch in the kynurenine pathway and pharmacological blockade of KMO leads to shunting of the metabolism of kynurenine towards KYNA (Rover et al., 1997; Russi et al., 1992; Speciale et al., 1996). Moreover, cyclo-oxygenase (COX)-1 inhibition with indometacin or diclofenac indirectly increases KYNA levels in the brain (Edwards et al., 2000; Schwieler et al., 2005; Schwieler et al.,

2006), whereas, systematic administration of parecoxib or meloxicam, which act as COX-2 inhibitors, decreases KYNA concentrations in the brain (Schwieler et al., 2005; Schwieler et al., 2006). Furthermore, KYNA is actively extruded from the CNS by organic anion transporters (Nemeth et al., 2005; Uwai et al., 2012). To avoid its extrusion, probenecid is used to block organic anion transporter activity (Cunningham et al., 1981; Moroni et al., 1988). Notably, pro-inflammatory cytokines are also able to induce the rate-limiting enzymes, IDO1 (Carlin et al., 1987; Yoshida et al., 1986) and TDO2 (Miller et al., 2006; Sellgren et al., 2015), which lead to increased levels of kynurenine and further induction of the entire pathway. Thus, this evidence implies that immune activation is involved in the regulation of the kynurenine pathway.

1.3.4 Regulation of the kynurenine pathway following immune activation

Cytokines are released from innate immune cells under immune response. The principal cytokine to induce IDO1 is interferon (IFN)-γ (Pemberton et al., 1997), which has been shown to increase IDO1 transcription in many cell types such as macrophages, microglia, dendritic cells, epithelial cells, smooth muscle cells and several tumor cell lines (Alberati-Giani et al., 1996; Chiarugi et al., 2001; Cuffy et al., 2007; Jeong et al., 2009; Mailankot and Nagaraj, 2010; Takikawa, 2005). Other cytokines including IFN-α, IFN-β and tumor necrosis factor (TNF)-α have also the capacity to induce IDO1, but to a less degree than IFN-γ (Guillemin et al., 2001a; Pemberton et al., 1997). Recent studies have shown that the other rate-limiting enzyme TDO2 is induced by IL-1β (Sellgren et al., 2015; Urata et al., 2014). Moreover, IFN-γ has been shown to induce KMO expression as well as its activity (Alberati-Giani et al., 1996; Chiarugi et al., 2001). Following systemic inflammatory stimulation by Lipopolysaccharide (LPS), KMO expression is enhanced together with increased levels of TNF-α and IL-6 in rat brains (Connor et al., 2008).

1.3.5 The kynurenine pathway in schizophrenia and depression

As described in the “kynurenic acid hypothesis of schizophrenia” section, KYNA levels are elevated in both CSF and postmortem brains of patients with schizophrenia (Erhardt et al., 2001a; Schwarcz et al., 2001). Other metabolites and enzymes involved in the kynurenine pathway have also been measured. Importantly, a reduction of KMO gene expression and enzyme activity has been discovered in patients with schizophrenia and bipolar disorder (Lavebratt et al., 2014; Sathyasaikumar et al., 2011; Wonodi et al., 2011). Thus, the reduced activity in KMO shunts the metabolism of kynurenine towards KYNA (Sathyasaikumar et al.,

2011). Interestingly, studies investigating rate-limiting enzymes of the kynurenine pathway revealed that only TDO2 expression, but not IDO1, is upregulated in brains of patients with schizophrenia and bipolar disorder with psychosis (Miller et al., 2004; Miller et al., 2006).

Participation of the kynurenine pathway in depression was suggested in 1970's (Lapin, 1973), and implied a connection between tryptophan, kynurenine and depression. However, due to the achievement of antidepressants using serotonin reuptake inhibitors, the focus of depression research shifted to serotonin. The involvement of the kynurenine pathway in depression is now gaining increased attention and for instance IDO1 activation is suggested to play a critical role in the induction of depressive symptoms. Pro-inflammatory cytokines, such as IFN-α, stimulate IDO1 and further induce depressive symptoms in patients with hepatitis C and malignant melanoma (Capuron et al., 2003; Raison et al., 2010). Moreover, animal studies have demonstrated that bacterial infection or upregulation of IDO1 by IFN-γ and TNF-α leads to long-lasting depressive-like behaviors (O'Connor et al., 2009a; O'Connor et al., 2009b). The elevation of downstream neurotoxic metabolites of the kynurenine pathway, such as 3-HK and QUIN, are implicated for the responsibility for depressive symptoms (Muller and Schwarz, 2007). In addition, post mortem studies have found microglia activation and decreased number of astrocytes in patients with depression (Si et al., 2004; Steiner et al., 2011). Furthermore, in recent studies, low CSF levels of KYNA associated to severe depressive symptoms (Bay-Richter et al., 2015) and low plasma KYNA has also been found in patients with depressive disorders (Myint et al., 2007; Schwieler et al., 2016). A reduction in the KYNA/QUIN ratio has also been found in both depressed patients and during the remitted phase of MDD (Savitz et al., 2015b). Interestingly, the KYNA/QUIN ratio has been found to positively correlate with larger hippocampal and amygdala volumes in patients with MDD (Savitz et al., 2015a)

1.4 Virus infections and CNS immune responses

Virus infections usually start in the periphery since viruses, including neurotropic viruses, hardly have access to the CNS directly. Viruses may enter the brain only in rare events, such as in cases of disrupted BBB or by release from circulating immune cells, having penetrated the BBB. Following virus invasions, viral molecules are recognized and subsequently trigger a cell-autonomous immune response, which increases the production of various anti-viral proteins and cytokines (Koyuncu et al., 2013).

Previously, the CNS was believed to be an immune privileged site within the entire body (Bailey et al., 2006; Galea et al., 2007), meaning that the CNS is segregated from the peripheral immune system by the BBB. However, recent studies have dramatically modified this idea and revealed a concept of a bidirectional communication between the brain and peripheral immune systems (Trakhtenberg and Goldberg, 2011; Wilson et al., 2010). Interestingly, a new discovery suggests a lymphatic system directly linking the peripheral immune system to the brain (Louveau et al., 2015). Cytokines are able to cross the BBB and serve as communication molecules between the two systems. The innate immune response in the CNS is initiated by microglia, astrocytes and neurons (Carson, 2002; Reiss et al., 2002), which is the first step of the host defense to viruses or infectious agents in the brain. Microglia are widespread in the brain and serve as monitors to identify foreign pathogens via Toll-like receptors (TLR) during CNS immunological process (Olson and Miller, 2004). Astrocytes also express TLRs and release cytokines, chemokines and neruotrophic factors following pathogen recognition (Falsig et al., 2008; Farina et al., 2007). These mediators trigger adjacent cells and further amplify the innate immune response in the CNS. They also alter BBB permeability, hereby allowing immune cells in the blood circulation to easier cross the BBB and participate in the adaptive immune response.

1.5 Immunological aspects

1.5.1 Immunological aspects in schizophrenia

The concept of immune activation involvement in psychotic disorders emerged in early 1900s by Julius Wagner-Jauregg who won the Nobel Prize for his discovery of the importance of therapeutic fever in mental patients (Tsay, 2013). During the last couple of years, this idea has gained increased support and recognition from numerous observations. Several risk factors, such as late winter birth, maternal infections and early childhood infections are all associated with immunological aspects and the development of schizophrenia (Brown and Derkits, 2010; Dalman et al., 2008; Davies et al., 2003). Based on genome-wide association studies (GWAS), a recent meta-analysis study revealed that immune related genes are the most significantly associated with schizophrenia (Aberg et al., 2013). Moreover, a Positron Emission Tomography (PET) study has revealed activation of microglia in schizophrenia (van Berckel et al., 2008). Furthermore, elevated levels of CSF cytokines including IL-1β (Soderlund et al., 2009) and IL-6 (Sasayama et al., 2013; Schwieler et al., 2015) have been observed in patients with schizophrenia. In analogy, postmortem studies have investigated

mRNA and protein levels of cytokines in the frontal cortex in human. Patients with schizophrenia exhibited higher expression levels of IL-1β and TNF-α in Brodmann area 10 (Rao et al., 2013). Similar results have been found in another postmortem study, which discovered upregulated mRNA levels of IL-1β, IL-6 and IL-8 in dorsolateral prefrontal cortex in patients with schizophrenia (Fillman et al., 2013).

1.5.2 Immunological aspects in depression

Depression often occurs together with other diseases, such as atherosclerosis, rheumatoid arthritis and congestive heart failure, all involving chronic inflammatory factors (Evans et al., 2005). Cytokine immunotherapy is a useful treatment strategy for some specific cancer and viral diseases (Ardolino et al., 2015; Mattiello et al., 2015). Patients with cancer or hepatitis C receiving cytokine immunotherapy frequently generate depressive symptoms (Capuron and Dantzer, 2003; Capuron et al., 2002). Moreover, elevated levels of pro-inflammatory cytokines, such as TNF-α, IL-1β and IL-6 have been found in peripheral blood in patients with depression (Miller et al., 2009; Zorrilla et al., 2001). Furthermore, induction of the immune system by administration of cytokines or cytokine inducers leads to depressive symptoms (Dantzer et al., 2008; Reichenberg et al., 2001) and anti-inflammatory therapy may ameliorate the symptoms of depression (Tyring et al., 2006).

1.6 Animal models

Animal models of mental disorders serve as valuable tools in preclinical research, supporting the investigation of underling neurobiological mechanisms of the disorder.

The criterion of animal models validation is generally categorized as construct, predictive and face validity. Construct validity emphasizes if the animal model provides a rational theoretical construct. Predictive validity refers to the potency of the animal model to predict the efficacy of pharmacological treatments. Face validity corresponds to the accuracy of the animal model to mimic the core symptoms of the disorder in a human condition.

1.6.1 Animal models of schizophrenia

The existing animal models of schizophrenia are not able to predict the whole spectrum of symptoms of schizophrenia. The design of these animal models are based on specific mechanistic and causative hypotheses that related to schizophrenia. A variety of animal behavioral tests, correlating to human conditions, have been established for validating animal

performance. Animal models of schizophrenia can be categorized into four groups: developmental, genetic, pharmacological and lesion models.

PPI of the startle reflex measures the sensorimotor gating and serves as an index in schizophrenia. It measures the ability of a non-startling auditory stimulus to inhibit the startle response to an intense auditory stimulus (Hoffman and Ison, 1980), i.e. the ability of the brain to filter out redundant information (Braff et al., 1978; Braff and Geyer, 1990). It is well known that patients with schizophrenia display disruptions in PPI (Braff et al., 1978; Swerdlow et al., 2008). PPI is a cross-species operation of measuring sensorimotor gating, it has thus been widely used in translational animal models (Swerdlow et al., 2000; Swerdlow et al., 1994).

1.6.1.1 Developmental models

Developmental models of schizophrenia are based on the fact that exposure of the neonate to environmental risk factors during prenatal or postnatal period increases the risk of schizophrenia development. Risk factors, such as infections or immune activation, maternal stress, and isolation rearing perturbed the neonate's CNS development (Asp et al., 2009; Bitanihirwe et al., 2010; Harms et al., 2008; Holloway et al., 2013; Koike et al., 2009; Macedo et al., 2012).

1.6.1.2 Genetic models

Genetic animal models are instructive for the understanding of the pathogenesis of the disorder. Creating animal models with targeted deletions of the genes of interest is a common strategy to explore the function of a single gene in the pathophysiology of a disorder. One of the historical candidate genes of schizophrenia is *disrupted-in-schizophrenia 1* (DISC1). DISC1 knockout (KO) mice display impairments in a broad spectrum of behaviors associated to psychiatric disorders including PPI (Lipina and Roder, 2014). As previously described, KYNA plays an essential role in schizophrenia pathophysiology. Thus, the main biosynthesis enzyme of KYNA (KAT II) has been investigated and KAT II KO mice were developed to study the phenotypic function of low KYNA levels. Indeed, KAT II KO mice display improved cognitive functions (Potter et al., 2010). Another genetic model associated with the kynurenine pathway is the KMO KO mouse model, which presents high levels of brain KYNA (Giorgini et al., 2013). In the present thesis, *Kmo*$^{-/-}$ mice have served as a platform to study subsequent influences of increased brain KYNA levels.

1.6.1.3 Pharmacological models

Pharmacological models are mainly designed from our current knowledge regarding neurotransmitter changes in schizophrenia. The most well known pharmacological model of schizophrenia is the amphetamine model, which reflect the mesolimbic dopaminergic hyperactivity (Breier et al., 1997; Laruelle, 1998). Administration of amphetamine induces hyperlocomotion and stereotypy in animals (Kokkinidis and Anisman, 1980; Sharp et al., 1987), which have been correlated with positive symptoms of schizophrenia. Moreover, NMDA receptor antagonist such as PCP and ketamine induce not only positive systems, but also partial negative and cognitive deficits of schizophrenia (Marcotte et al., 2001; Monte et al., 2013).

1.6.1.4 Lesion models

Lesion models are generated from the theory that neurodevelopment and neurodegeneration are linked to schizophrenia. Specific brain regions including the prefrontal cortex, the hippocampal formation and the thalamus have been selected when constructing lesion models of schizophrenia (Bardgett et al., 1997; Jaskiw et al., 1990; Kodsi and Swerdlow, 1997; Mittleman et al., 1993; Swerdlow et al., 1995). Lesion models typically involve excitotoxic substances, which lead to the release of excitatory glutamate and cause damage to neuronal tissue. The most acknowledged lesion model is the neonatal lesion model, which is also categorized into developmental models. It is widely used to study the neurodevelopmental theory of schizophrenia (Brake et al., 2000).

1.6.2 Animal models of depression

1.6.2.1 Genetic models

The monoamine hypothesis of depression states a depletion of monoamine neurotransmitters, such as serotonin, norepinephrine, and/or dopamine in the CNS leads to depression (Delgado, 2000). Based on this theory, traditional genetic models of depression such as serotonin receptor 1A knockout mouse model (Heisler et al., 1998; Ramboz et al., 1998) or the noradrenaline transporter knockout mouse model (Xu et al., 2000) were generated. Other transgenic animal models, targeting on corticotropin-releasing hormone receptor-1 (Muller et al., 2003) or the type II glucocorticoid receptor (Montkowski et al., 1995), are based on a theory suggesting a dysregulation of the hypothalamic-pituitary-adrenal (HPA) axis in depression.

Selective breeding is another method to develop animal models of depression. It consists of selection of animals according to their specific features and several generations of breeding. Because of these reasons, animals are genotypically identical to each other in inbred strains and show specific behavioral or physiological abnormalities. One example of genetic models by selective breeding is the Flinders Sensitive Line (FSL) and the Flinders Resistant Line (FRL) rats, which were originally generated towards increased sensitivity to an anticholinesterase agent (diisopropyl fluorophosphates, DFP). Compared to the FRL rats, the FSL rats exhibit partial depressive-like behavior as well as certain neurochemical and pharmacological features involved in depression (Overstreet et al., 2005). The FSL rats display reduced appetite, psychomotor retardation as well as deficits in sleep and immune system (Overstreet et al., 2005; Overstreet and Wegener, 2013).

1.6.2.2 Stress Models

Stress models of depression are based on different types of stress including social stress, chronic mild stress, early life stress and learned helplessness. These different types of stress models offer distinctive information for various subsets of the disorder (Czeh et al., 2016).

1.6.2.3 Immune models

The hypothesis of immunological abnormalities in depression has gained increased attentions. Activation of the peripheral immune system in humans and rodents induces sickness behavior including fever, fatigue, anhedonia, nausea and malaise (McCusker and Kelley, 2013), which have phenomenological similarities with depressive symptoms. LPS, a main component of the out membrane of gram-negative bacteria, has been found to induce sickness behavior in rodents (Bluthe et al., 1994). Administration of LPS induces the expression of pro-inflammatory cytokines in the brain, subsequently activates IDO1 and disrupts the balance of the kynurenine pathway (Parrott et al., 2016). Thus, the LPS model has been used as a reliable immune animal model to study the pathophysiological role of immune activation in depression (O'Connor et al., 2009c; Salazar et al., 2012).

2 AIMS OF THE THESIS

1) To investigate locomotor response to a d-amphetamine challenge in adult wild-type mice neonatally infected with influenza A virus.

2) To investigate whether neonatally elevated brain KYNA is associated with the disturbed behavior in adulthood seen after infection in early life.

3) To investigate if the concentrations of kynurenine pathway metabolites are changed in different brain regions in Flinders Sensitive Line rats, an animal model of depression.

4) To investigate possible changes in gene expression in the brain of mice with a targeted deletion of *Kmo* (*Kmo*$^{-/-}$ mice).

5) To assess cerebral and cerebellar variations in KYNA levels in *Kmo* deficient mice.

6) To characterize *Kmo* deficient mice behaviorally.

7) To investigate if KAT II KO mice, which have less capacity to generate KYNA, are prevented from elevation of brain KYNA induced by an LPS triggered immune response.

3 MATERIALS AND METHODS

3.1 Animals

In **paper I,** wild-type C57BL/6 mice (Scanbur AB, Sweden) were used in all experiments. The virus-infected mice and their counterparts were bred at the Department of Neuroscience, Karolinska Institutet, Sweden. The kynurenine-treated mice and their respective controls were bred at the Department of Physiology and Pharmacology, Karolinska Institutet, Sweden. Female FSL and FRL rats were used in **paper II** and bred at the Department of Clinical Neuroscience, Karolinska Institutet, Sweden. In **paper III,** male *Kmo*$^{-/-}$ mice and their control wild-type mice were bred on C57BL/6 background at the Department of Genetics, University of Leicester, UK or FVB/N background at the Department of Physiology and Pharmacology, Karolinska Institutet, Sweden as well as the Department of Psychiatry, University of Maryland School of Medicine, Baltimore, USA. The whole genome gene-expression analysis was carried out on *Kmo*$^{-/-}$ and wide-type mice on a C57BL/6 background. Metabolite analyses, enzyme activity and behavior tests were performed on *Kmo*$^{-/-}$ and wild-type mice on a FVB/N background. In **paper IV,** male KAT II knockout and wild-type mice on a FVB/N background were used in all experiments and bred at the Department of Physiology and Pharmacology, Karolinska Institutet, Sweden.

All animals were maintained under standard laboratory conditions with free access to food and tap water in a light controlled room (12 h light/dark cycle, light on at 6.00 a.m.). Experiments were approved by and performed in accordance with the guidelines of the Ethical Committee of Northern Stockholm, Sweden. In paper III, all experiments were approved and issued by the Home Office (UK), the Institutional Animal Care and Use Committee of the University of Maryland School of Medicine (USA), or the Ethical Committee of Northern Stockholm (Sweden). All efforts were made to minimize the number of animals used and their suffering.

3.2 Drugs and chemicals

L-kynurenine sulfate salt (Sigma Aldrich) was dissolved in sterile dH_2O and adjusted pH with NaOH to approximate 8.2 (**paper I and IV**). d-amphetamine (Sigma Aldrich) was dissolved in sterile dH_2O (**paper I and III**). LPS (Escherichia coli serotype O111:B4) was purchased from Sigma Aldrich and dissolved in sterile saline (**paper IV**).

3.3 Treatment

3.3.1 Influenza A/WSN/33 virus infection (paper I)

The mouse adapted neurotropic influenza A/WSN/33 virus was obtained from Dr. S Nakajima (The Institute of Public Health, Tokyo, Japan). Mice were infected intraperitonatally (i.p.) at postnatal day (PND) 3 or PND 4 with 2400 plaque-forming units of the influenza A/WSN/33 virus suspended in 30 µl of phosphate buffered saline (PBS; Gibco) to mimic a hematogenous route of infection.

3.3.2 L-kynurenine treatment (paper I and IV)

In **paper I**, L-kynurenine was injected i.p. to C57BL/6 mice from PND 7 to PND 16, every 12^{th} hour to mimic a transient increase in brain KYNA levels following influenza virus infection (Holtze et al., 2008). In **paper IV**, acute L-kynurenine injection was performed i.p. in KAT II KO and wild-type mice.

3.3.3 LPS injection (paper IV)

Both KAT II KO and wild-type mice received two LPS injections (i.p.) at PND 21. The second injection was administrated 16 hours after the first injection and mice were sacrificed 24 hours after the first injection at PND 22. In another set of animals, LPS was injected twice in both wild-type and KAT II KO mice as adults.

3.4 Behavior tests (paper I and III)

Several behavior tests were utilized to investigate behavioral alterations in different animal models. Locomotor activity, elevated plus maze, light-dark box and PPI were investigated in both **paper I and III.** Trace fear conditioning was used in **paper I**. Passive avoidance and social interaction were investigated in **paper III**.

3.4.1 Locomotor activity

Locomotor activity was studied in an open field apparatus for mice. Each mouse was placed in a square Plexiglas box (50 x 50 x 21.6 cm) within a sound-dampened and solid chamber. To detect the movement, the chamber was equipped with two rows of photocells sensitive to infrared light forming a two-layer grid over the open field arena. All mice were habituated

during three sessions of 60 min, 24 h apart. General locomotor activity was examined during the first session of the first day. The last habituation session was immediately followed by a test session of 90 min. d-amphetamine or vehicle was injected i.p. approximately 1 min prior to the test session. All habituation sessions and test sessions were performed in the dark during the animal's light cycle.

