PS126
DAPK1 interaction with NMDA receptor NR2B subunit underlies the rapid antidepressant effect
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Abstract
The limitations of current pharmacotherapies for depression highlight the need for rapid acting antidepressants. The gluta-mate mechanisms in major depressive disorder have attracted much attention in recent years as a promising target for develop-ing novel antidepressants. NR2A- and NR2B-containing NMDA receptors are considered as the major isoforms of functional NMDA receptor channels in CNS neurons. Death-associated protein kinase 1 (DAPK1) couples NR2B subunits at extrasynaptic sites to regulate the NMDA receptor channel conductance. However, still unknown is the involvement of DAPK1 and NR2B subunit interaction in the rapid antidepressant effect. Here we found high glutamate abundance accompanied by high expression of DAPK1, p-NR2B at Ser1303 and low expression of p-CREB, BDNF and synaptic proteins in the medial prefrontal cortex of rats that were subjected to chronic unpredictable mild stress (CUS). Blockade of astrocytic glutamate transporter-1 in the mPFC is sufficient to induce depressant-like behavior and cause similar molecular changes. Administration of DAPK1 inhibitor and selective NR2B antagonist but not NR2A antagonist produced a rapid antidepressant effect. Uncoupling DAPK1 from NR2B subunit by application of cell membrane permeable Tat-NR2Bct peptide also produced a rapid antidepressant effect by reducing the immobility in the forced swim test and reversing CUS-induced decrease in sucrose preference. Moreover, we found that selective NR2B antagonist did not produce rewarding effect measured with conditioned place preference paradigm. Together, our findings suggest that DAPK1 interaction with NMDA receptor NR2B subunit acts as a critical component in the rapid antidepressant actions.

Keywords: depression; glutamate; NMDA receptor; NR2B subunit; DAPK1

PS127
Antidepressant amitriptyline activates matrix metalloproteinase in astroglial cells: involvement in glial cell line-derived neurotrophic factor expression
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Abstract
Background: Glial cells