3.4.2 Elevated plus maze

The protocol of elevated plus maze was based on the method of Lister (Lister, 1987). The maze was shaped like a "plus" sign and consisted of two open arms (30 × 5 cm) and two equal-sized closed (30 × 5 × 15 cm) arms opposite to each other. Open and closed arms extended from a central platform (5 × 5 cm). The maze was made of stainless steel and mounted on a base, raising it to a height of 50 cm above the floor. Mice were individually placed on the central platform facing an open arm, and allowed to freely explore the maze for 5 min. The number of entries into different arms and the time spent in each arm was recorded by a video camera and then analyzed using the top-view based behavior analysis software TopScan Lite (Clever Sys Inc., Reston, VA, USA). The number of entries and percent time spent in different arms was measured as an index of exploratory behavior. After each test, the maze was cleaned with 70% ethanol solution and dried for 10 min.

3.4.3 Light-dark box

The light-dark box was made of Plexiglass (50 x 25 x 25 cm) and was equally divided into an open white compartment and a darkened closed black compartment. The two compartments were separated by a partition with a 10 x 5 cm opening in the center. Each mouse was placed in the center of the light section facing away from the dark chamber and allowed to explore the box freely for 5 min while being recorded with an overhead digital video camera. The box was cleaned with 70% ethanol solution between subjects. Time spent in each compartment and total number of transitions was analyzed.

3.4.4 Trace fear conditioning

The trace fear conditioning test was carried out by means of a fear conditioning chamber (Med Associates Inc., St. Albans, VT, USA). During training section, mice were allowed to explore the chamber for 100 s before a 20 s tone cue (90 dB). After 18 s interval a foot shock (2 s duration, 0.5 mA intensity) was delivered through the stainless steel rods on the floor of the apparatus. A second tone-shock pairing was repeated after 100 s inter trial interval and the

following 5 s after the second shock was defined as working memory. Working memory is generally defined as cognitive entities (or ‘‘central executive’’ mechanisms) relating to temporary storage and operation of information in both humans and animals (Yoon et al., 2008). The mouse was removed 30 s after the last shock. Three days later, freezing was recorded in the same context, with no tone or shock exposures. Freezing was scored automatically by the Med Associate software and defined as the absence of movement except that required for respiration. Approximately 3h after the contextual assessment, mice were again placed in the apparatus and freezing was recorded in a novel environment (a plastic floor covered the metal grid and a pyramidal shape was inserted in the rectangular box) and in response to the cue (tone). After 100 s exploration, the auditory cue was presented for 20 s followed by a 120 s inter trial interval, then another 20s cue presented. The percentage of time spent freezing was used to score learning and memory. A decrease in percent freezing represents impairment of these abilities.

3.4.5 Passive avoidance

The passive avoidance paradigm (PAP) had two compartments of equal size (each 22 cm high, 18 cm wide and 16 cm deep), one illuminated and the other in darkness, separated by a guillotine door. During the acquisition trial, on day 1, the mouse was first placed in the illuminated compartment. The door was then opened, prompting the mouse to move rapidly into the preferred dark compartment. The latency to enter the dark compartment was recorded as the “approach latency” and the guillotine door was immediately closed. An inescapable foot shock (0.56 mA for 1 sec) was delivered through metal rods of the floor. Twenty-four hours later, in the retention trial, the mouse was again placed in the light compartment, and the guillotine door was opened. The time from opening the guillotine door to the time of entering the dark compartment, which defined as “avoidance latency”, was recorded.

3.4.6 Social interaction

The social interaction test was performed in a three-chambered box. All chambers were equal in size (each 17 cm wide and 29.5 cm deep). The compartments were separated by clear walls with openings to allow access into each chamber. The test mouse was placed into the center chamber and allowed to explore for 5 min, the two side chambers were obstructed by plastic boxes. After acclimating to the testing environment, mouse was given 10 min to explore all three chambers of the testing apparatus, this was termed the habituation phase and the amount of time spent in each chamber was recorded. After the habituation period, an unfamiliar

129Sv/Ev male (stranger) was placed in one of the side chambers. The location of the stranger mouse systematically alternated between testing sessions. The stranger mouse was enclosed in a wire cup (9.5 cm in diameter), which allowed nose touching but no fighting. In the alternate chamber, an empty wire cup (termed the novel object) was placed. The test mouse was allowed to explore the entire testing apparatus for a 10 min session. The amount of time spent in each chamber was recorded by two blinded human observers seated five feet away. Data between these two observers were averaged. Additionally, the testing was video recorded, and a third blinded human observer scored the direct interaction between the test mouse and stranger mouse.

3.4.7 Prepulse inhibition

PPI of the startle response is used as an operational measure of sensorimotor gating and is analyzed by measuring the ability of a non-startling ‘‘prepulse’’ to inhibit the response to a startling stimulus (Hoffman and Ison, 1980). Startle response and PPI testing were performed in commercial startle chambers (35 x 33 x 46 cm, SR-LAB™ system, San Diego Instruments, San Diego, CA). The mouse was placed into a Plexiglas cylinder (3.7 cm in diameter), which was mounted in each chamber. Sudden movements by the mouse were detected by a piezoelectric accelerometer attached below the cylinder. A standard personal computer recorded the signals and controlled the presentations of acoustic stimuli and broadband background noise.

The experimental session consisted of a 5 min habituation period to a 65 dB background noise (continuous throughout all blocks), followed by five 120 dB single trials. This was immediately followed by three different test blocks: 1) A variable stimulus intensity block including five trial types: a 40 msec 80, 90, 100, 110, or 120 dB startle pulse, each trial presented 4 times. 2) A variable prepulse intensity block including 12 single 120 dB trials, and 10 trials each of a 69, 72, or 81 dB, 20 msec long prepulse followed 80 msec later by a 120 dB pulse. 3) A variable interstimulus interval block consisting of a 72 dB prepulse followed by a 120 dB pulse with a 25, 50, 100, 200 or 500 msec delay from the start of the prepulse. Eight single 120 dB trials were interspersed within the block.

3.5 Microdialysis (paper I)

3.5.1 Microdialysis surgery

Mice were anesthetized in a Plexiglas chamber continuously ventilated with 4.8% isoflurane in air using a vaporizer (Univentor 400 Anesthesia Unit; Univentor Ltd, Zejtun, Malta) and then mounted onto the ear bars of a conventional stereotaxic frame (David Kopf Instruments, Tujunga, CA, USA). Anesthesia was maintained using a nose cone delivering 2.4% isoflurane. Body temperature was maintained at 37 °C throughout the surgery, by a thermometer and a heating pad (Homeothermic Blanket Control Unit 50–7053-F, Harvard Apparatus, Holliston, MA, USA). An ocular lubricant was applied and 0.5 ml sterile saline was given s.c. to prevent dehydration. The skull was exposed and cleaned from adhesions. Next, a thin layer of quick-setting cyanoacrylate glue (BT AB, Stockholm, Sweden) was applied to the exposed skull, serving as an adhesion surface to the dental cement. A small hole was then drilled over striatum (AP: 0.5 mm anterior to bregma, L: 2 mm from midline), and, following careful removal of the dura, a guide cannula (AT4.7.IC, AgnTho's AB) containing a dummy probe (outer diameter: 0.2 mm) was implanted (V: 1.5 mm below the brain surface), and secured to the skull with acrylic dental cement (Dentalon® plus, Heraeus, Hanau, Germany). Before each guide cannula implantation the incisor bar was adjusted so that the skull was set in a horizontal flat plane. The wound was then sutured and mice were allowed to recover single-housed for 48 h with food and water ad libitum.

3.5.2 In vivo microdialysis

On the day of experiment, microdialysis was performed in the home cage in unanaesthetized freely moving mice. Mice were tethered to a swivel, the guide was removed and a microdialysis probe (AT4.7.2.PES, shaft length: 7 mm, membrane length: 2 mm, molecular cut-off: 6 kDa, AgnTho's AB) was inserted through the guide cannula. Probes were perfused with perfusion fluid (Ringer solution containing 148 mM NaCl, 4 mM KCl, 0.8 mM $MgCl_2$, 1.4 mM $CaCl_2$), delivered via polyethylene tubing from a microinfusion pump (Univentor 864, Univentor Ltd) at a flow rate of 1 µl/min. All samples were collected in plastic tubes attached to the tether in 30 min (for the analysis of dopamine) intervals throughout the experiment and immediately manually injected (Rheodyne, Cotati, CA, USA) into a high performance liquid chromatography (HPLC) system. To minimize the inter-individual variation due to differences in probe recovery, the dialysate concentrations were transformed to percent of baseline before statistical analysis. A stable baseline, consisting of three

consecutive samples with a maximal variation of 10%, was usually obtained after 2–3 h and defined as 100%. These three samples were taken for determination of basal extracellular dopamine concentrations. Results for subsequent samples were calculated as percentages of this average basal release. The mice then received an i.p. injection of d-amphetamine (2 mg/kg), and dopamine concentrations were measured for up to 300 min. After the session, the mice were sedated with isoflurane and sacrificed with cervical dislocation and tissues were frozen for later analysis and histological verification of probe placement.

3.6 High performance liquid chromatography (paper I - IV)

3.6.1 Fluorescent detection (paper I – IV)

The HPLC system consist of a dual-piston, high-pressure liquid delivery pump Shimadzu LC-10AD (Shimadzu Corporation, Kyoto, Japan), a ReproSil-Pur C18 column (4 × 100 mm; Dr. Maisch GmbH, Ammerbuch, Germany) and a fluorescence detector (Jasco Ltd., Hachioji City, Japan) with an excitation wavelength of 344 nm and an emission wavelength of 398 nm (18 nm bandwidth). A mobile phase of 50 mM sodium acetate (pH 6.2, adjusted with acetic acid) and 7.0% acetonitrile was pumped through the reversed-phase column at a flow rate of 0.5 ml/min. Brain samples (50 µl) were manually injected by a Rheodyne® 7725i injector (IDEX, Oak Harbor, WA, USA) into a 100 µl loop. A second mobile phase containing Zinc acetate (0.5 M, not pH adjusted) was delivered after the column at a flow rate of 10 ml/hour by a peristaltic pump (P-500; Pharmacia, Uppsala, Sweden). Signals from the fluorescence detector were transferred to a computer for analysis using Datalys Azur software (Grenoble, France). The retention time of KYNA was about 7–8 min. The sensitivity of the system was verified throughout the session by analysis of KYNA standards.

3.6.2 Electrochemical detection (paper I and II)

The HPLC system is coupled to an electrochemical detector (Coulochem III; ESA Inc., Chelmsford, MA, USA). A mobile phase consisting of 20mM sodium phosphate, 0.7 mM octanesulfonic acid and 10% acetonitrile (pH set to 3.2 using acetic acid) was pumped through a ReproSil-Pur C18 column (4 × 150 mm, Dr. Maisch GmbH), at a flow rate of 0.6ml/min, delivered by a LC-20AD VP HPLC pump (Shimadzu Corporation, Kyoto, Japan). Samples of 20 µl (kept at –25°C until analysis) were manually injected through a Rheodyne® 7725i injector (IDEX) into a 100µl loop. The retention time of 3-HK was about 8.5 min. Signals from the detector were transferred to a computer for analysis with Clarity (DataApex

Ltd, Prague, The Czech Republic).

3.7 Microarray analysis (paper III)

Animals were killed by cervical dislocation, and their brains were rapidly removed and placed on ice. The cerebellum was then separated from the cerebrum, and all samples were snap-frozen in liquid nitrogen and stored at -80°C until use. Tissues were homogenized in TRIzol reagent (Invitrogen, Fisher Scientific, UK Ltd, Loughborough, UK), and RNA was extracted following the manufacturer's protocol. Integrity of total RNA samples was assessed on an Agilent 2100 Bioanalyzer. All samples had an RNA Integrity Number (RIN) >7.5. 250 ng of total RNA were reverse-transcribed to cDNA from which biotin-labelled cRNA was synthesized and purified using an Illumina TotalPrep RNA Amplification kit (Ambion, Fisher Scientific) according to the manufacturer's instructions. The cRNA was hybridized to MouseWG-6 v2.0 Expression BeadChips, which allow the simultaneous quantification of 45,200 transcripts. The beadchips were scanned on an Illumina BeadArray Reader, and the raw microarray data were extracted using Illumina Genome Studio V2011.1 software. The data were normalized and background subtracted using ArrayTrack.1 Transcripts with low mean expression (fluorescent intensity <150 units, which was equivalent to the mean background fluorescent intensity across the whole slide) across all samples were removed. Data sets were compared by Welch t-test using Arraytrack, and only differentially expressed transcripts with $p < 0.05$ and >1.2-fold changes were included for subsequent analyses. The differentially-expressed transcript list was annotated with Arraytrack. In order to visualize groups of similarly differentially-expressed transcripts, hierarchically clustered heat maps were produced using GENE-E software.

3.8 Statistics

Data analyses were formed with the statistical software package GraphPad Prism® 6 (GraphPad Software Inc., San Diego, CA, USA) for Mac OS X. A p-value <0.05 was considered statistically significant.

Behavior tests including locomotor activity, trace fear conditioning, passive avoidance and social interaction were analyzed with two-way repeated measure (RM) analysis of variance (ANOVA) followed by Bonferroni post-hoc test. Elevated plus maze and light-dark box tests were analyzed by *t*-test or Mann–Whitney *U* test. Biochemical data were analyzed with Mann–Whitney *U* test in **paper I and II**, and one-way or two-way ANOVA followed by

Bonferroni post-hoc correction in **paper III and IV**. Microdialysis data were analyzed with two-way RM ANOVA followed by Bonferroni multiple comparison test. Percent PPI was calculated as follows: ((A-B)/A)*100, where A is the average startle magnitude of the 120 dB trials within each block, and B is the average startle magnitude of the prepulse trial in the same block. Basal average startle magnitude between groups was evaluated with a two-tailed unpaired *t*-test. Differences in percent PPI were calculated with two-way RM ANOVA (genotype x prepulse level or interstimulus interval) followed by Bonferroni post-hoc test.

4 RESULTS AND DISCUSSION

4.1 Paper I

Exposure to infection in early life increases the risk for schizophrenia development. Previous studies reported transient increases of brain KYNA concentration in neonatal influenza A/WSN/33 virus infected wild-type mice (Holtze et al., 2008) as well as in immunodeficient mice (Asp et al., 2010). While the neonatal infection was related to anxiety and impairments in working memory and PPI in adult immunodeficient mice (Asp et al., 2009) no such effects were observed in the wild-type mice (Asp et al., 2010). The aim of this paper is therefore to investigate locomotor response to a d-amphetamine challenge in adult wild-type mice neonatally infected with influenza A virus as well as to investigate whether neonatally elevated brain KYNA is associated with long-term behavior disturbances seen after infection in early life.

4.1.1 Neonatal virus infection or neonatal L-kynurenine administration enhance locomotor responsiveness to d-amphetamine in adult mice

The present study demonstrates that acute administration of d-amphetamine (5 mg/kg, i.p.) in adult life increased horizontal activity in neonatally influenza A/WSN/33 virus-infected mice and their respective controls. As compared to the uninfected control mice, neonatal infected mice displayed a more pronounced increase in d-amphetamine-induced locomotor response (**Figure 2**). The basal horizontal activity did not differ between neonatally virus-infected mice and their uninfected controls. Similar results were obtained in adult mice neonatally injected with kynurenine (**Figure 3**).

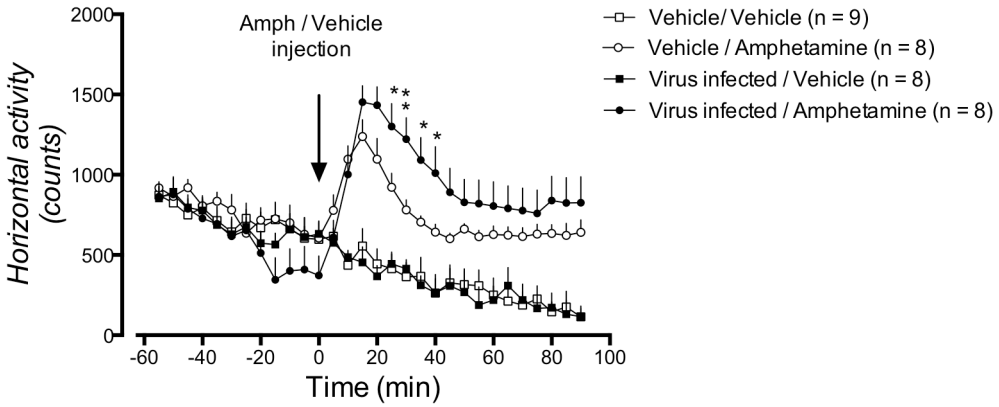

Figure 2. Horizontal activity during habituation three and following acutely administered d-amphetamine (5 mg/kg) or vehicle to 5–6 months old mice, injected with influenza A virus (2400 plaque-forming units) or phosphate buffered saline (PBS) at PND 3 or 4. Each point represents the mean ± S.E.M. of counts recorded during five-minute intervals. Statistical analysis was performed by a two-way ANOVA for repeated measurements (time × treatment) followed by Bonferroni's multiple comparison test (Interaction: $F(51, 493) = 3.629$, $p < 0.001$; Time: $F(17, 493) = 25.34$, $p < 0.001$; Treatment: $F(3, 29) = 16.29$, $p < 0.001$).

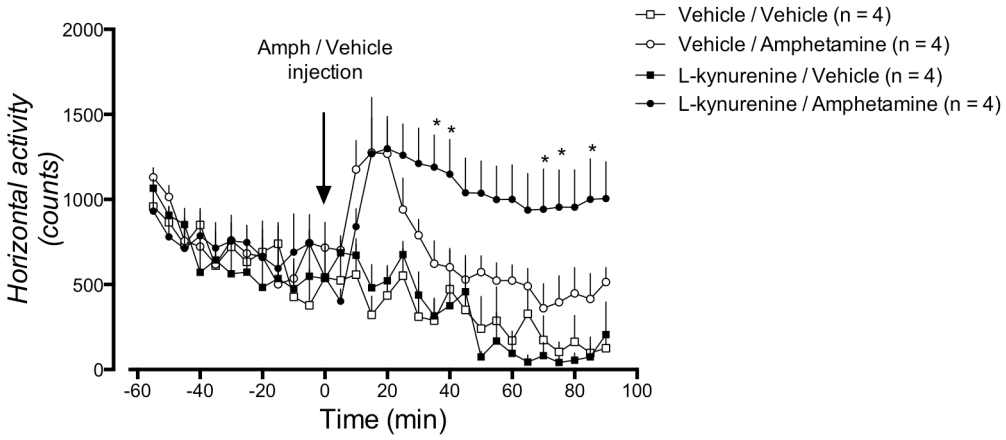

Figure 3. Horizontal activity during habituation three and following acutely administered d-amphetamine (5 mg/kg) or vehicle to 3–4 months old mice, injected with L- kynurenine (2 × 200 mg/kg/day) or saline from PND 7 to 16. Each point represents the mean ± S.E.M of counts recorded during five-minute intervals. Statistical analysis was performed by a two-way ANOVA for repeated measurements (time × treatment) followed by Bonferroni's multiple comparison test (Interaction: $F(51, 204) = 2.631$, $p < 0.001$; Time: $F(17, 204) = 10.61$, $p < 0.001$; Treatment: $F(3, 12) = 8.788$, $p < 0.01$).

4.1.2 Neonatal kynurenine treatment disrupts prepulse inhibition and working memory in adult mice

Neonatally L-kynurenine treated mice displayed mild reductions in PPI as adults (**Figure 4**). Significant disruption in PPI was only seen at the 500 ms interstimulus interval, suggesting reduced processing time in mice neonatally exposed to L-kynurenine.

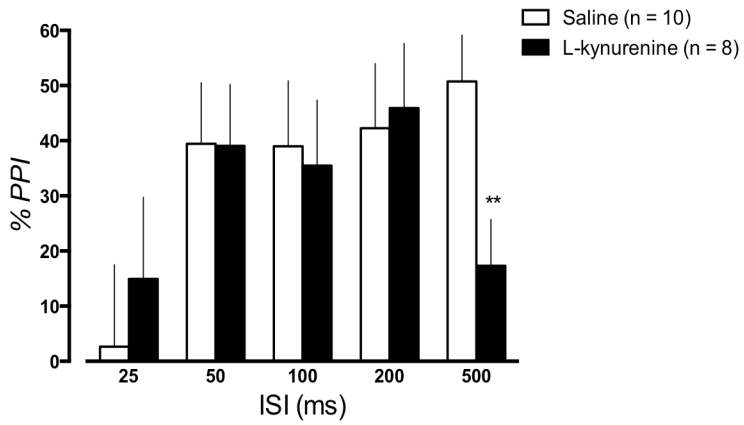

Figure 4. Prepulse inhibition (PPI) during the varied interstimulus interval (ISI) block of the startle session in 3–4 months old mice, neonatally injected with L-kynurenine (2 × 200 mg/kg/day, n = 8) or saline at PND 7–16 (n = 10). Data are presented as mean ± S.E.M. **p < 0.01.

Furthermore, neonatally L-kynurenine treated mice showed a reduction in working memory in trace fear conditioning test as adult (**Figure 5)**. However, no significant difference in contextual memory was found, implying a partial disruption of the hippocampus and/or other related brain regions in this developmental animal model. In line with the paper by Chess and colleagues (Chess et al., 2009), no tone-cued memory alterations were observed.

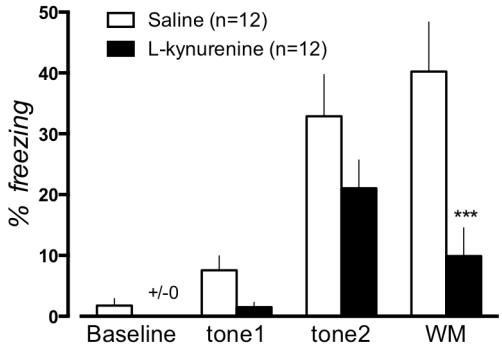

Figure 5. Trace fear conditioning of L-kynurenine treated and saline treated C57BL/6 female mice. Percent component time freezing during the training trial was analyzed in the following time periods: Baseline, first tone cue (Tone1), Inter-trial interval (ITI), second tone cue (Tone2) and working memory (WM). Data were analyzed with two-way ANOVA for repeated measurements (time × pretreatment) followed by Bonferroni multiple comparison test, which revealed a significant impairment of working memory in L-kynurenine treated mice, as compared to saline treated controls, (Treatment: F (1, 22) = 24.82, ***p < 0.001).

4.2 Decreased levels of prefrontal cortex KYNA in a genetic animal model of depression (Paper II)

In this paper, two key metabolites of the kynurenine pathway were analyzed in different brain regions of a rat model of depression, i.e. the FSL and FRL. Varied KYNA levels were detected in the brain areas of both FSL and FRL rats. No changes in KYNA levels in the frontal cortex, striatum, hippocampus or cerebellum were observed between the two strains. However, in the prefrontal cortex the concentration of KYNA levels was significantly lower in the FSL compared to FRL rats (**Table 1**).

Levels of 3-HK were analyzed in the prefrontal cortex and the frontal cortex in these rats and no differences were found between the two strains. As a result, the KYNA/3-HK ratio was significantly lower in the prefrontal cortex in FSL rats compared to FRL rats. These results indicate an imbalanced metabolism between the two main branches of the kynurenine pathway in this animal model of depression. The region specific abnormality of the kynurenine pathway metabolism implies an essential role of the prefrontal cortex in MDD. Indeed, several imaging studies have demonstrated abnormalities in neurophysiology of the prefrontal cortex in patients with MDD (Dutta et al., 2014; Johnstone et al., 2007; Liu et al., 2014b).

Table 1. Levels of KYNA, 3-HK and KYNA/3HK ratio in different brain regions of FSL and FRL rats

	KYNA		3-HK		KYNA/3-HK ratio	
	FSL	FRL	FSL	FRL	FSL	FRL
Frontal Cortex	10.0 [4.3-15.6](10)	8.5 [6.5-27.8] (10)	21.6 [16.3-27.8] (8)	19.0 [7.4-31.5] (10)	0.39 [0.2-0.6] (8)	0.66 [0.2-3.6](10)
Prefrontal cortex	3.8 [3.1-5.9]* (10)	14.5 [5.1-32.1] (10)	14.2 [12.5-19.1] (10)	12.8 [9.1-17.8] (10)	0.25 [0.2-0.4]** (10)	1.0 [0.4-2.1] (10)
Striatum	3.5 [2.7-4.7] (10)	4.0 [3.1-16.6] (10)	-	-	-	-
Hippocampus	10.3 [2.5-21.1](10)	8.5 [5.5-30.6] (10)	-	-	-	-
Cerebellum	4.9 [3.6-6.4] (10)	5.3 [3.4-7.6] (10)	-	-	-	-

Median concentrations [IQR] of KYNA and 3-HK are expressed in nM. Numbers of animals are given in brackets. Differences in KYNA, 3-HK levels and KYNA/3-HK ratios between FSL and FRL rats were evaluated by Mann-Whitney *U* test followed by the Bonferroni test for multiple comparison, *$p<0.05$, **$p<0.01$.

4.3 Paper III

In this study, mice with a targeted deletion of KMO were recruited to explore gene expression changes and behavioral alterations compared to wild-type mice.

4.3.1 Differential gene expression profiling identifies a network of schizophrenia-related genes in *Kmo*$^{-/-}$ mice

In order to explore the regulatory changes in the *Kmo*$^{-/-}$ mice, an unbiased screen for differentially expressed genes (DEGs) was performed. Gene profiling identified a number of DEGs in both cerebrum and cerebellum ($p \leq 0.05$). Of the two samples, the cerebrum exhibited a greater number of DEGs in *Kmo*$^{-/-}$ mice than in wild-type mice, with a total of 120 DEGs. In the cerebellum, a set of 24 genes was identified (**Figure 6**). However, only 6 genes overlapped between the forebrain and the cerebellum: CNIH4, FCER1G, LYPLAL1, MGST3, MYOC and SLC22A6. Interestingly, several identified genes have previously been implicated as dysfunctional in schizophrenia. In the cerebellum, we identified more upregulated genes associated with schizophrenia, but, in the cerebrum, identified DEGs that are linked to schizophrenia were more evenly distributed between up- and down-regulated categories.

Figure 6. Hierarchical clustering (Euclidian distance) was used to visualize the gene-expression changes relative to other genes in the dataset. Expression changes were sorted into clusters of similarly expressed genes both inter- and intra-dataset, across different brain regions in *Kmo*$^{-/-}$ relative to wild-type mice. Blue indicates a relative decrease whereas red indicates an increase. A greater number of significant gene-expression changes were observed in the cerebrum. Genes with significant ($p<0.05$) fold changes >1.2 are included in the analysis (n=4 per genotype).

Network analysis revealed a single robust interaction network in several functional clusters (**Figure 7**), supporting the idea that a large amount of the DEGs arise from genomic abolition of KMO activity – hereby linked to increased KYNA levels – and act in a common network.

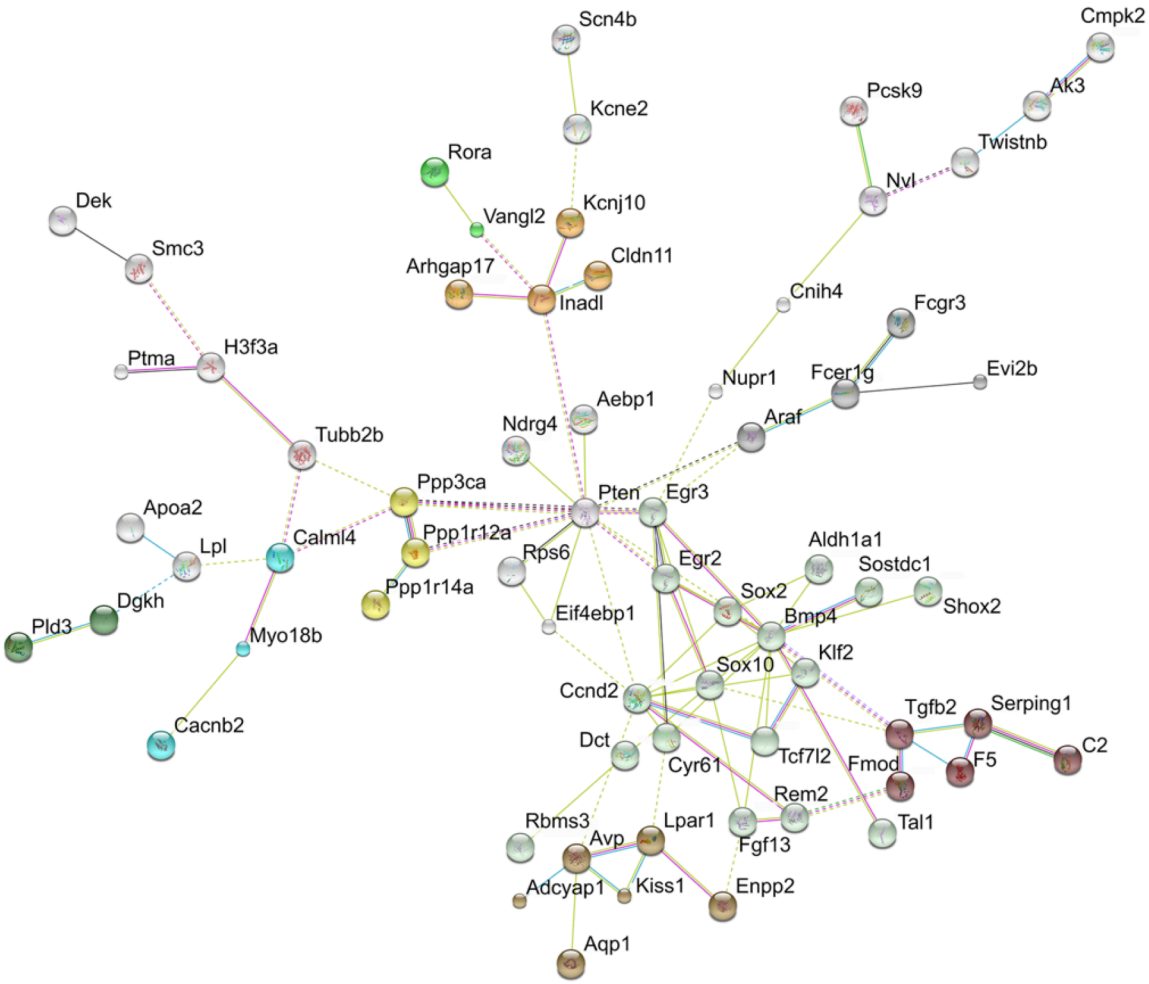

Figure 7. Differentially expressed genes (DEGs) form a highly interconnected network. Network analysis determined the DEGs identified in *Kmo*$^{-/-}$ mice form a robust network containing 67/144 of the candidates. The network is characterized by several functional clusters highlighted with different colors.

4.3.2 Brain KYNA levels are elevated in *Kmo*$^{-/-}$ mice

KMO activity, 3-HK levels and KYNA levels were measured in both cerebrum and cerebellum of *Kmo*$^{-/-}$ mice in order to compare kynurenine pathway changes in these two brain areas. KMO activity (**Figure 8A**) and the amount of its enzymatic product 3-HK (**Figure 8B**) were dramatically reduced in both cerebrum and cerebellum of *Kmo*$^{-/-}$ mice. Conversely, KYNA levels were significantly elevated in both brain areas in *Kmo*$^{-/-}$ mice, and unexpectedly, we observed higher KYNA levels in the cerebellum than in the cerebrum (**Figure 8C**).

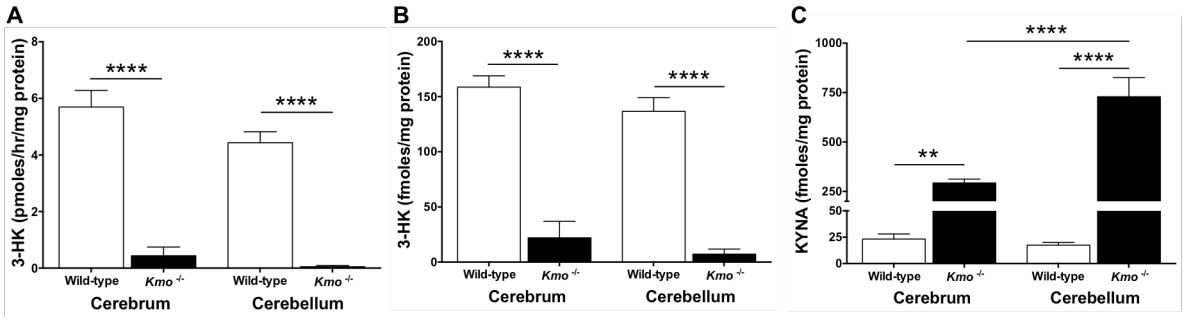

Figure 8. Kynurenine pathway metabolism in cerebrum and cerebellum of adult wild-type and *Kmo*$^{-/-}$ mice. (A) KMO activity is eliminated in both tissues in *Kmo*$^{-/-}$ mice. (B) Levels of 3-HK are reduced in both tissues in *Kmo*$^{-/-}$ compared to wild-type mice. (C) Levels of KYNA are elevated in *Kmo*$^{-/-}$ mice. KYNA levels are significantly more elevated in the cerebellum than in the cerebrum. All data are the mean ± SEM. **p<0.01; ***p<0.001; n=5-8 per group.

4.3.3 *Kmo*$^{-/-}$ mice display deficits in contextual memory and social interaction

Contextual memory was assessed in the passive avoidance task, a hippocampus-mediated behavioral test. *Kmo*$^{-/-}$ mice didn't display behavioral difference with wild-type mice in approach latencies during the acquisition trial (**Figure 9A**). However, after 24 hours, the avoidance latencies of *Kmo*$^{-/-}$ mice were significantly shorter than those of wild-type mice (**Figure 9A**). Deficits in hippocampus-dependent contextual memory are in line with the demonstration that elevated brain KYNA is associated with abnormalities in hippocampus-dependent learning and memory (Chess et al., 2009; Pocivavsek et al., 2012; Pocivavsek et al., 2011).

Social interaction was characterized with the three-chamber social approach apparatus. Both wild-type and *Kmo*$^{-/-}$ mice preferred to spend more time with the stranger than the novel object. Notably, when compare with wild-type animals, *Kmo*$^{-/-}$ mice spent a lower percentage

of time with the stranger mouse versus the novel object, suggesting a deficit in social interaction (**Figure 9B**). This observation is in agreement with data showing impaired social behavior in rats after kynurenine injection during early postnatal development or adolescence (Iaccarino et al., 2013; Trecartin and Bucci, 2011).

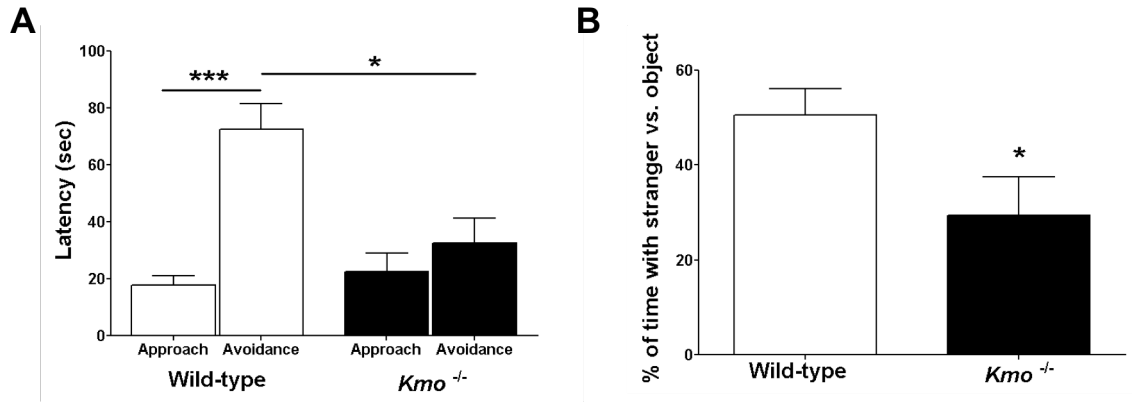

Figure 9. Contextual memory and social interaction. (**A**) Wild-type (n=14) and *Kmo*$^{-/-}$ (n=7) mice were tested in the passive avoidance paradigm. No genotypic difference in approach latency was observed on the training day. On Day 2, only wild-type animals showed contextual memory, i.e. a significant difference between avoidance and approach latency. Avoidance latency differed significantly between wild-type and *Kmo*$^{-/-}$ animals. (**B**) Performance of wild-type (n=12) and *Kmo*$^{-/-}$ (n=12) mice in the three-chambered social interaction paradigm. Compared to wild-type animals, mutant animals spent a lower proportion of time with the stranger mouse than with the novel object. All data are the mean ± SEM. *p<0.05; *** p<0.001.

4.3.4 *Kmo*$^{-/-}$ mice display anxiety-like behaviors

Similar with previous studies demonstrating an increase in anxiety-like phenotypes after acute or repeated systemic kynurenine administration in rodents (Olsson et al., 2012a; Salazar et al., 2012; Vecsei and Beal, 1990), *Kmo*$^{-/-}$ mice also displayed increased anxiety-like behaviors in the elevated plus-maze, the light-dark box, and open field tests. In the elevated plus-maze, *Kmo*$^{-/-}$ mice showed significant reductions in the percentage of time spent in open arms (**Figure 10A**) and in the number of entries into open arms (**Figure 10B**) compared to wild-type mice. In the light-dark box test, *Kmo*$^{-/-}$ mice spent significantly less time in the light compartment compared to their wild-type counterparts (**Figure 10C**) and made a decreased number of entries into the light compartment (**Figure 10D**). Furthermore, we observed a significant increase in the corner time of the *Kmo*$^{-/-}$ animals in a general assessment of locomotion (**Figure 10E**). Together, these data demonstrate increased anxiety-like behaviors in *Kmo*$^{-/-}$ mice compared to wild-type mice.

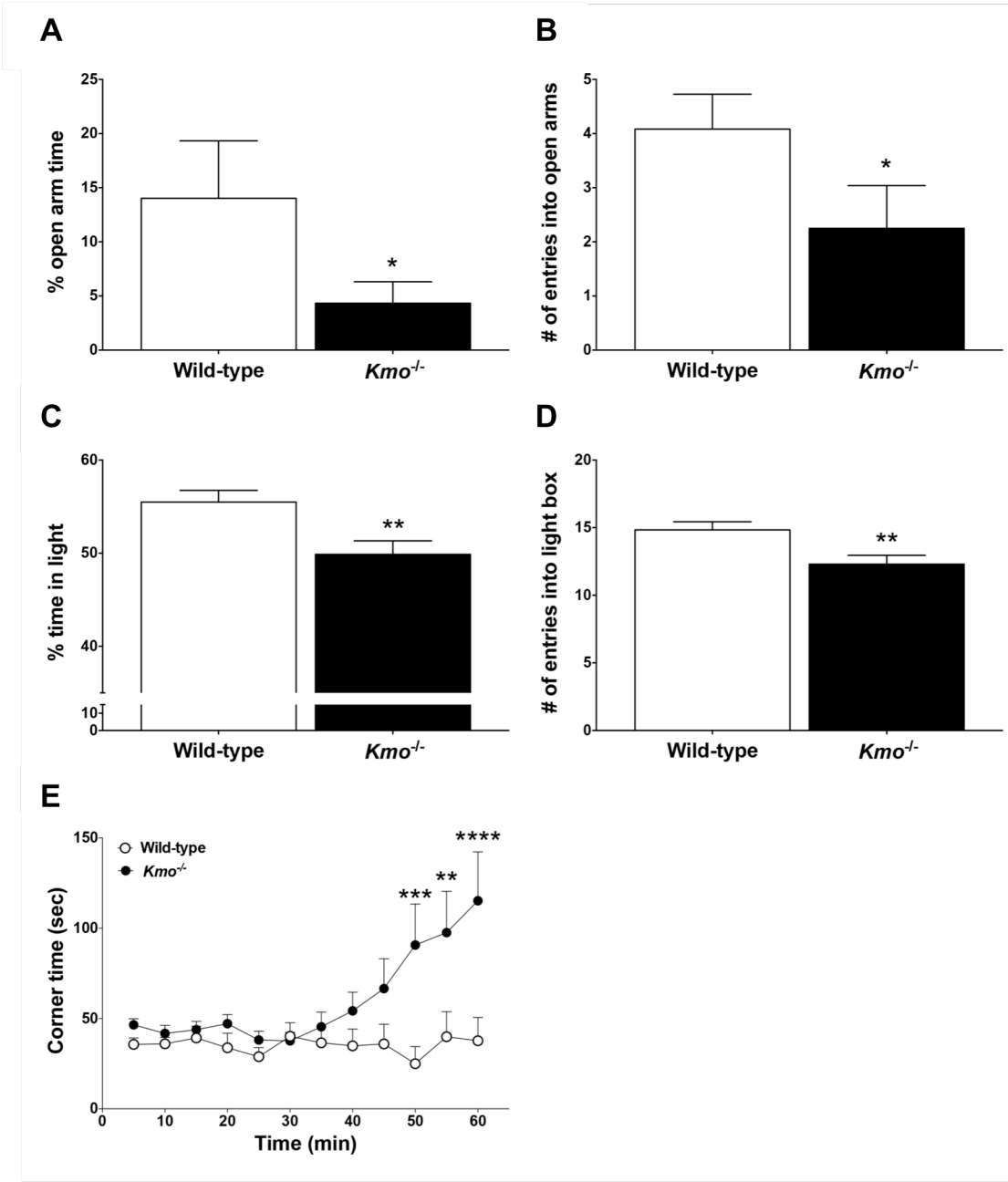

Figure 10. Anxiety behavior in elevated plus maze (**A, B**), light-dark box (**C, D**), and open field (**E**). In the elevated plus maze, *Kmo*$^{-/-}$ mice (n=12) spent significantly less time in the open arm (**A**) and entered the open arms less frequently (**B**) than wild-type animals (n=12); In the light-dark box, *Kmo*$^{-/-}$ mice (n=17) spent significantly less time in the light compartment (**C**) and entered the light compartment less frequently (**D**) than wild-type mice (n=24); (**E**) In the open field, *Kmo*$^{-/-}$ mice (n=21) spent more time in the corners than wild-type animals (n=23). All data are the mean ± SEM. *p<0.05, **p<0.01, ***p<0.001, ****p<0.0001.

4.3.5 *Kmo*$^{-/-}$ mice show enhanced locomotor response to d-amphetamine

Acute administration of d-amphetamine (5 mg/kg, i.p.) produced increased horizontal activity compared to saline treated mice and potentiated the increase in horizontal activity in *Kmo*$^{-/-}$ mice as compared to wild-type mice (Time: $F(29, 1160)=15.24$, $p<0.0001$; Genotype: $F(3, 40)=35.55$, $p<0.0001$; Interaction: $F(87, 1160)=17.88$, $p<0.0001$, **Figure 11A**). Central activity, indicating the movement of the animal in the center of the cage, in *Kmo*$^{-/-}$ mice was enhanced by acute administration of d-amphetamine compared to wild-type mice (Time: $F(29, 1160)=6.416$, $p<0.0001$; Genotype: $F(3, 40)=11.85$, $P<0.0001$; Interaction: $F(87, 1160)=6.117$, $p<0.0001$, **Figure 11B**). This enhanced response is also seen in mice with experimentally induced chronic elevations in brain KYNA levels (Olsson et al., 2009).

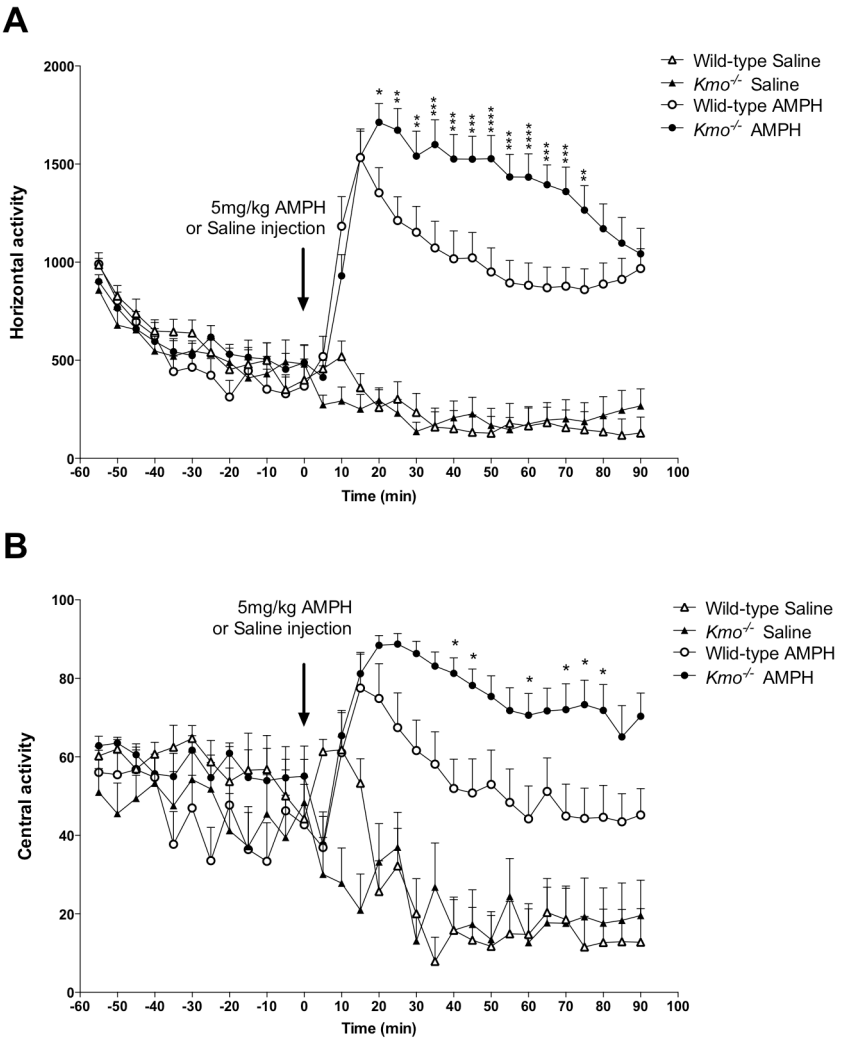

Figure 11. Increased locomotor activity after d-amphetamine (AMPH; 5 mg/kg). At time 0 (arrows), animals received an i.p. injection of either AMPH (wild-type: n=12; *Kmo*$^{-/-}$: n=11) or saline (wild-type: n=11; *Kmo*$^{-/-}$: n=10). AMPH increased both horizontal (**A**) and central (**B**) activity significantly more in *Kmo*$^{-/-}$ mice than in wild-type animals. All data are the mean ± SEM. *p<0.05, **p<0.01, ***p<0.001, ****p<0.0001 versus wild-type.

4.4 Paper IV

In the present study, mice with a targeted deletion of KAT II, hereby having less capacity to generate KYNA, were used as an experimental model to investigate if these mice are prevented from elevation of brain KYNA induced by an LPS triggered immune response. It has been shown that KAT II KO mice have lower levels of KYNA than wild-type mice before PND 28 (Yu et al., 2004).

4.4.1 Brain KYNA levels are lower in KAT II KO mice than in wild-type mice at PND 22

Baseline KYNA levels were measured in wild-type and KAT II KO mouse brains at PND 22 as well as in adults. KAT II KO mice exhibited lower brain KYNA levels compared to wild-type mice at PND 22 (mean ±SEM: wild-type: 1.42 ±0.41 nM (n=16); KAT II KO: 0.25 ±0.05 nM (n=16), **p<0.01, **Figure 12**), but no difference in baseline brain KYNA was found in the adult animals.

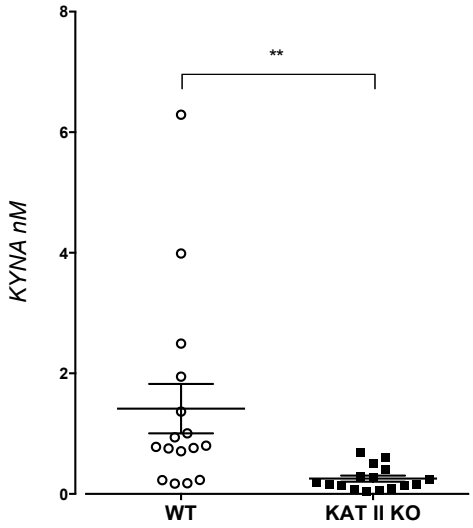

Figure 12. Baseline brain KYNA levels were measured in 22 days old wild-type (WT) and KAT II KO mice. n=16 in each group. Data are presented as mean ± SEM and analyzed with unpaired *t* test. **p<0.01

4.4.2 Administration of kynurenine or repeated LPS injections elevate brain KYNA levels in both wild-type and KAT II KO mice at PND 22.

Brain KYNA levels were measured after kynurenine injection (20mg/kg or 40mg/kg) or repeated LPS injections (2 × 0.83mg/kg). Surprisingly, brain KYNA levels were elevated in both wild-type and KAT II KO mice after kynurenine injection **(Figure 13A)** or repeated LPS injections **(Figure 13B)**. These results imply that other enzymes aside from KAT II might be responsible for the transamination of kynurenine to KYNA. In order to explore the

underlying mechanisms behind this unexpected observation, mRNA expression levels of the four KAT enzymes were measured.

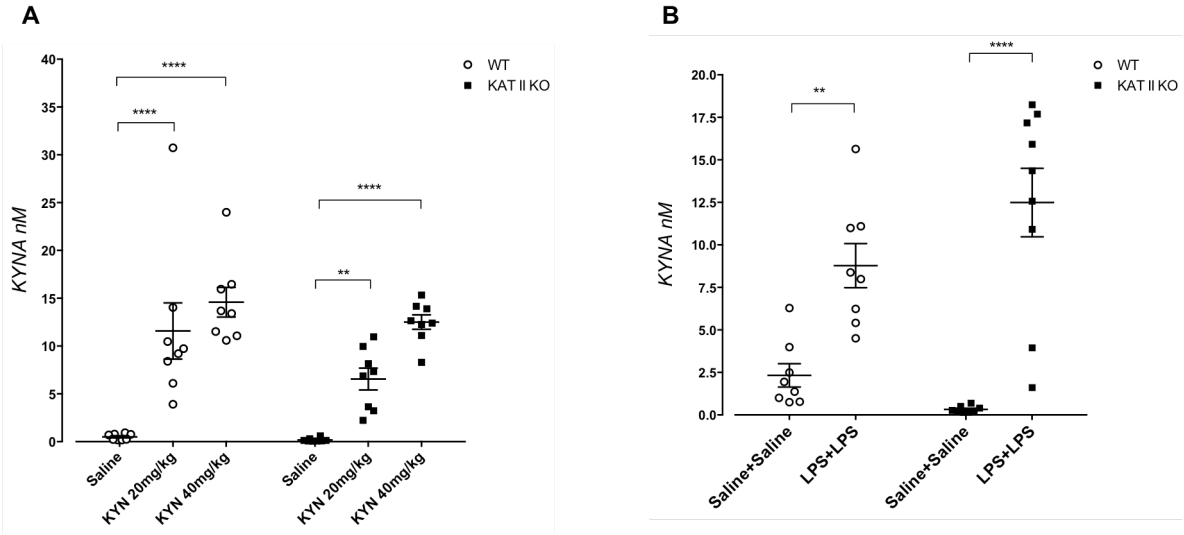

Figure 13. Brain KYNA levels were measured in wild-type (WT) and KAT II KO mice after kynurenine injection (**A**) or repeated LPS injections (**B**) at PND 22. n=8-9 in each group. Levels are presented as mean ± SEM. Data were analyzed using two-way ANOVA followed by Bonferroni's multiple comparisons test. **p<0.01, ****p<0.0001

4.4.3 Repeated LPS injections upregulate KAT III mRNA expression in wild-type and KAT II KO mice at PND 22.

By examining the mRNA expression level of KAT I/II/III and IV enzymes, significant effects of genotype were found in KAT I ($F(1, 20) = 116.0$, $p<0.0001$, **Figure 14A**), KAT III ($F(1, 20) = 57.07$, $p<0.0001$, **Figure 14C**) and KAT IV expression levels ($F(1, 20) = 9.750$, $p=0.0054$, **Figure 14D**). As expected, no KAT II mRNA was detected in KAT II KO mice (**Figure 14B**). Repeated LPS injections did not affect KAT I, KAT II or KAT IV expression levels. Rather, this treatment was associated with increased KAT III mRNA expression (Treatment: $F(1, 20) = 34.34$, $p<0.0001$, **Figure 14C**) in both wild-type and KAT II KO mice at PND 22 (Wild-type LPS+LPS vs Wild-type saline+saline: ***p<0.001; KO LPS+LPS vs KO saline+saline: *p<0.05, **Figure 14C**).

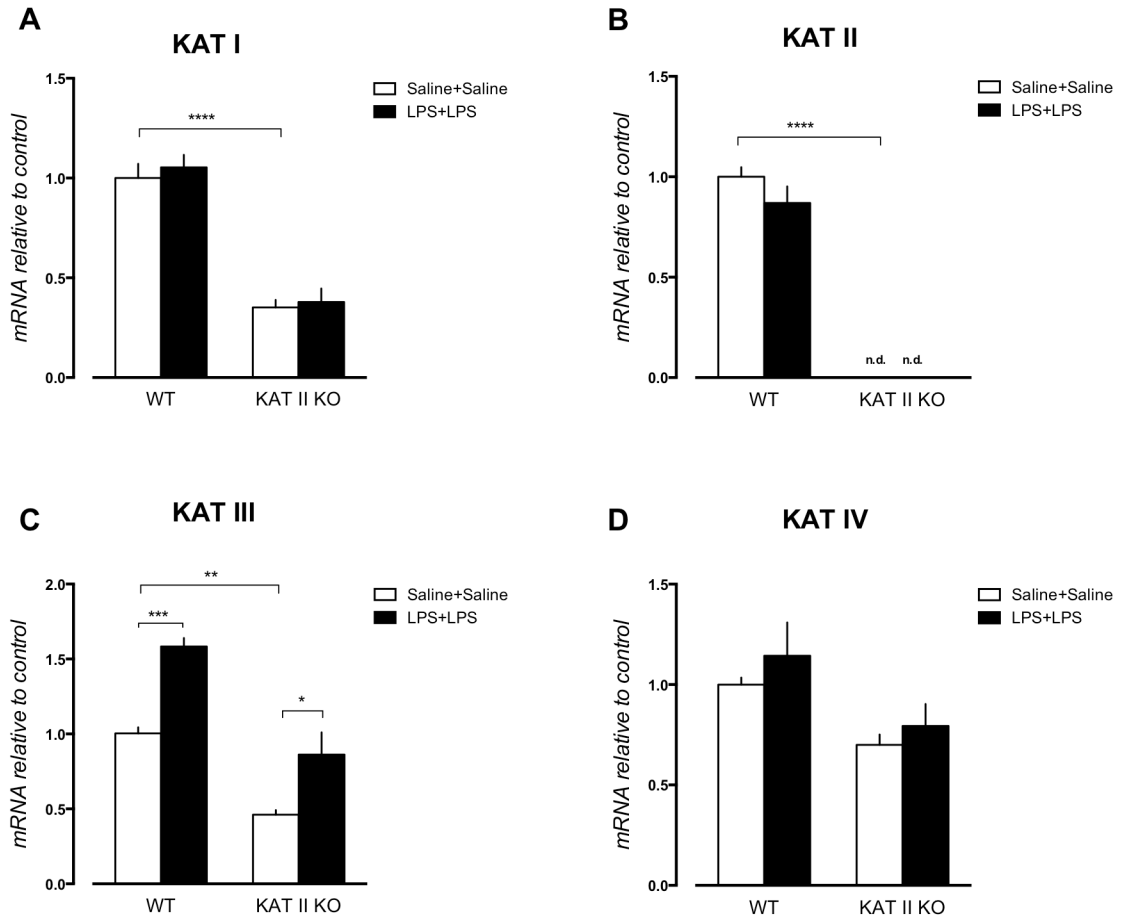

Figure 14. Brain mRNA expression levels of KAT I (**A**), KAT II (**B**), KAT III (**C**) and KAT IV (**D**) enzymes in wild-type (WT) and KAT II KO mice brain following repeated LPS injections (2 × 0.83mg/kg) at adult age. n=5-7 per group. Data are presented as mean ± SEM and analyzed using two-way ANOVA followed by Bonferroni's multiple comparisons test. *p<0.05, **p<0.01, ***p<0.001, ****p<0.0001.

Further, in order to investigate if LPS induces mRNA expression of KAT III or if increased concentration of kynurenine per se serves as the inducer, we investigated mRNA expression of KAT III following kynurenine administration. mRNA expression levels of all the four KAT enzymes were analyzed following administration of kynurenine and no significant changes were found between mice administered kynurenine and saline-treated mice (**Figure 15**).

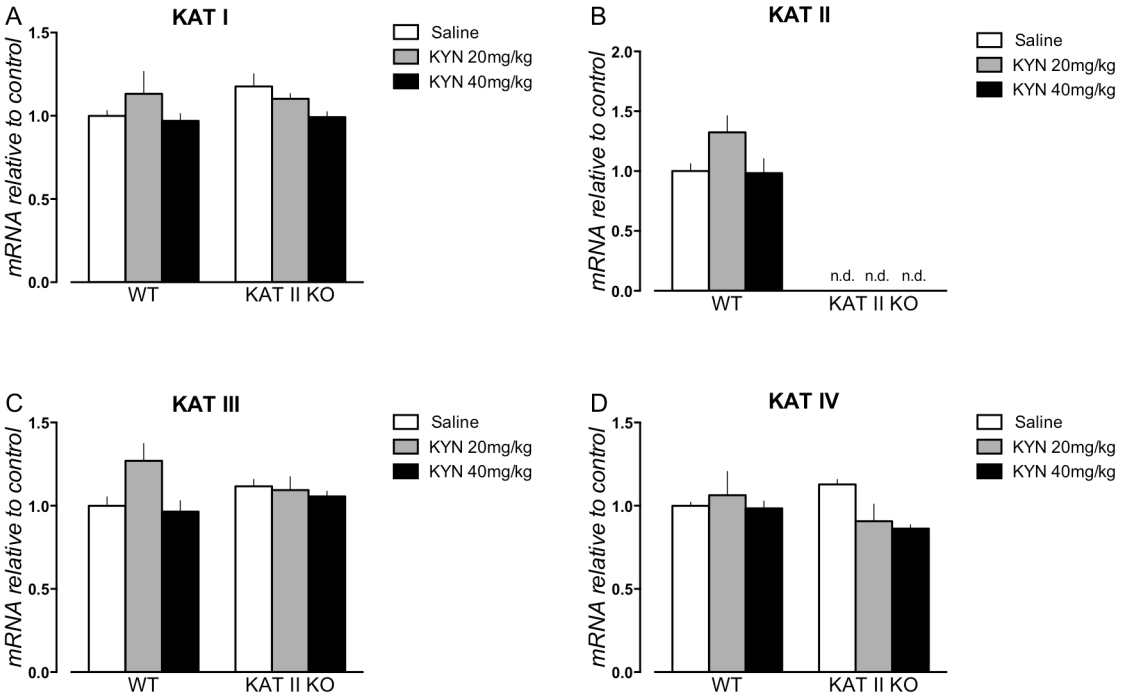

Figure 15. Brain mRNA expression levels of KAT I (A), KAT II (B), KAT III (C) and KAT IV (D) enzymes in wild-type and KAT II KO mice brain following kynurenine (KYN) injection (20mg/kg or 40mg/kg) at PND 22. n=3-6 per group. Data are presented as mean ± SEM and analyzed using two-way ANOVA followed by Bonferroni's multiple comparisons test.

4.4.4 Repeated LPS injections upregulate KAT III mRNA expression in KAT II KO and wild-type mice at adult age.

mRNA expression levels of KAT I/II/III and IV enzymes were measured in adult wild-type and KAT II KO mice brains. Significant genotype effects were found in KAT I ($F(1, 20) = 12.50$, $p=0.0021$, **Figure 16A**), KAT III ($F(1, 20) = 15.00$, $p=0.0009$, **Figure 16C**) and KAT IV ($F(1, 20) = 25.68$, $p<0.0001$, **Figure 16D**) mRNA expression levels. KAT II mRNA was not detected in KAT II KO mice (**Figure 16B**). Similar as the result from PND 22, repeated LPS administration induced KAT III expression (Treatment: $F(1, 20) = 27.66$, $p<0.0001$, **Figure 16C**) in both genotypes at this age (Wild-type LPS+LPS vs Wild-type saline+saline: *$p<0.05$; KO LPS+LPS vs KO saline+saline: **$p<0.01$, **Figure 16C**). However, when comparing the baseline levels of the four enzymes between the two genotypes, KAT II KO mice showed higher KAT III (Wild-type saline+saline vs KAT II KO saline+saline: *$p<0.05$, **Figure 16C**) and KAT IV (Wild-type saline+saline vs KAT II KO saline+saline: **$p<0.05$, **Figure 16D**) mRNA expression levels than their wild-type counterparts. These results are in

line with a previous study, showing an increased KAT III mRNA levels in KAT II KO mice at PND 60 (Yu et al., 2006).

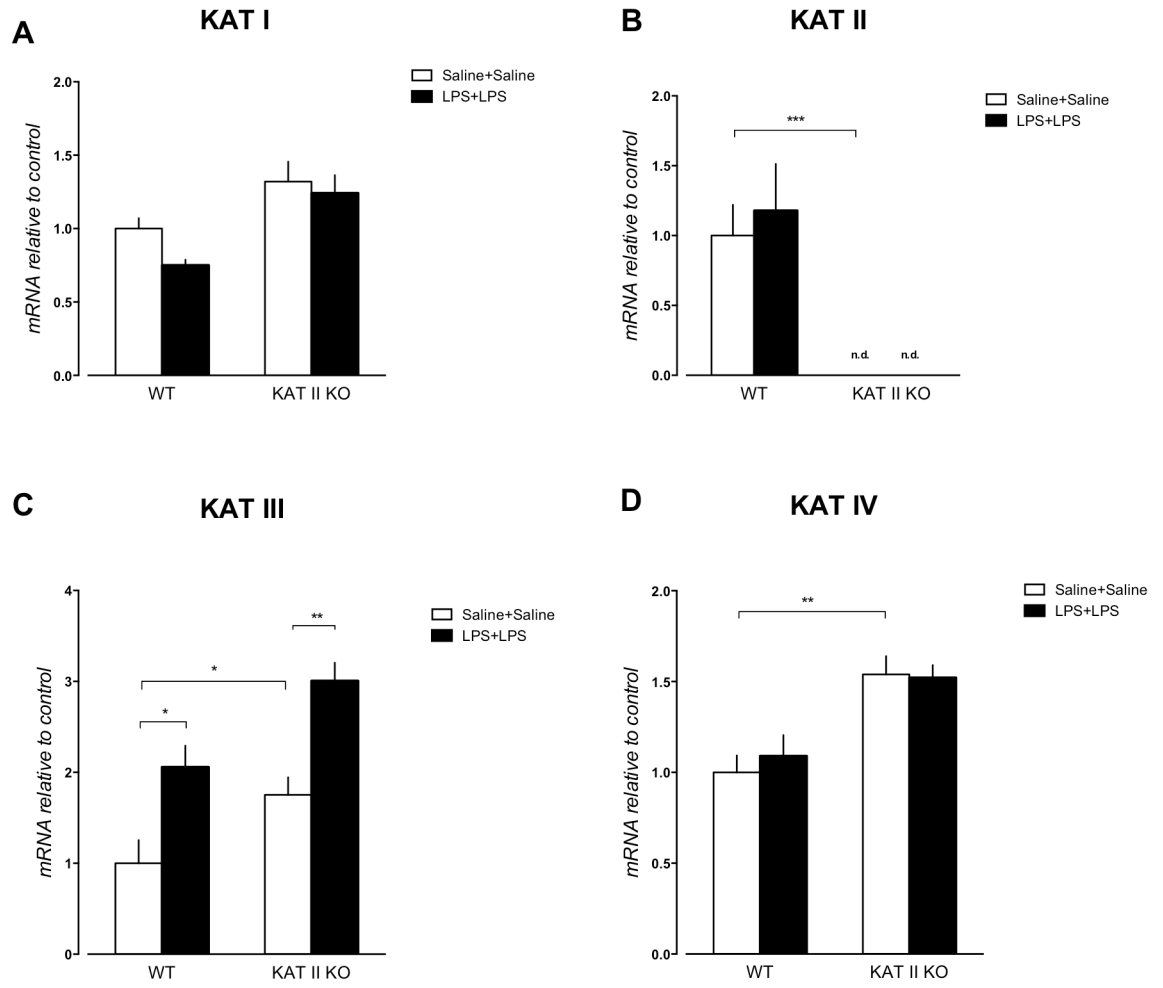

Figure 16. Brain mRNA expression levels of KAT I (**A**), KAT II (**B**), KAT III (**C**) and KAT IV (**D**) enzymes in wild-type (WT) and KAT II KO mice brain following repeated LPS injections (2 × 0.83mg/kg) at adult age. n=5-7 per group. Data are presented as mean ± SEM and analyzed using two-way ANOVA followed by Bonferroni's multiple comparisons test. *p<0.05, **p<0.01, ***p<0.001.

5 GENERAL DISCUSSION

During the last 15 years, growing evidence of an imbalanced regulation of the kynurenine pathway in the pathophysiology of psychiatric disorders has emerged. Thus, as shown in numerous studies, elevated levels of brain KYNA are found in patients with schizophrenia and bipolar disorder (Erhardt et al., 2001a; Linderholm et al., 2012; Miller et al., 2006; Nilsson et al., 2005; Olsson et al., 2010; Sathyasaikumar et al., 2011; Schwarcz et al., 2001) and shown to specifically correlate with psychotic features (Lavebratt et al., 2014; Olsson et al., 2012b; Sellgren et al., 2015) and impairments in cognitive functions (Sellgren et al., 2015). Reductions in both peripheral and brain KYNA concentrations have been observed in depressed patients (Bay-Richter et al., 2015; Savitz et al., 2015b; Schwarcz et al., 2012). In suicidal attempters, increased QUIN and decreased picolinic acid (PIC) has been observed (Bay-Richter et al., 2015; Brundin et al., 2016; Erhardt et al., 2013). Thus, the balance between the neuroprotective branch (i.e. KYNA) and the neurotoxic branch (i.e. QUIN), in all probability in combination with a genetic vulnerability, seems to be important for the development of either psychosis or depression.

Experimental studies support the theory that KYNA associates to psychosis and cognitive dysfunctions and that QUIN associates to depression. Thus, it has previously been shown in rodent models that increased levels of brain KYNA disrupt PPI (Erhardt et al., 2004), increase midbrain dopaminergic activity (Erhardt and Engberg, 2002; Erhardt et al., 2001b; Linderholm et al., 2007; Nilsson et al., 2006) and enhance amphetamine-induced dopamine release (Olsson et al., 2009) as well as the locomotor activity (Olsson et al., 2012a). Also cognitive dysfunctions such as impairments in contextual processing, spatial working memory and contextual memory have been found to associate with elevated levels of KYNA in several animal studies (Alexander et al., 2012; Chess et al., 2009; Chess et al., 2007; Pocivavsek et al., 2011). A previous study has shown that KAT II KO mice, i.e. mice with reduced capacity to produce KYNA, display improved cognitive function (Potter et al., 2010). With regard to animal models related to depression, it has been shown that IFN-α induces depressive-like behavior through the induction of the kynurenine pathway, and that this effect is accompanied by elevated hippocampal QUIN levels in the rat (Fischer et al., 2015). Also, chronic stress increases hippocampal QUIN levels and induces depressive-like behaviors in rats. Local administration of QUIN in the hippocampus results in similar depressive-like behaviors (Chen et al., 2013). In addition, LPS has been used as a reliable stimulator to construct an animal model of depression. Thus, several studies have shown that

both peripheral and local administration of LPS induces depressive-like behaviors in rodent (Fu et al., 2010; Lawson et al., 2013; O'Connor et al., 2009c; Salazar et al., 2012). In fact, also administration of a low dose of kynurenine has been shown to induce depressive-like behaviors (Agudelo et al., 2014). The same study also found that physical exercise induces antidepressant effect by inducing the expression of KATs, the enzymes responsible for the conversion of kynurenine towards KYNA, in skeletal muscle in both human and mouse (Agudelo et al., 2014).

In the present thesis, several animal models were used to investigate the role of kynurenine pathway in schizophrenia and depression. The results show that *Kmo*$^{-/-}$ mice, with high brain KYNA levels, display impairments in cognitive functions and social interaction as well as increased anxiety-like behaviors and enhanced locomotor activity following d-amphetamine administration. These data are in line with our findings in mice neonatally infected with influenza A virus or neonatally treated with kynurenine. Thus, neonatal administrations of neurotropic influenza A virus in wild-type mice lead to enhanced locomotor responsiveness to d-amphetamine. We have previously shown that such a neonatal infections causes a transient elevation of brain KYNA concentrations in mice (Asp et al., 2010; Holtze et al., 2008).

Here, we further investigated the association between elevated KYNA levels in early life and disturbed behaviors in adulthood. Thus, kynurenine, the precursor of KYNA was administrated to wild-type mice neonatally. Indeed, brain KYNA elevation in neonatal period was found to induce impairments in PPI, working memory as well as a tendency to induce hyper dopaminergic responsiveness to d-amphetamine.

In summary, findings of the present thesis reveal a critical role of KYNA in the development of schizophrenia. In support, several studies have shown that elevated levels of KYNA contribute to the neurochemical and behavioral abnormalities in neurodevelopmental animal models (Alexander et al., 2013; Liu et al., 2014a; Pershing et al., 2015; Pocivavsek et al., 2012). Taken together, these findings are in line with the neurodevelopmental theory of schizophrenia, suggesting that environmental insults in early life is a risk factor for schizophrenia development and neonatal elevation of brain KYNA levels might serve as a link between early life environmental insults and disturbed behaviors in adulthood.

The underlying reason for the increase in brain KYNA concentration in schizophrenia and bipolar disorder with psychotic features may be related to polymorphisms in the KMO gene (Holtze et al., 2012; Lavebratt et al., 2014; Wonodi et al., 2011). Interestingly, our genome-

wide differential gene expression analyses in *Kmo*$^{-/-}$ mice identified a network of schizophrenia- and psychosis- related genes.

In addition, enhanced secretion of pro-inflammatory cytokines may also be important for the observed increase in brain KYNA. Thus, elevated levels of IL-1β have been observed in first-episode schizophrenia patients and in patients with bipolar disorder (Beumer et al., 2012; Dowlati et al., 2010; Lindqvist et al., 2009; Schwieler et al., 2015; Sellgren et al., 2015; Soderlund et al., 2011; Soderlund et al., 2009), whereas elevated levels of IL-6 have been shown in chronic schizophrenia, depression and suicidal attempters (Erhardt et al., 2013; Sasayama et al., 2013; Schwieler et al., 2015). Indeed, pro-inflammatory cytokines induce the kynurenine pathway by activating rate limiting enzymes, IDO1 and TDO2 (Carlin et al., 1987; Miller et al., 2006; Sellgren et al., 2015; Yoshida et al., 1986). Cytokines, such as IFN-γ, IFN-α, IFN-β and TNF-α, have been shown to induce IDO1 in several studies (Guillemin et al., 2001a; Pemberton et al., 1997). Recently, studies have shown that IL-1β has the potency to induce TDO2 (Sellgren et al., 2015; Urata et al., 2014). In fact, post mortem studies of patients with schizophrenia and patients with bipolar disorder with psychotic features have found increased expression of TDO2. In the present study, LPS was used as a tool to induce the secretion of pro-inflammatory cytokines. We have recently shown that a double-shot of LPS induces not only the kynurenine pathway but also increase brain KYNA levels (Larsson et al., 2016). Here, KAT II KO mice were used to investigate if loss of KAT II could prevent the increase of KYNA induced by a double-shot of LPS. To our big surprise, repeated injections of LPS up-regulated brain KYNA levels in not only wild-type mice but also in KAT II KO mice. Indeed, mRNA expression data show evidence of increased KAT III mRNA levels in both KAT II KO and wild-type mice. These data imply that under pathophysiological conditions, where the central immune system is triggered, KAT III might be the most prominent enzyme converting kynurenine towards KYNA.

The outcome of the present thesis provides novel information regarding underlying pathological mechanisms in schizophrenia and depression. These findings may have the potential to lay grounds for novel pharmacological interventions, targeting on specific kynurenine pathway enzymes.

6 ACKNOWLEDGEMENTS

I would like to express my deepest gratitude to everyone who has helped, supported and inspired me during my Ph.D. life at Karolinska Institutet.

In particular, I would like to thank:

My main supervisor, Professor **Sophie Erhardt**. Your great encouragement and inspiration provided me with lots of motivation throughout my Ph.D. life. Thank you for believing in me, so I could rid myself of self-doubt, and persevere through any challenges. Your energy, enthusiasm and passion for science showed me how a real scientist should be! And I'm very appreciative for all the visits and conferences to the rest scientific world.

Professor **Göran Engberg**, for giving me an opportunity for an interview and accepting me as a project student in this lab in 2012. Your optimistic spirit and creative thinking made my challenging journey much more enjoyable. Thank you for teaching me how to properly write papers. I learned a lot from working with you!

My co-supervisor, Dr. **Lilly Schwieler**, for all the fun talks, your patience, and sharing all your detailed knowledge with me. I am especially grateful for your help and advice on preforming enzyme activity.

My co-supervisor, Associate Professor **Simon Cervenka**, for all the wonderful discussions and considerations. Your clinical perspectives made my laboratory work much more meaningful.

My mentor, **Hong Li**, for introducing me to Sweden, Karolinska, science and research. You are the best mentor and a very good friend to me. I really enjoy all the conversations we had. I owe you a big thank you for your help in all aspects of my life here. I would also like to thank **Tao Liu** for all the interesting and inspirational stories, as well as all the tasty food at your place. You are like a family for me in Sweden, without you I would have never started my journey to this wonderful country.

All present and former members of the Erhardt/Engberg research group for your support and help. Special thanks to:

Michel Goiny, for all your helps, explanations and troubleshooting regarding HPLC.

Funda Orhan, for your super kind heart, sweet and innocent attitude, as well as all your companionship inside and outside of work.

Anthi Faka, for being concerned about me all the time, and providing an energetic presence every day.

Sophie Imbeault, for being cool and wise, and answering all my questions regarding science, language and Canada.

Maximilian Tufvesson Alm, for all the Swedish translations and encouraging me to start writing my thesis early.

Markus Larsson, for helping me to start using Mac, for all the funny conversations and laughers we shared and for all your patience in answering my questions.

Alexandra Andersson, for your understanding and encouragement when I was feeling stressed.

Magdalena Kegel, for making me feel like home when I had just joined the group.

Maria Hotlze, for sharing your knowledge and guiding me into the behavior world.

Sara Olsson, for teaching me experimental skills and for sharing all your interesting stories.

Klas Lindholm, for being kind, wise and supportive.

Carl Sellgren, for helping me with statistical issues.

Additional members and collaborators: **Anna Malmqvist, Micke Hedberg, Vincent Millischer, Oscar Jungholm**, **Dag Holmberg**, for nice discussions and being supportive.

The AstraZeneca Science Translational Center, for financial support throughout the thesis and all researchers involved in the AstraZeneca - Karolinska Institutet Joint Research Program: **Maria Bhat, Kristian Sandberg, Sara Ståhl, John Dunlop, Nick Brandon, Marie Svedberg, Fredrik Piehl, Jenny Häggkvist, Ida Nilsson, Peter Johnström, Magnus Schou, Lars Farde, Karin Collste, Miklós Tóth, Lenke Tari, Evgeny Revunov as well as Ratan Bhat and Kaushik Sengupta** at AZ**.**

All co-authors: **Robert Schwarcz, Flaviano Giorgini, Aleksander Mathé, Ana Pocivavsek, Paul Muchowski, Susan Powell, Niccolò Terrando, Anna Persson, Linnea Asp, Håkan Karlsson, Mariaelena Repici, Daniel Maddison, Marian Thomas, Joshua Smalley,** for nice collaborations and discussions.

Professor **Doo-sup Choi, Sun Choi** and **Alfredo Oliveros**, for nice discussions and for welcoming me in your lab at Mayo Clinic in Rochester, USA.

All the former and present colleagues and friends in the Department of Physiology and Pharmacology: **Ming Liu, Tianle Gao, Lei Li, Meng Xie, Kent Jardemark, Jana Valnohova, Adi Zheng, Boxi Zhang, Zhengbing Zhuge, Varsha Prakash, Miyoung Lee, Phillip Newton, Jie Su, Meishan Li** for the pleasant conversations and support. **Jingxia Hao** and **Xiaojun Xu** for the great food, valuable advice and generosity. **Camilla I. Svensson** for sharing your lab and skills.

The staff at the Department of Physiology and Pharmacology: **Stefan Eriksson, Camilla Fors Holmberg, Sara Kullgren, Sofia Schilken, Sofia Pettersson, Katariina Välimäki, Inger Johansson, Micke Elm, Hina Mohsin, Renee Andersson, Ylva Haraldsdotter** for all your help with everything. Also a big thanks to the staff at the animal department, especially **Josefin Lilja, Martina Andersson, Per-Arne Åberg, Paulina Lenngren Hysing.**

Professor **Ulf Eriksson**, for inviting me to Sweden and starting my research life.

Financial support from the Swedish Medical Research Council, the Karolinska Institutets stiftelse för virus forskning, Stiftelsen Sigurd och Elsa Golje Minne, Svenska Läkaresällskapet, Åhléns-stiftelsen, MH091407 (SBP), the Stanley Medical Research Institute, NIH grants, Impact Award from Higher Education Innovation Fund, Swedish Brain Foundation, Torsten Söderbergs Stiftelse, Petrus och Augusta Hedlunds Stiftelse, Professor Bror Gadelius Minne.

My lovely friends: **Jia Guo, Tingting Guan, Yan Chen, Qing Shen, Tiansheng Shi, Xiaoxiao Peng, Zi Ning, Kewei Zhang, Du Liu, Shisong Gao, Xun Wang, Ying Sun, Liqun He, Xiaoyuan Ren, Jiangrong Wang, Shuting Gao, Bin Xiao, Yi Jin, Xiaojing Tang, Wenming Hao**, for all the wonderful memories we created together. You really brightened my life in Stockholm!

Last and foremost, my dear **parents,** thank you for your endless love and all the effort you made to bring me up, for always believing in me, encouraging me and supporting me! I love you so much!

感谢我的父母和家人给予我无限的爱, 你们的支持和鼓励是我前进的动力!

7 REFERENCES

Aberg, K.A., Liu, Y., Bukszar, J., McClay, J.L., Khachane, A.N., Andreassen, O.A., Blackwood, D., Corvin, A., Djurovic, S., Gurling, H., Ophoff, R., Pato, C.N., Pato, M.T., Riley, B., Webb, T., Kendler, K., O'Donovan, M., Craddock, N., Kirov, G., Owen, M., Rujescu, D., St Clair, D., Werge, T., Hultman, C.M., Delisi, L.E., Sullivan, P., van den Oord, E.J., 2013. A comprehensive family-based replication study of schizophrenia genes. JAMA Psychiatry 70, 573-581.

Abi-Dargham, A., Gil, R., Krystal, J., Baldwin, R.M., Seibyl, J.P., Bowers, M., van Dyck, C.H., Charney, D.S., Innis, R.B., Laruelle, M., 1998. Increased striatal dopamine transmission in schizophrenia: confirmation in a second cohort. Am J Psychiatry 155, 761-767.

Abi-Dargham, A., Laruelle, M., 2005. Mechanisms of action of second generation antipsychotic drugs in schizophrenia: insights from brain imaging studies. Eur Psychiatry 20, 15-27.

Agudelo, L.Z., Femenia, T., Orhan, F., Porsmyr-Palmertz, M., Goiny, M., Martinez-Redondo, V., Correia, J.C., Izadi, M., Bhat, M., Schuppe-Koistinen, I., Pettersson, A.T., Ferreira, D.M., Krook, A., Barres, R., Zierath, J.R., Erhardt, S., Lindskog, M., Ruas, J.L., 2014. Skeletal muscle PGC-1alpha1 modulates kynurenine metabolism and mediates resilience to stress-induced depression. Cell 159, 33-45.

Alberati-Giani, D., Ricciardi-Castagnoli, P., Kohler, C., Cesura, A.M., 1996. Regulation of the kynurenine metabolic pathway by interferon-gamma in murine cloned macrophages and microglial cells. J Neurochem 66, 996-1004.

Albers, G.W., Clark, W.M., Atkinson, R.P., Madden, K., Data, J.L., Whitehouse, M.J., 1999. Dose escalation study of the NMDA glycine-site antagonist licostinel in acute ischemic stroke. Stroke 30, 508-513.

Alexander, E., 1904. Die entstehung der Kynurensäure. Z. Physiol.Chem. 43, 325-337.

Alexander, K.S., Pocivavsek, A., Wu, H.Q., Pershing, M.L., Schwarcz, R., Bruno, J.P., 2013. Early developmental elevations of brain kynurenic acid impair cognitive flexibility in adults: reversal with galantamine. Neuroscience 238, 19-28.

Alexander, K.S., Wu, H.Q., Schwarcz, R., Bruno, J.P., 2012. Acute elevations of brain kynurenic acid impair cognitive flexibility: normalization by the alpha7 positive modulator galantamine. Psychopharmacology (Berl) 220, 627-637.

Allen, R.M., Young, S.J., 1978. Phencyclidine-induced psychosis. Am J Psychiatry 135, 1081-1084.

Andreasen, N.C., Olsen, S., 1982. Negative v positive schizophrenia. Definition and validation. Arch Gen Psychiatry 39, 789-794.

Angrist, B., Sathananthan, G., Wilk, S., Gershon, S., 1974. Amphetamine psychosis: behavioral and biochemical aspects. J Psychiatr Res 11, 13-23.

Anis, N.A., Berry, S.C., Burton, N.R., Lodge, D., 1983. The dissociative anaesthetics, ketamine and phencyclidine, selectively reduce excitation of central mammalian neurones by N-methyl-aspartate. Br J Pharmacol 79, 565-575.

Ardolino, M., Hsu, J., Raulet, D.H., 2015. Cytokine treatment in cancer immunotherapy. Oncotarget 6, 19346-19347.

Asp, L., Beraki, S., Kristensson, K., Ogren, S.O., Karlsson, H., 2009. Neonatal infection with neurotropic influenza A virus affects working memory and expression of type III Nrg1 in adult mice. Brain Behav Immun 23, 733-741.

Asp, L., Holtze, M., Powell, S.B., Karlsson, H., Erhardt, S., 2010. Neonatal infection with neurotropic influenza A virus induces the kynurenine pathway in early life and disrupts sensorimotor gating in adult Tap1-/- mice. Int J Neuropsychopharmacol 13, 475-485.

Bailey, S.L., Carpentier, P.A., McMahon, E.J., Begolka, W.S., Miller, S.D., 2006. Innate and adaptive immune responses of the central nervous system. Crit Rev Immunol 26, 149-188.

Bardgett, M.E., Jacobs, P.S., Jackson, J.L., Csernansky, J.G., 1997. Kainic acid lesions enhance locomotor responses to novelty, saline, amphetamine, and MK-801. Behav Brain Res 84, 47-55.

Bay-Richter, C., Linderholm, K.R., Lim, C.K., Samuelsson, M., Traskman-Bendz, L., Guillemin, G.J., Erhardt, S., Brundin, L., 2015. A role for inflammatory metabolites as modulators of the glutamate N-methyl-D-aspartate receptor in depression and suicidality. Brain Behavior and Immunity 43, 110-117.

Beadle, G.W., Mitchell, H.K., Nyc, J.F., 1947. Kynurenine as an Intermediate in the Formation of Nicotinic Acid from Tryptophane by Neurospora. Proc Natl Acad Sci U S A 33, 155-158.

Berlinguer-Palmini, R., Masi, A., Narducci, R., Cavone, L., Maratea, D., Cozzi, A., Sili, M., Moroni, F., Mannaioni, G., 2013. GPR35 Activation Reduces Ca2+ Transients and Contributes to the Kynurenic Acid-Dependent Reduction of Synaptic Activity at CA3-CA1 Synapses. Plos One 8.

Beumer, W., Gibney, S.M., Drexhage, R.C., Pont-Lezica, L., Doorduin, J., Klein, H.C., Steiner, J., Connor, T.J., Harkin, A., Versnel, M.A., Drexhage, H.A., 2012. The immune theory of psychiatric diseases: a key role for activated microglia and circulating monocytes. J Leukoc Biol 92, 959-975.

Birch, P.J., Grossman, C.J., Hayes, A.G., 1988. Kynurenic acid antagonises responses to NMDA via an action at the strychnine-insensitive glycine receptor. Eur J Pharmacol 154, 85-87.

Bitanihirwe, B.K., Peleg-Raibstein, D., Mouttet, F., Feldon, J., Meyer, U., 2010. Late prenatal immune activation in mice leads to behavioral and neurochemical abnormalities relevant to the negative symptoms of schizophrenia. Neuropsychopharmacology 35, 2462-2478.

Black, D.W., Fisher, R., 1992. Mortality in Dsm-Iiir Schizophrenia. Schizophrenia Research 7, 109-116.

Bleuler, E., 1911. Dementia praecox, oder Gruppe der Schizophrenien. Deuticke, Leipzig.

Bluthe, R.M., Walter, V., Parnet, P., Laye, S., Lestage, J., Verrier, D., Poole, S., Stenning, B.E., Kelley, K.W., Dantzer, R., 1994. Lipopolysaccharide induces sickness behaviour in rats by a vagal mediated mechanism. C R Acad Sci III 317, 499-503.

Braff, D., Stone, C., Callaway, E., Geyer, M., Glick, I., Bali, L., 1978. Prestimulus effects on human startle reflex in normals and schizophrenics. Psychophysiology 15, 339-343.

Braff, D.L., Geyer, M.A., 1990. Sensorimotor gating and schizophrenia. Human and animal model studies. Arch Gen Psychiatry 47, 181-188.

Brake, W.G., Flores, G., Francis, D., Meaney, M.J., Srivastava, L.K., Gratton, A., 2000. Enhanced nucleus accumbens dopamine and plasma corticosterone stress responses in adult rats with neonatal excitotoxic lesions to the medial prefrontal cortex. Neuroscience 96, 687-695.

Breier, A., 1999. Cognitive deficit in schizophrenia and its neurochemical basis. Br J Psychiatry Suppl, 16-18.

Breier, A., Adler, C.M., Weisenfeld, N., Su, T.P., Elman, I., Picken, L., Malhotra, A.K., Pickar, D., 1998. Effects of NMDA antagonism on striatal dopamine release in healthy subjects: application of a novel PET approach. Synapse 29, 142-147.

Breier, A., Su, T.P., Saunders, R., Carson, R.E., Kolachana, B.S., de Bartolomeis, A., Weinberger, D.R., Weisenfeld, N., Malhotra, A.K., Eckelman, W.C., Pickar, D., 1997. Schizophrenia is associated with elevated amphetamine-induced synaptic dopamine concentrations: Evidence from a novel positron emission tomography method. P Natl Acad Sci USA 94, 2569-2574.

Bromet, E., Andrade, L.H., Hwang, I., Sampson, N.A., Alonso, J., de Girolamo, G., de Graaf, R., Demyttenaere, K., Hu, C., Iwata, N., Karam, A.N., Kaur, J., Kostyuchenko, S., Lepine, J.P., Levinson, D., Matschinger, H., Mora, M.E., Browne, M.O., Posada-Villa, J., Viana, M.C., Williams, D.R., Kessler, R.C., 2011. Cross-national epidemiology of DSM-IV major depressive episode. BMC Med 9, 90.

Brown, A.S., Derkits, E.J., 2010. Prenatal infection and schizophrenia: a review of epidemiologic and translational studies. Am J Psychiatry 167, 261-280.

Brown, A.S., Hooton, J., Schaefer, C.A., Zhang, H., Petkova, E., Babulas, V., Perrin, M., Gorman, J.M., Susser, E.S., 2004. Elevated maternal interleukin-8 levels and risk of schizophrenia in adult offspring. Am J Psychiatry 161, 889-895.

Brown, A.S., Susser, E.S., Butler, P.D., Richardson Andrews, R., Kaufmann, C.A., Gorman, J.M., 1996. Neurobiological plausibility of prenatal nutritional deprivation as a risk factor for schizophrenia. J Nerv Ment Dis 184, 71-85.

Brundin, L., Sellgren, C.M., Lim, C.K., Grit, J., Palsson, E., Landen, M., Samuelsson, M., Lundgren, K., Brundin, P., Fuchs, D., Postolache, T.T., Traskman-Bendz, L., Guillemin, G.J., Erhardt, S., 2016. An enzyme in the kynurenine pathway that governs vulnerability to suicidal behavior by regulating excitotoxicity and neuroinflammation. Transl Psychiatry 6, e865.

Buka, S.L., Tsuang, M.T., Torrey, E.F., Klebanoff, M.A., Wagner, R.L., Yolken, R.H., 2001. Maternal cytokine levels during pregnancy and adult psychosis. Brain Behav Immun 15, 411-420.

Burton, N., 2012. Living With Schizophrenia. Acheron Press, 3.

Cannon, T.D., Kaprio, J., Lonnqvist, J., Huttunen, M., Koskenvuo, M., 1998. The genetic epidemiology of schizophrenia in a Finnish twin cohort. A population-based modeling study. Arch Gen Psychiatry 55, 67-74.

Capuron, L., Dantzer, R., 2003. Cytokines and depression: the need for a new paradigm. Brain Behav Immun 17 Suppl 1, S119-124.

Capuron, L., Gumnick, J.F., Musselman, D.L., Lawson, D.H., Reemsnyder, A., Nemeroff, C.B., Miller, A.H., 2002. Neurobehavioral effects of interferon-alpha in cancer patients: phenomenology and paroxetine responsiveness of symptom dimensions. Neuropsychopharmacology 26, 643-652.

Capuron, L., Neurauter, G., Musselman, D.L., Lawson, D.H., Nemeroff, C.B., Fuchs, D., Miller, A.H., 2003. Interferon-alpha-induced changes in tryptophan metabolism: Relationship to depression and paroxetine treatment. Biol Psychiat 54, 906-914.

Cardno, A.G., Marshall, E.J., Coid, B., Macdonald, A.M., Ribchester, T.R., Davies, N.J., Venturi, P., Jones, L.A., Lewis, S.W., Sham, P.C., Gottesman, II, Farmer, A.E., McGuffin, P., Reveley, A.M., Murray, R.M., 1999. Heritability estimates for psychotic disorders: the Maudsley twin psychosis series. Arch Gen Psychiatry 56, 162-168.

Carlin, J.M., Borden, E.C., Sondel, P.M., Byrne, G.I., 1987. Biologic-response-modifier-induced indoleamine 2,3-dioxygenase activity in human peripheral blood mononuclear cell cultures. J Immunol 139, 2414-2418.

Carlsson, A., Lindqvist, M., 1963. Effect of Chlorpromazine or Haloperidol on Formation of 3-Methoxytyramine and Normetanephrine in Mouse Brain. Acta Pharmacol Tox 20, 140-&.

Carson, M.J., 2002. Microglia as liaisons between the immune and central nervous systems: Functional implications for multiple sclerosis. Glia 40, 218-231.

Chang, Y.F., Cauley, R.K., Chang, J.D., Rao, V.V., 1997. L-alpha-aminoadipate inhibits kynurenate synthesis in rat brain hippocampus and tissue culture. Neurochem Res 22, 825-829.

Chen, H.B., Li, F., Wu, S., An, S.C., 2013. [Hippocampus quinolinic acid modulates glutamate and NMDAR/mGluR1 in chronic unpredictable mild stress-induced depression]. Sheng Li Xue Bao 65, 577-585.

Chess, A.C., Bucci, D.J., 2006. Increased concentration of cerebral kynurenic acid alters stimulus processing and conditioned responding. Behav Brain Res 170, 326-332.

Chess, A.C., Landers, A.M., Bucci, D.J., 2009. L-kynurenine treatment alters contextual fear conditioning and context discrimination but not cue-specific fear conditioning. Behav Brain Res 201, 325-331.

Chess, A.C., Simoni, M.K., Alling, T.E., Bucci, D.J., 2007. Elevations of endogenous kynurenic acid produce spatial working memory deficits. Schizophr Bull 33, 797-804.

Chiarugi, A., Calvani, M., Meli, E., Traggiai, E., Moroni, F., 2001. Synthesis and release of neurotoxic kynurenine metabolites by human monocyte-derived macrophages. J Neuroimmunol 120, 190-198.

Connor, T.J., Starr, N., O'Sullivan, J.B., Harkin, A., 2008. Induction of indolamine 2,3-dioxygenase and kynurenine 3-monooxygenase in rat brain following a systemic inflammatory challenge: a role for IFN-gamma? Neurosci Lett 441, 29-34.

Creese, I., Burt, D.R., Snyder, S.H., 1976. Dopamine receptor binding predicts clinical and pharmacological potencies of antischizophrenic drugs. Science 192, 481-483.

Crump, C., Winkleby, M.A., Sundquist, K., Sundquist, J., 2013. Comorbidities and mortality in persons with schizophrenia: a Swedish national cohort study. Am J Psychiatry 170, 324-333.

Cuffy, M.C., Silverio, A.M., Qin, L., Wang, Y., Eid, R., Brandacher, G., Lakkis, F.G., Fuchs, D., Pober, J.S., Tellides, G., 2007. Induction of indoleamine 2,3-dioxygenase in vascular smooth muscle cells by interferon-gamma contributes to medial immunoprivilege. J Immunol 179, 5246-5254.

Cunningham, R.F., Israili, Z.H., Dayton, P.G., 1981. Clinical pharmacokinetics of probenecid. Clin Pharmacokinet 6, 135-151.

Czeh, B., Fuchs, E., Wiborg, O., Simon, M., 2016. Animal models of major depression and their clinical implications. Prog Neuro-Psychoph 64, 293-310.

Dalman, C., Allebeck, P., Gunnell, D., Harrison, G., Kristensson, K., Lewis, G., Lofving, S., Rasmussen, F., Wicks, S., Karlsson, H., 2008. Infections in the CNS during childhood and the risk of subsequent psychotic illness: a cohort study of more than one million Swedish subjects. Am J Psychiatry 165, 59-65.

Dantzer, R., O'Connor, J.C., Freund, G.G., Johnson, R.W., Kelley, K.W., 2008. From inflammation to sickness and depression: when the immune system subjugates the brain. Nature Reviews Neuroscience 9, 46-57.

Davies, G., Welham, J., Chant, D., Torrey, E.F., McGrath, J., 2003. A systematic review and meta-analysis of Northern Hemisphere season of birth studies in schizophrenia. Schizophr Bull 29, 587-593.

Davis, K.L., Kahn, R.S., Ko, G., Davidson, M., 1991. Dopamine in schizophrenia: a review and reconceptualization. Am J Psychiatry 148, 1474-1486.

Delgado, P.L., 2000. Depression: the case for a monoamine deficiency. J Clin Psychiatry 61 Suppl 6, 7-11.

DiNatale, B.C., Murray, I.A., Schroeder, J.C., Flaveny, C.A., Lahoti, T.S., Laurenzana, E.M., Omiecinski, C.J., Perdew, G.H., 2010. Kynurenic acid is a potent endogenous aryl hydrocarbon receptor ligand that synergistically induces interleukin-6 in the presence of inflammatory signaling. Toxicol Sci 115, 89-97.

Dowlati, Y., Herrmann, N., Swardfager, W., Liu, H., Sham, L., Reim, E.K., Lanctot, K.L., 2010. A meta-analysis of cytokines in major depression. Biol Psychiatry 67, 446-457.

Dutta, A., Mckie, S., Deakin, J.F.W., 2014. Resting state networks in major depressive disorder. Psychiat Res-Neuroim 224, 139-151.

Edwards, S.R., Mather, L.E., Lin, Y., Power, I., Cousins, M.J., 2000. Glutamate and kynurenate in the rat central nervous system following treatments with tail ischaemia or diclofenac. J Pharm Pharmacol 52, 59-66.

Erhardt, S., Blennow, K., Nordin, C., Skogh, E., Lindstrom, L.H., Engberg, G., 2001a. Kynurenic acid levels are elevated in the cerebrospinal fluid of patients with schizophrenia. Neurosci Lett 313, 96-98.

Erhardt, S., Engberg, G., 2002. Increased phasic activity of dopaminergic neurones in the rat ventral tegmental area following pharmacologically elevated levels of endogenous kynurenic acid. Acta Physiol Scand 175, 45-53.

Erhardt, S., Lim, C.K., Linderholm, K.R., Janelidze, S., Lindqvist, D., Samuelsson, M., Lundberg, K., Postolache, T.T., Traskman-Bendz, L., Guillemin, G.J., Brundin, L., 2013. Connecting inflammation with glutamate agonism in suicidality. Neuropsychopharmacology 38, 743-752.

Erhardt, S., Oberg, H., Mathe, J.M., Engberg, G., 2001b. Pharmacological elevation of endogenous kynurenic acid levels activates nigral dopamine neurons. Amino Acids 20, 353-362.

Erhardt, S., Schwieler, L., Emanuelsson, C., Geyer, M., 2004. Endogenous kynurenic acid disrupts prepulse inhibition. Biol Psychiatry 56, 255-260.

Evans, D.L., Charney, D.S., Lewis, L., Golden, R.N., Gorman, J.M., Krishnan, K.R., Nemeroff, C.B., Bremner, J.D., Carney, R.M., Coyne, J.C., Delong, M.R., Frasure-Smith, N., Glassman, A.H., Gold, P.W., Grant, I., Gwyther, L., Ironson, G., Johnson, R.L., Kanner, A.M., Katon, W.J., Kaufmann, P.G., Keefe, F.J., Ketter, T., Laughren, T.P., Leserman, J., Lyketsos, C.G., McDonald, W.M., McEwen, B.S., Miller, A.H., Musselman, D., O'Connor, C., Petitto, J.M., Pollock, B.G., Robinson, R.G., Roose, S.P., Rowland, J., Sheline, Y., Sheps, D.S., Simon, G., Spiegel, D., Stunkard, A., Sunderland, T., Tibbits, P., Jr., Valvo, W.J., 2005. Mood disorders in the medically ill: scientific review and recommendations. Biol Psychiatry 58, 175-189.

Falsig, J., van Beek, J., Hermann, C., Leist, M., 2008. Molecular basis for detection of invading pathogens in the brain. J Neurosci Res 86, 1434-1447.

Farina, C., Aloisi, F., Meinl, E., 2007. Astrocytes are active players in cerebral innate immunity. Trends Immunol 28, 138-145.

Fillman, S.G., Cloonan, N., Catts, V.S., Miller, L.C., Wong, J., McCrossin, T., Cairns, M., Weickert, C.S., 2013. Increased inflammatory markers identified in the dorsolateral prefrontal cortex of individuals with schizophrenia. Mol Psychiatry 18, 206-214.

Fischer, C.W., Eskelund, A., Budac, D.P., Tillmann, S., Liebenberg, N., Elfving, B., Wegener, G., 2015. Interferon-alpha treatment induces depression-like behaviour

accompanied by elevated hippocampal quinolinic acid levels in rats. Behav Brain Res 293, 166-172.

French, E.D., 1994. Phencyclidine and the midbrain dopamine system: electrophysiology and behavior. Neurotoxicol Teratol 16, 355-362.

French, E.D., Mura, A., Wang, T., 1993. MK-801, phencyclidine (PCP), and PCP-like drugs increase burst firing in rat A10 dopamine neurons: comparison to competitive NMDA antagonists. Synapse 13, 108-116.

Fu, X., Zunich, S.M., O'Connor, J.C., Kavelaars, A., Dantzer, R., Kelley, K.W., 2010. Central administration of lipopolysaccharide induces depressive-like behavior in vivo and activates brain indoleamine 2,3 dioxygenase in murine organotypic hippocampal slice cultures. J Neuroinflammation 7, 43.

Gal, E.M., Sherman, A.D., 1980. L-Kynurenine - Its Synthesis and Possible Regulatory Function in Brain. Neurochemical Research 5, 223-239.

Galea, I., Bechmann, I., Perry, V.H., 2007. What is immune privilege (not)? Trends Immunol 28, 12-18.

Giorgini, F., Huang, S.Y., Sathyasaikumar, K.V., Notarangelo, F.M., Thomas, M.A., Tararina, M., Wu, H.Q., Schwarcz, R., Muchowski, P.J., 2013. Targeted deletion of kynurenine 3-monooxygenase in mice: a new tool for studying kynurenine pathway metabolism in periphery and brain. J Biol Chem 288, 36554-36566.

Goldman-Rakic, P.S., Muly, E.C., 3rd, Williams, G.V., 2000. D(1) receptors in prefrontal cells and circuits. Brain Res Brain Res Rev 31, 295-301.

Grotta, J., Clark, W., Coull, B., Pettigrew, L.C., Mackay, B., Goldstein, L.B., Meissner, I., Murphy, D., LaRue, L., 1995. Safety and tolerability of the glutamate antagonist CGS 19755 (Selfotel) in patients with acute ischemic stroke. Results of a phase IIa randomized trial. Stroke 26, 602-605.

Guidetti, P., Amori, L., Sapko, M.T., Okuno, E., Schwarcz, R., 2007. Mitochondrial aspartate aminotransferase: a third kynurenate-producing enzyme in the mammalian brain. J Neurochem 102, 103-111.

Guidetti, P., Okuno, E., Schwarcz, R., 1997. Characterization of rat brain kynurenine aminotransferases I and II. J Neurosci Res 50, 457-465.

Guillemin, G.J., Kerr, S.J., Pemberton, L.A., Smith, D.G., Smythe, G.A., Armati, P.J., Brew, B.J., 2001a. IFN-beta1b induces kynurenine pathway metabolism in human macrophages: potential implications for multiple sclerosis treatment. J Interferon Cytokine Res 21, 1097-1101.

Guillemin, G.J., Kerr, S.J., Smythe, G.A., Smith, D.G., Kapoor, V., Armati, P.J., Croitoru, J., Brew, B.J., 2001b. Kynurenine pathway metabolism in human astrocytes: a paradox for neuronal protection. J Neurochem 78, 842-853.

Guillemin, G.J., Smith, D.G., Smythe, G.A., Armati, P.J., Brew, B.J., 2003. Expression of the kynurenine pathway enzymes in human microglia and macrophages. Developments in Tryptophan and Serotonin Metabolism 527, 105-112.

Han, Q., Cai, T., Tagle, D.A., Li, J., 2010a. Structure, expression, and function of kynurenine aminotransferases in human and rodent brains. Cell Mol Life Sci 67, 353-368.

Han, Q., Cai, T., Tagle, D.A., Li, J., 2010b. Thermal stability, pH dependence and inhibition of four murine kynurenine aminotransferases. BMC Biochem 11, 19.

Harms, L.R., Eyles, D.W., McGrath, J.J., Mackay-Sim, A., Burne, T.H., 2008. Developmental vitamin D deficiency alters adult behaviour in 129/SvJ and C57BL/6J mice. Behav Brain Res 187, 343-350.

Harrison, P.J., Owen, M.J., 2003. Genes for schizophrenia? Recent findings and their pathophysiological implications. Lancet 361, 417-419.

Heisler, L.K., Chu, H.M., Brennan, T.J., Danao, J.A., Bajwa, P., Parsons, L.H., Tecott, L.H., 1998. Elevated anxiety and antidepressant-like responses in serotonin 5-HT1A receptor mutant mice. Proc Natl Acad Sci U S A 95, 15049-15054.

Hilmas, C., Pereira, E.F., Alkondon, M., Rassoulpour, A., Schwarcz, R., Albuquerque, E.X., 2001. The brain metabolite kynurenic acid inhibits alpha7 nicotinic receptor activity and increases non-alpha7 nicotinic receptor expression: physiopathological implications. J Neurosci 21, 7463-7473.

Hoffman, H.S., Ison, J.R., 1980. Reflex modification in the domain of startle: I. Some empirical findings and their implications for how the nervous system processes sensory input. Psychol Rev 87, 175-189.

Holloway, T., Moreno, J.L., Umali, A., Rayannavar, V., Hodes, G.E., Russo, S.J., Gonzalez-Maeso, J., 2013. Prenatal stress induces schizophrenia-like alterations of serotonin 2A and metabotropic glutamate 2 receptors in the adult offspring: role of maternal immune system. J Neurosci 33, 1088-1098.

Holtze, M., Asp, L., Schwieler, L., Engberg, G., Karlsson, H., 2008. Induction of the kynurenine pathway by neurotropic influenza A virus infection. J Neurosci Res 86, 3674-3683.

Holtze, M., Saetre, P., Engberg, G., Schwieler, L., Werge, T., Andreassen, O.A., Hall, H., Terenius, L., Agartz, I., Jonsson, E.G., Schalling, M., Erhardt, S., 2012. Kynurenine 3-monooxygenase polymorphisms: relevance for kynurenic acid synthesis in patients with schizophrenia and healthy controls. J Psychiatry Neurosci 37, 53-57.

Huttunen, M.O., Niskanen, P., 1978. Prenatal loss of father and psychiatric disorders. Arch Gen Psychiatry 35, 429-431.

Iaccarino, H.F., Suckow, R.F., Xie, S., Bucci, D.J., 2013. The effect of transient increases in kynurenic acid and quinolinic acid levels early in life on behavior in adulthood: Implications for schizophrenia. Schizophr Res 150, 392-397.

Insel, B.J., Schaefer, C.A., McKeague, I.W., Susser, E.S., Brown, A.S., 2008. Maternal iron deficiency and the risk of schizophrenia in offspring. Arch Gen Psychiat 65, 1136-1144.

Itil, T., Keskiner, A., Kiremitci, N., Holden, J.M.C., 1967. Effect of Phencyclidine in Chronic Schizophrenics. Can Psychiat Assoc J 12, 209-212.

Jaskiw, G.E., Karoum, F., Freed, W.J., Phillips, I., Kleinman, J.E., Weinberger, D.R., 1990. Effect of ibotenic acid lesions of the medial prefrontal cortex on amphetamine-induced locomotion and regional brain catecholamine concentrations in the rat. Brain Res 534, 263-272.

Jauch, D.A., Sethy, V.H., Weick, B.G., Chase, T.N., Schwarcz, R., 1993. Intravenous administration of L-kynurenine to rhesus monkeys: effect on quinolinate and kynurenate levels in serum and cerebrospinal fluid. Neuropharmacology 32, 467-472.

Jentsch, J.D., Roth, R.H., 1999. The neuropsychopharmacology of phencyclidine: from NMDA receptor hypofunction to the dopamine hypothesis of schizophrenia. Neuropsychopharmacology 20, 201-225.

Jeong, Y.I., Kim, S.W., Jung, I.D., Lee, J.S., Chang, J.H., Lee, C.M., Chun, S.H., Yoon, M.S., Kim, G.T., Ryu, S.W., Kim, J.S., Shin, Y.K., Lee, W.S., Shin, H.K., Lee, J.D., Park, Y.M., 2009. Curcumin suppresses the induction of indoleamine 2,3-dioxygenase by blocking the Janus-activated kinase-protein kinase Cdelta-STAT1 signaling pathway in interferon-gamma-stimulated murine dendritic cells. J Biol Chem 284, 3700-3708.

Johnstone, T., van Reekum, C.M., Urry, H.L., Kalin, N.H., Davidson, R.J., 2007. Failure to regulate: counterproductive recruitment of top-down prefrontal-subcortical circuitry in major depression. J Neurosci 27, 8877-8884.

Kegeles, L.S., Abi-Dargham, A., Zea-Ponce, Y., Rodenhiser-Hill, J., Mann, J.J., Van Heertum, R.L., Cooper, T.B., Carlsson, A., Laruelle, M., 2000. Modulation of amphetamine-induced striatal dopamine release by ketamine in humans: implications for schizophrenia. Biol Psychiatry 48, 627-640.

Kessler, M., Terramani, T., Lynch, G., Baudry, M., 1989. A glycine site associated with N-methyl-D-aspartic acid receptors: characterization and identification of a new class of antagonists. J Neurochem 52, 1319-1328.

Khashan, A.S., Abel, K.M., McNamee, R., Pedersen, M.G., Webb, R.T., Baker, P.N., Kenny, L.C., Mortensen, P.B., 2008. Higher risk of offspring schizophrenia following antenatal maternal exposure to severe adverse life events. Arch Gen Psychiatry 65, 146-152.

Kim, J.S., Kornhuber, H.H., Schmid-Burgk, W., Holzmuller, B., 1980. Low cerebrospinal fluid glutamate in schizophrenic patients and a new hypothesis on schizophrenia. Neurosci Lett 20, 379-382.

King, D.J., 1998. Drug treatment of the negative symptoms of schizophrenia. Eur Neuropsychopharmacol 8, 33-42.

Kodsi, M.H., Swerdlow, N.R., 1997. Regulation of prepulse inhibition by ventral pallidal projections. Brain Res Bull 43, 219-228.

Koike, H., Ibi, D., Mizoguchi, H., Nagai, T., Nitta, A., Takuma, K., Nabeshima, T., Yoneda, Y., Yamada, K., 2009. Behavioral abnormality and pharmacologic response in social isolation-reared mice. Behav Brain Res 202, 114-121.

Kokkinidis, L., Anisman, H., 1980. Amphetamine models of paranoid schizophrenia: an overview and elaboration of animal experimentation. Psychol Bull 88, 551-579.

Koyuncu, O.O., Hogue, I.B., Enquist, L.W., 2013. Virus infections in the nervous system. Cell Host Microbe 13, 379-393.

Kozak, R., Campbell, B.M., Strick, C.A., Horner, W., Hoffmann, W.E., Kiss, T., Chapin, D.S., McGinnis, D., Abbott, A.L., Roberts, B.M., Fonseca, K., Guanowsky, V., Young, D.A., Seymour, P.A., Dounay, A., Hajos, M., Williams, G.V., Castner, S.A., 2014. Reduction of brain kynurenic acid improves cognitive function. J Neurosci 34, 10592-10602.

Kraepelin, E., 1896. Lehrbuch der psychiatrie. Barth, Leipzig.

Kristensen, J.D., Svensson, B., Gordh, T., Jr., 1992. The NMDA-receptor antagonist CPP abolishes neurogenic 'wind-up pain' after intrathecal administration in humans. Pain 51, 249-253.

Krystal, J.H., Karper, L.P., Seibyl, J.P., Freeman, G.K., Delaney, R., Bremner, J.D., Heninger, G.R., Bowers, M.B., Jr., Charney, D.S., 1994. Subanesthetic effects of the noncompetitive NMDA antagonist, ketamine, in humans. Psychotomimetic, perceptual, cognitive, and neuroendocrine responses. Arch Gen Psychiatry 51, 199-214.

Lahti, A.C., Weiler, M.A., Tamara Michaelidis, B.A., Parwani, A., Tamminga, C.A., 2001. Effects of ketamine in normal and schizophrenic volunteers. Neuropsychopharmacology 25, 455-467.

Lapin, I.P., 1973. Kynurenines as Probable Participants of Depression. Pharmakopsych Neuro 6, 273-279.

Larsson, M.K., Faka, A., Bhat, M., Imbeault, S., Goiny, M., Orhan, F., Oliveros, A., Stahl, S., Liu, X.C., Choi, D.S., Sandberg, K., Engberg, G., Schwieler, L., Erhardt, S., 2016. Repeated LPS Injection Induces Distinct Changes in the Kynurenine Pathway in Mice. Neurochem Res.

Laruelle, M., 1998. Imaging dopamine transmission in schizophrenia. A review and meta-analysis. Q J Nucl Med 42, 211-221.

Laruelle, M., Abi-Dargham, A., 1999. Dopamine as the wind of the psychotic fire: new evidence from brain imaging studies. J Psychopharmacol 13, 358-371.

Lavebratt, C., Olsson, S., Backlund, L., Frisen, L., Sellgren, C., Priebe, L., Nikamo, P., Traskman-Bendz, L., Cichon, S., Vawter, M.P., Osby, U., Engberg, G., Landen, M., Erhardt, S., Schalling, M., 2014. The KMO allele encoding Arg452 is associated with psychotic features in bipolar disorder type 1, and with increased CSF KYNA level and reduced KMO expression. Mol Psychiatry 19, 334-341.

Lawson, M.A., Parrott, J.M., McCusker, R.H., Dantzer, R., Kelley, K.W., O'Connor, J.C., 2013. Intracerebroventricular administration of lipopolysaccharide induces indoleamine-2,3-dioxygenase-dependent depression-like behaviors. J Neuroinflammation 10, 87.

Leklem, J.E., 1971. Quantitative aspects of tryptophan metabolism in humans and other species: a review. Am J Clin Nutr 24, 659-672.

Lewis, S.W., 1989. Congenital risk factors for schizophrenia. Psychol Med 19, 5-13.

Lichtenstein, P., Bjork, C., Hultman, C.M., Scolnick, E., Sklar, P., Sullivan, P.F., 2006. Recurrence risks for schizophrenia in a Swedish national cohort. Psychol Med 36, 1417-1425.

Liebig, J., 1853. Uber Kynurensäure. Justus Liebigs Ann. Chem. 86, 125-126.

Linderholm, K.R., Alm, M.T., Larsson, M.K., Olsson, S.K., Goiny, M., Hajos, M., Erhardt, S., Engberg, G., 2016. Inhibition of kynurenine aminotransferase II reduces activity of midbrain dopamine neurons. Neuropharmacology 102, 42-47.

Linderholm, K.R., Andersson, A., Olsson, S., Olsson, E., Snodgrass, R., Engberg, G., Erhardt, S., 2007. Activation of rat ventral tegmental area dopamine neurons by endogenous kynurenic acid: a pharmacological analysis. Neuropharmacology 53, 918-924.

Linderholm, K.R., Skogh, E., Olsson, S.K., Dahl, M.L., Holtze, M., Engberg, G., Samuelsson, M., Erhardt, S., 2012. Increased levels of kynurenine and kynurenic acid in the CSF of patients with schizophrenia. Schizophr Bull 38, 426-432.

Lindqvist, D., Janelidze, S., Hagell, P., Erhardt, S., Samuelsson, M., Minthon, L., Hansson, O., Bjorkqvist, M., Traskman-Bendz, L., Brundin, L., 2009. Interleukin-6 is elevated in the cerebrospinal fluid of suicide attempters and related to symptom severity. Biol Psychiatry 66, 287-292.

Lipina, T.V., Roder, J.C., 2014. Disrupted-In-Schizophrenia-1 (DISC1) interactome and mental disorders: impact of mouse models. Neurosci Biobehav Rev 45, 271-294.

Lister, R.G., 1987. The Use of a Plus-Maze to Measure Anxiety in the Mouse. Psychopharmacology 92, 180-185.

Liu, X.C., Holtze, M., Powell, S.B., Terrando, N., Larsson, M.K., Persson, A., Olsson, S.K., Orhan, F., Kegel, M., Asp, L., Goiny, M., Schwieler, L., Engberg, G., Karlsson, H., Erhardt, S., 2014a. Behavioral disturbances in adult mice following neonatal virus infection or kynurenine treatment--role of brain kynurenic acid. Brain Behav Immun 36, 80-89.

Liu, X.M., Sun, G.X., Zhang, X.Q., Xu, B., Shen, C.Y., Shi, L.J., Ma, X.Y., Ren, X.J., Feng, K., Liu, P.Z., 2014b. Relationship between the prefrontal function and the severity of the emotional symptoms during a verbal fluency task in patients with major depressive disorder: A multi-channel NIRS study. Prog Neuro-Psychoph 54, 114-121.

Lodge, D., Anis, N.A., 1982. Effects of phencyclidine on excitatory amino acid activation of spinal interneurones in the cat. Eur J Pharmacol 77, 203-204.

Louveau, A., Smirnov, I., Keyes, T.J., Eccles, J.D., Rouhani, S.J., Peske, J.D., Derecki, N.C., Castle, D., Mandell, J.W., Lee, K.S., Harris, T.H., Kipnis, J., 2015. Structural and functional features of central nervous system lymphatic vessels. Nature 523, 337-341.

Luby, E.D., Cohen, B.D., Rosenbaum, G., Gottlieb, J.S., Kelley, R., 1959. Study of a New Schizophrenomimetic Drug - Sernyl. Arch Neuro Psychiatr 81, 363-369.

Lyon, M., Barr, C.E., Cannon, T.D., Mednick, S.A., Shore, D., 1989. Fetal neural development and schizophrenia. Schizophr Bull 15, 149-161.

Macedo, D.S., Araujo, D.P., Sampaio, L.R.L., Vasconcelos, S.M.M., Sales, P.M.G., Sousa, F.C.F., Hallak, J.E., Crippa, J.A., Carvalho, A.F., 2012. Animal models of prenatal immune challenge and their contribution to the study of schizophrenia: a systematic review. Braz J Med Biol Res 45, 179-186.

Mailankot, M., Nagaraj, R.H., 2010. Induction of indoleamine 2,3-dioxygenase by interferon-gamma in human lens epithelial cells: apoptosis through the formation of 3-hydroxykynurenine. Int J Biochem Cell Biol 42, 1446-1454.

Malhotra, A.K., Pinals, D.A., Adler, C.M., Elman, I., Clifton, A., Pickar, D., Breier, A., 1997. Ketamine-induced exacerbation of psychotic symptoms and cognitive impairment in neuroleptic-free schizophrenics. Neuropsychopharmacology 17, 141-150.

Marcelis, M., Takei, N., van Os, J., 1999. Urbanization and risk for schizophrenia: does the effect operate before or around the time of illness onset? Psychol Med 29, 1197-1203.

Marcotte, E.R., Pearson, D.M., Srivastava, L.K., 2001. Animal models of schizophrenia: a critical review. J Psychiatry Neurosci 26, 395-410.

Mattiello, R., D'Ambrosio, E., Wilda, M., Saye, M., Miranda, M.R., Maure, P., Pereira, C.A., Digirolamo, F.A., 2015. A Preventive Cytokine Treatment of the Viral Infectious Bursal Disease (IBD) of Chickens. J Poult Sci 52, 145-150.

McCusker, R.H., Kelley, K.W., 2013. Immune-neural connections: how the immune system's response to infectious agents influences behavior. Journal of Experimental Biology 216, 84-98.

McGrath, J., 1999. Hypothesis: Is low prenatal vitamin D a risk-modifying factor for schizophrenia? Schizophrenia Research 40, 173-177.

McGrath, J., Saha, S., Chant, D., Welham, J., 2008. Schizophrenia: a concise overview of incidence, prevalence, and mortality. Epidemiol Rev 30, 67-76.

Meltzer, H.Y., 2002. Suicidality in schizophrenia: a review of the evidence for risk factors and treatment options. Curr Psychiatry Rep 4, 279-283.

Meyer, U., 2013. Developmental neuroinflammation and schizophrenia. Prog Neuropsychopharmacol Biol Psychiatry 42, 20-34.

Miller, A.H., Maletic, V., Raison, C.L., 2009. Inflammation and its discontents: the role of cytokines in the pathophysiology of major depression. Biol Psychiatry 65, 732-741.

Miller, C.L., Llenos, I.C., Dulay, J.R., Barillo, M.M., Yolken, R.H., Weis, S., 2004. Expression of the kynurenine pathway enzyme tryptophan 2,3-dioxygenase is increased in the frontal cortex of individuals with schizophrenia. Neurobiology of Disease 15, 618-629.

Miller, C.L., Llenos, I.C., Dulay, J.R., Weis, S., 2006. Upregulation of the initiating step of the kynurenine pathway in postmortem anterior cingulate cortex from individuals with schizophrenia and bipolar disorder. Brain Res 1073-1074, 25-37.

Mittleman, G., LeDuc, P.A., Whishaw, I.Q., 1993. The role of D1 and D2 receptors in the heightened locomotion induced by direct and indirect dopamine agonists in rats with hippocampal damage: an animal analogue of schizophrenia. Behav Brain Res 55, 253-267.

Monte, A.S., de Souza, G.C., McIntyre, R.S., Soczynska, J.K., dos Santos, J.V., Cordeiro, R.C., Ribeiro, B.M., de Lucena, D.F., Vasconcelos, S.M., de Sousa, F.C., Carvalho, A.F., Macedo, D.S., 2013. Prevention and reversal of ketamine-induced schizophrenia related behavior by minocycline in mice: Possible involvement of antioxidant and nitrergic pathways. J Psychopharmacol 27, 1032-1043.

Montkowski, A., Barden, N., Wotjak, C., Stec, I., Ganster, J., Meaney, M., Engelmann, M., Reul, J.M., Landgraf, R., Holsboer, F., 1995. Long-term antidepressant treatment reduces behavioural deficits in transgenic mice with impaired glucocorticoid receptor function. J Neuroendocrinol 7, 841-845.

Morgan, C., Fisher, H., 2007. Environment and schizophrenia: environmental factors in schizophrenia: childhood trauma--a critical review. Schizophr Bull 33, 3-10.

Moroni, F., 1999. Tryptophan metabolism and brain function: focus on kynurenine and other indole metabolites. Eur J Pharmacol 375, 87-100.

Moroni, F., Russi, P., Lombardi, G., Beni, M., Carla, V., 1988. Presence of kynurenic acid in the mammalian brain. J Neurochem 51, 177-180.

Mortensen, P.B., Pedersen, C.B., Westergaard, T., Wohlfahrt, J., Ewald, H., Mors, O., Andersen, P.K., Melbye, M., 1999. Effects of family history and place and season of birth on the risk of schizophrenia. N Engl J Med 340, 603-608.

Muller, M.B., Zimmermann, S., Sillaber, I., Hagemeyer, T.P., Deussing, J.M., Timpl, P., Kormann, M.S., Droste, S.K., Kuhn, R., Reul, J.M., Holsboer, F., Wurst, W., 2003. Limbic corticotropin-releasing hormone receptor 1 mediates anxiety-related behavior and hormonal adaptation to stress. Nat Neurosci 6, 1100-1107.

Muller, N., Schwarz, M.J., 2007. The immune-mediated alteration of serotonin and glutamate: towards an integrated view of depression. Mol Psychiatry 12, 988-1000.

Murray, C.J., Lopez, A.D., 1996. Evidence-based health policy--lessons from the Global Burden of Disease Study. Science 274, 740-743.

Murray, C.J., Lopez, A.D., 1997. Alternative projections of mortality and disability by cause 1990-2020: Global Burden of Disease Study. Lancet 349, 1498-1504.

Murray, R.M., Lewis, S.W., 1988. Is schizophrenia a neurodevelopmental disorder? Br Med J (Clin Res Ed) 296, 63.

Myint, A.M., Kim, Y.K., Verkerk, R., Scharpe, S., Steinbusch, H., Leonard, B., 2007. Kynurenine pathway in major depression: evidence of impaired neuroprotection. J Affect Disord 98, 143-151.

Nemeth, H., Toldi, J., Vecsei, L., 2005. Role of kynurenines in the central and peripheral nervous systems. Curr Neurovasc Res 2, 249-260.

Nilsson, L.K., Linderholm, K.R., Engberg, G., Paulson, L., Blennow, K., Lindstrom, L.H., Nordin, C., Karanti, A., Persson, P., Erhardt, S., 2005. Elevated levels of kynurenic acid in the cerebrospinal fluid of male patients with schizophrenia. Schizophr Res 80, 315-322.

Nilsson, L.K., Linderholm, K.R., Erhardt, S., 2006. Subchronic treatment with kynurenine and probenecid: effects on prepulse inhibition and firing of midbrain dopamine neurons. J Neural Transm (Vienna) 113, 557-571.

Nuechterlein, K.H., Barch, D.M., Gold, J.M., Goldberg, T.E., Green, M.F., Heaton, R.K., 2004. Identification of separable cognitive factors in schizophrenia. Schizophr Res 72, 29-39.

O'Connor, J.C., Andre, C., Wang, Y., Lawson, M.A., Szegedi, S.S., Lestage, J., Castanon, N., Kelley, K.W., Dantzer, R., 2009a. Interferon-gamma and tumor necrosis factor-alpha mediate the upregulation of indoleamine 2,3-dioxygenase and the induction of depressive-like behavior in mice in response to bacillus Calmette-Guerin. J Neurosci 29, 4200-4209.

O'Connor, J.C., Lawson, M.A., Andre, C., Briley, E.M., Szegedi, S.S., Lestage, J., Castanon, N., Herkenham, M., Dantzer, R., Kelley, K.W., 2009b. Induction of IDO by Bacille Calmette-Guerin Is Responsible for Development of Murine Depressive-Like Behavior. Journal of Immunology 182, 3202-3212.

O'Connor, J.C., Lawson, M.A., Andre, C., Moreau, M., Lestage, J., Castanon, N., Kelley, K.W., Dantzer, R., 2009c. Lipopolysaccharide-induced depressive-like behavior is mediated by indoleamine 2,3-dioxygenase activation in mice. Mol Psychiatr 14, 511-522.

Okuno, E., Nakamura, M., Schwarcz, R., 1991. Two kynurenine aminotransferases in human brain. Brain Res 542, 307-312.

Olson, J.K., Miller, S.D., 2004. Microglia initiate central nervous system innate and adaptive immune responses through multiple TLRs. J Immunol 173, 3916-3924.

Olsson, S.K., Andersson, A.S., Linderholm, K.R., Holtze, M., Nilsson-Todd, L.K., Schwieler, L., Olsson, E., Larsson, K., Engberg, G., Erhardt, S., 2009. Elevated levels of kynurenic acid change the dopaminergic response to amphetamine: implications for schizophrenia. Int J Neuropsychopharmacol 12, 501-512.

Olsson, S.K., Larsson, M.K., Erhardt, S., 2012a. Subchronic elevation of brain kynurenic acid augments amphetamine-induced locomotor response in mice. J Neural Transm (Vienna) 119, 155-163.

Olsson, S.K., Samuelsson, M., Saetre, P., Lindstrom, L., Jonsson, E.G., Nordin, C., Engberg, G., Erhardt, S., Landen, M., 2010. Elevated levels of kynurenic acid in the cerebrospinal fluid of patients with bipolar disorder. J Psychiatry Neurosci 35, 195-199.

Olsson, S.K., Sellgren, C., Engberg, G., Landen, M., Erhardt, S., 2012b. Cerebrospinal fluid kynurenic acid is associated with manic and psychotic features in patients with bipolar I disorder. Bipolar Disord 14, 719-726.

Overstreet, D.H., Friedman, E., Mathe, A.A., Yadid, G., 2005. The Flinders Sensitive Line rat: a selectively bred putative animal model of depression. Neurosci Biobehav Rev 29, 739-759.

Overstreet, D.H., Wegener, G., 2013. The flinders sensitive line rat model of depression--25 years and still producing. Pharmacol Rev 65, 143-155.

Palmer, B.A., Pankratz, V.S., Bostwick, J.M., 2005. The lifetime risk of suicide in schizophrenia: a reexamination. Arch Gen Psychiatry 62, 247-253.

Parrott, J.M., Redus, L., O'Connor, J.C., 2016. Kynurenine metabolic balance is disrupted in the hippocampus following peripheral lipopolysaccharide challenge. J Neuroinflamm 13.

Parsons, C.G., Danysz, W., Quack, G., Hartmann, S., Lorenz, B., Wollenburg, C., Baran, L., Przegalinski, E., Kostowski, W., Krzascik, P., Chizh, B., Headley, P.M., 1997. Novel systemically active antagonists of the glycine site of the N-methyl-D-aspartate receptor: electrophysiological, biochemical and behavioral characterization. J Pharmacol Exp Ther 283, 1264-1275.

Paus, T., 2005. Mapping brain maturation and cognitive development during adolescence. Trends Cogn Sci 9, 60-68.

Paus, T., Keshavan, M., Giedd, J.N., 2008. Why do many psychiatric disorders emerge during adolescence? Nat Rev Neurosci 9, 947-957.

Pemberton, L.A., Kerr, S.J., Smythe, G., Brew, B.J., 1997. Quinolinic acid production by macrophages stimulated with IFN-gamma, TNF-alpha, and IFN-alpha. J Interferon Cytokine Res 17, 589-595.

Perälä, J., Suvisaari, J., Saarni, S.I., Kuoppasalmi, K., Isometsa, E., Pirkola, S., Partonen, T., Tuulio-Henriksson, A., Hintikka, J., Kieseppa, T., Harkanen, T., Koskinen, S., Lonnqvist, J., 2007. Lifetime prevalence of psychotic and bipolar I disorders in a general population. Arch Gen Psychiatry 64, 19-28.

Pershing, M.L., Bortz, D.M., Pocivavsek, A., Fredericks, P.J., Jorgensen, C.V., Vunck, S.A., Leuner, B., Schwarcz, R., Bruno, J.P., 2015. Elevated levels of kynurenic acid during gestation produce neurochemical, morphological, and cognitive deficits in adulthood: implications for schizophrenia. Neuropharmacology 90, 33-41.

Pocivavsek, A., Wu, H.Q., Elmer, G.I., Bruno, J.P., Schwarcz, R., 2012. Pre- and postnatal exposure to kynurenine causes cognitive deficits in adulthood. Eur J Neurosci 35, 1605-1612.

Pocivavsek, A., Wu, H.Q., Potter, M.C., Elmer, G.I., Pellicciari, R., Schwarcz, R., 2011. Fluctuations in endogenous kynurenic acid control hippocampal glutamate and memory. Neuropsychopharmacology 36, 2357-2367.

Potter, M.C., Elmer, G.I., Bergeron, R., Albuquerque, E.X., Guidetti, P., Wu, H.Q., Schwarcz, R., 2010. Reduction of endogenous kynurenic acid formation enhances

extracellular glutamate, hippocampal plasticity, and cognitive behavior. Neuropsychopharmacology 35, 1734-1742.

Raison, C.L., Dantzer, R., Kelley, K.W., Lawson, M.A., Woolwine, B.J., Vogt, G., Spivey, J.R., Saito, K., Miller, A.H., 2010. CSF concentrations of brain tryptophan and kynurenines during immune stimulation with IFN-alpha: relationship to CNS immune responses and depression. Mol Psychiatry 15, 393-403.

Ramboz, S., Oosting, R., Amara, D.A., Kung, H.F., Blier, P., Mendelsohn, M., Mann, J.J., Brunner, D., Hen, R., 1998. Serotonin receptor 1A knockout: an animal model of anxiety-related disorder. Proc Natl Acad Sci U S A 95, 14476-14481.

Rao, J.S., Kim, H.W., Harry, G.J., Rapoport, S.I., Reese, E.A., 2013. Increased neuroinflammatory and arachidonic acid cascade markers, and reduced synaptic proteins, in the postmortem frontal cortex from schizophrenia patients. Schizophr Res 147, 24-31.

Rapoport, J.L., Addington, A.M., Frangou, S., Psych, M.R., 2005. The neurodevelopmental model of schizophrenia: update 2005. Mol Psychiatry 10, 434-449.

Reichenberg, A., Yirmiya, R., Schuld, A., Kraus, T., Haack, M., Morag, A., Pollmacher, T., 2001. Cytokine-associated emotional and cognitive disturbances in humans. Arch Gen Psychiatry 58, 445-452.

Reiss, C.S., Chesler, D.A., Hodges, J., Ireland, D.D., Chen, N., 2002. Innate immune responses in viral encephalitis. Curr Top Microbiol Immunol 265, 63-94.

Resta, F., Masi, A., Sili, M., Laurino, A., Moroni, F., Mannaioni, G., 2016. Kynurenic acid and zaprinast induce analgesia by modulating HCN channels through GPR35 activation. Neuropharmacology 108, 136-143.

Rosso, P.R., 1990. Prenatal Nutrition and Brain Growth. Neurol Neur 58, 25-40.

Rover, S., Cesura, A.M., Huguenin, P., Kettler, R., Szente, A., 1997. Synthesis and biochemical evaluation of N-(4-phenylthiazol-2-yl)benzenesulfonamides as high-affinity inhibitors of kynurenine 3-hydroxylase. J Med Chem 40, 4378-4385.

Russi, P., Alesiani, M., Lombardi, G., Davolio, P., Pellicciari, R., Moroni, F., 1992. Nicotinylalanine increases the formation of kynurenic acid in the brain and antagonizes convulsions. J Neurochem 59, 2076-2080.

Saha, S., Chant, D., Welham, J., McGrath, J., 2005. A systematic review of the prevalence of schizophrenia. PLoS Med 2, e141.

Salazar, A., Gonzalez-Rivera, B.L., Redus, L., Parrott, J.M., O'Connor, J.C., 2012. Indoleamine 2,3-dioxygenase mediates anhedonia and anxiety-like behaviors caused by peripheral lipopolysaccharide immune challenge. Horm Behav 62, 202-209.

Sasayama, D., Hattori, K., Wakabayashi, C., Teraishi, T., Hori, H., Ota, M., Yoshida, S., Arima, K., Higuchi, T., Amano, N., Kunugi, H., 2013. Increased cerebrospinal fluid interleukin-6 levels in patients with schizophrenia and those with major depressive disorder. J Psychiatr Res 47, 401-406.

Sathyasaikumar, K.V., Stachowski, E.K., Wonodi, I., Roberts, R.C., Rassoulpour, A., McMahon, R.P., Schwarcz, R., 2011. Impaired kynurenine pathway metabolism in the prefrontal cortex of individuals with schizophrenia. Schizophr Bull 37, 1147-1156.

Savitz, J., Drevets, W.C., Smith, C.M., Victor, T.A., Wurfel, B.E., Bellgowan, P.S., Bodurka, J., Teague, T.K., Dantzer, R., 2015a. Putative neuroprotective and neurotoxic kynurenine pathway metabolites are associated with hippocampal and amygdalar volumes in subjects with major depressive disorder. Neuropsychopharmacology 40, 463-471.

Savitz, J., Drevets, W.C., Wurfel, B.E., Ford, B.N., Bellgowan, P.S., Victor, T.A., Bodurka, J., Teague, T.K., Dantzer, R., 2015b. Reduction of kynurenic acid to quinolinic acid ratio in both the depressed and remitted phases of major depressive disorder. Brain Behav Immun 46, 55-59.

Schmidt, W., Guidetti, P., Okuno, E., Schwarcz, R., 1993. Characterization of human brain kynurenine aminotransferases using [3H]kynurenine as a substrate. Neuroscience 55, 177-184.

Schwarcz, R., Bruno, J.P., Muchowski, P.J., Wu, H.Q., 2012. Kynurenines in the mammalian brain: when physiology meets pathology. Nat Rev Neurosci 13, 465-477.

Schwarcz, R., Rassoulpour, A., Wu, H.Q., Medoff, D., Tamminga, C.A., Roberts, R.C., 2001. Increased cortical kynurenate content in schizophrenia. Biol Psychiatry 50, 521-530.

Schwieler, L., Engberg, G., Erhardt, S., 2004. Clozapine modulates midbrain dopamine neuron firing via interaction with the NMDA receptor complex. Synapse 52, 114-122.

Schwieler, L., Erhardt, S., Erhardt, C., Engberg, G., 2005. Prostaglandin-mediated control of rat brain kynurenic acid synthesis--opposite actions by COX-1 and COX-2 isoforms. J Neural Transm (Vienna) 112, 863-872.

Schwieler, L., Erhardt, S., Nilsson, L., Linderholm, K., Engberg, G., 2006. Effects of COX-1 and COX-2 inhibitors on the firing of rat midbrain dopaminergic neurons--possible involvement of endogenous kynurenic acid. Synapse 59, 290-298.

Schwieler, L., Larsson, M.K., Skogh, E., Kegel, M.E., Orhan, F., Abdelmoaty, S., Finn, A., Bhat, M., Samuelsson, M., Lundberg, K., Dahl, M.L., Sellgren, C., Schuppe-Koistinen, I., Svensson, C., Erhardt, S., Engberg, G., 2015. Increased levels of IL-6 in the cerebrospinal fluid of patients with chronic schizophrenia--significance for activation of the kynurenine pathway. J Psychiatry Neurosci 40, 126-133.

Schwieler, L., Linderholm, K.R., Nilsson-Todd, L.K., Erhardt, S., Engberg, G., 2008. Clozapine interacts with the glycine site of the NMDA receptor: electrophysiological studies of dopamine neurons in the rat ventral tegmental area. Life Sci 83, 170-175.

Schwieler, L., Samuelsson, M., Frye, M.A., Bhat, M., Schuppe-Koistinen, I., Jungholm, O., Johansson, A.G., Landen, M., Sellgren, C.M., Erhardt, S., 2016. Electroconvulsive

therapy suppresses the neurotoxic branch of the kynurenine pathway in treatment-resistant depressed patients. J Neuroinflammation 13, 51.

Seeman, P., Lee, T., 1975. Antipsychotic drugs: direct correlation between clinical potency and presynaptic action on dopamine neurons. Science 188, 1217-1219.

Seeman, P., Lee, T., Chau-Wong, M., Wong, K., 1976. Antipsychotic drug doses and neuroleptic/dopamine receptors. Nature 261, 717-719.

Sellgren, C.M., Kegel, M.E., Bergen, S.E., Ekman, C.J., Olsson, S., Larsson, M., Vawter, M.P., Backlund, L., Sullivan, P.F., Sklar, P., Smoller, J.W., Magnusson, P.K., Hultman, C.M., Walther-Jallow, L., Svensson, C.I., Lichtenstein, P., Schalling, M., Engberg, G., Erhardt, S., Landen, M., 2015. A genome-wide association study of kynurenic acid in cerebrospinal fluid: implications for psychosis and cognitive impairment in bipolar disorder. Mol Psychiatry.

Sharp, T., Zetterstrom, T., Ljungberg, T., Ungerstedt, U., 1987. A Direct Comparison of Amphetamine-Induced Behaviors and Regional Brain Dopamine Release in the Rat Using Intracerebral Dialysis. Brain Research 401, 322-330.

Si, X., Miguel-Hidalgo, J.J., O'Dwyer, G., Stockmeier, C.A., Rajkowska, G., 2004. Age-dependent reductions in the level of glial fibrillary acidic protein in the prefrontal cortex in major depression. Neuropsychopharmacology 29, 2088-2096.

Snyder, S.H., 1973. Amphetamine psychosis: a "model" schizophrenia mediated by catecholamines. Am J Psychiatry 130, 61-67.

Soderlund, J., Olsson, S.K., Samuelsson, M., Walther-Jallow, L., Johansson, C., Erhardt, S., Landen, M., Engberg, G., 2011. Elevation of cerebrospinal fluid interleukin-1ss in bipolar disorder. J Psychiatry Neurosci 36, 114-118.

Soderlund, J., Schroder, J., Nordin, C., Samuelsson, M., Walther-Jallow, L., Karlsson, H., Erhardt, S., Engberg, G., 2009. Activation of brain interleukin-1beta in schizophrenia. Mol Psychiatry 14, 1069-1071.

Solomon, D.A., Keller, M.B., Leon, A.C., Mueller, T.I., Lavori, P.W., Shea, M.T., Coryell, W., Warshaw, M., Turvey, C., Maser, J.D., Endicott, J., 2000. Multiple recurrences of major depressive disorder. Am J Psychiatry 157, 229-233.

Speciale, C., Schwarcz, R., 1990. Uptake of kynurenine into rat brain slices. J Neurochem 54, 156-163.

Speciale, C., Wu, H.Q., Cini, M., Marconi, M., Varasi, M., Schwarcz, R., 1996. (R,S)-3,4-dichlorobenzoylalanine (FCE 28833A) causes a large and persistent increase in brain kynurenic acid levels in rats. Eur J Pharmacol 315, 263-267.

Steiner, J., Walter, M., Gos, T., Guillemin, G.J., Bernstein, H.G., Sarnyai, Z., Mawrin, C., Brisch, R., Bielau, H., Schwabedissen, L.M.Z., Bogerts, B., Myint, A.M., 2011. Severe depression is associated with increased microglial quinolinic acid in subregions of the anterior cingulate gyrus: Evidence for an immune-modulated glutamatergic neurotransmission? J Neuroinflamm 8.

Stone, T.W., 1993. Neuropharmacology of quinolinic and kynurenic acids. Pharmacol Rev 45, 309-379.

Stone, T.W., 2007. Kynurenic acid blocks nicotinic synaptic transmission to hippocampal interneurons in young rats. Eur J Neurosci 25, 2656-2665.

Stone, T.W., Perkins, M.N., 1981. Quinolinic acid: a potent endogenous excitant at amino acid receptors in CNS. Eur J Pharmacol 72, 411-412.

Sullivan, P.F., Kendler, K.S., Neale, M.C., 2003. Schizophrenia as a complex trait: evidence from a meta-analysis of twin studies. Arch Gen Psychiatry 60, 1187-1192.

Svensson, T.H., 2000. Dysfunctional brain dopamine systems induced by psychotomimetic NMDA-receptor antagonists and the effects of antipsychotic drugs. Brain Res Brain Res Rev 31, 320-329.

Swartz, K.J., During, M.J., Freese, A., Beal, M.F., 1990. Cerebral synthesis and release of kynurenic acid: an endogenous antagonist of excitatory amino acid receptors. J Neurosci 10, 2965-2973.

Swerdlow, N.R., Braff, D.L., Geyer, M.A., 2000. Animal models of deficient sensorimotor gating: what we know, what we think we know, and what we hope to know soon. Behav Pharmacol 11, 185-204.

Swerdlow, N.R., Braff, D.L., Taaid, N., Geyer, M.A., 1994. Assessing the validity of an animal model of deficient sensorimotor gating in schizophrenic patients. Arch Gen Psychiatry 51, 139-154.

Swerdlow, N.R., Lipska, B.K., Weinberger, D.R., Braff, D.L., Jaskiw, G.E., Geyer, M.A., 1995. Increased sensitivity to the sensorimotor gating-disruptive effects of apomorphine after lesions of medial prefrontal cortex or ventral hippocampus in adult rats. Psychopharmacology (Berl) 122, 27-34.

Swerdlow, N.R., Weber, M., Qu, Y., Light, G.A., Braff, D.L., 2008. Realistic expectations of prepulse inhibition in translational models for schizophrenia research. Psychopharmacology (Berl) 199, 331-388.

Takikawa, O., 2005. Biochemical and medical aspects of the indoleamine 2,3-dioxygenase-initiated L-tryptophan metabolism. Biochem Biophys Res Commun 338, 12-19.

Timofeeva, O.A., Levin, E.D., 2011. Glutamate and nicotinic receptor interactions in working memory: importance for the cognitive impairment of schizophrenia. Neuroscience 195, 21-36.

Torrey, E.F., Miller, J., Rawlings, R., Yolken, R.H., 1997. Seasonality of births in schizophrenia and bipolar disorder: a review of the literature. Schizophr Res 28, 1-38.

Trakhtenberg, E.F., Goldberg, J.L., 2011. Immunology. Neuroimmune communication. Science 334, 47-48.

Trecartin, K.V., Bucci, D.J., 2011. Administration of kynurenine during adolescence, but not during adulthood, impairs social behavior in rats. Schizophr Res 133, 156-158.

Tsay, C.J., 2013. Julius Wagner-Jauregg and the legacy of malarial therapy for the treatment of general paresis of the insane. Yale J Biol Med 86, 245-254.

Tyring, S., Gottlieb, A., Papp, K., Gordon, K., Leonardi, C., Wang, A., Lalla, D., Woolley, M., Jahreis, A., Zitnik, R., Cella, D., Krishnan, R., 2006. Etanercept and clinical outcomes, fatigue, and depression in psoriasis: double-blind placebo-controlled randomised phase III trial. Lancet 367, 29-35.

Urata, Y., Koga, K., Hirota, Y., Akiyama, I., Izumi, G., Takamura, M., Nagai, M., Harada, M., Hirata, T., Yoshino, O., Kawana, K., Fujii, T., Osuga, Y., 2014. IL-1beta increases expression of tryptophan 2,3-dioxygenase and stimulates tryptophan catabolism in endometrioma stromal cells. Am J Reprod Immunol 72, 496-503.

Uwai, Y., Honjo, H., Iwamoto, K., 2012. Interaction and transport of kynurenic acid via human organic anion transporters hOAT1 and hOAT3. Pharmacol Res 65, 254-260.

van Berckel, B.N., Bossong, M.G., Boellaard, R., Kloet, R., Schuitemaker, A., Caspers, E., Luurtsema, G., Windhorst, A.D., Cahn, W., Lammertsma, A.A., Kahn, R.S., 2008. Microglia activation in recent-onset schizophrenia: a quantitative (R)-[11C]PK11195 positron emission tomography study. Biol Psychiatry 64, 820-822.

van Os, J., Selten, J.P., 1998. Prenatal exposure to maternal stress and subsequent schizophrenia. The May 1940 invasion of The Netherlands. Br J Psychiatry 172, 324-326.

Vecsei, L., Beal, M.F., 1990. Influence of kynurenine treatment on open-field activity, elevated plus-maze, avoidance behaviors and seizures in rats. Pharmacol Biochem Behav 37, 71-76.

Vollenweider, F.X., Vontobel, P., Oye, I., Hell, D., Leenders, K.L., 2000. Effects of (S)-ketamine on striatal dopamine: a [11C]raclopride PET study of a model psychosis in humans. J Psychiatr Res 34, 35-43.

Weinberger, D.R., 1987. Implications of normal brain development for the pathogenesis of schizophrenia. Arch Gen Psychiatry 44, 660-669.

Welham, J., Isohanni, M., Jones, P., McGrath, J., 2009. The antecedents of schizophrenia: a review of birth cohort studies. Schizophr Bull 35, 603-623.

Wilson, E.H., Weninger, W., Hunter, C.A., 2010. Trafficking of immune cells in the central nervous system. J Clin Invest 120, 1368-1379.

Wonodi, I., Stine, O.C., Sathyasaikumar, K.V., Roberts, R.C., Mitchell, B.D., Hong, L.E., Kajii, Y., Thaker, G.K., Schwarcz, R., 2011. Downregulated kynurenine 3-monooxygenase gene expression and enzyme activity in schizophrenia and genetic association with schizophrenia endophenotypes. Arch Gen Psychiatry 68, 665-674.

Wu, H.Q., Ungerstedt, U., Schwarcz, R., 1992. Regulation of kynurenic acid synthesis studied by microdialysis in the dorsal hippocampus of unanesthetized rats. Eur J Pharmacol 213, 375-380.

Xu, F., Gainetdinov, R.R., Wetsel, W.C., Jones, S.R., Bohn, L.M., Miller, G.W., Wang, Y.M., Caron, M.G., 2000. Mice lacking the norepinephrine transporter are supersensitive to psychostimulants. Nat Neurosci 3, 465-471.

Yenari, M.A., Bell, T.E., Kotake, A.N., Powell, M., Steinberg, G.K., 1998. Dose escalation safety and tolerance study of the competitive NMDA antagonist selfotel (CGS 19755) in neurosurgery patients. Clin Neuropharmacol 21, 28-34.

Yoon, T., Okada, J., Jung, M.W., Kim, J.J., 2008. Prefrontal cortex and hippocampus subserve different components of working memory in rats. Learn Mem 15, 97-105.

Yoshida, R., Oku, T., Imanishi, J., Kishida, T., Hayaishi, O., 1986. Interferon: a mediator of indoleamine 2,3-dioxygenase induction by lipopolysaccharide, poly(I) X poly(C), and pokeweed mitogen in mouse lung. Arch Biochem Biophys 249, 596-604.

Yu, P., Di Prospero, N.A., Sapko, M.T., Cai, T., Chen, A., Melendez-Ferro, M., Du, F., Whetsell, W.O., Jr., Guidetti, P., Schwarcz, R., Tagle, D.A., 2004. Biochemical and phenotypic abnormalities in kynurenine aminotransferase II-deficient mice. Mol Cell Biol 24, 6919-6930.

Yu, P., Li, Z., Zhang, L., Tagle, D.A., Cai, T., 2006. Characterization of kynurenine aminotransferase III, a novel member of a phylogenetically conserved KAT family. Gene 365, 111-118.

Zorrilla, E.P., Luborsky, L., McKay, J.R., Rosenthal, R., Houldin, A., Tax, A., McCorkle, R., Seligman, D.A., Schmidt, K., 2001. The relationship of depression and stressors to immunological assays: a meta-analytic review. Brain Behav Immun 15, 199-226.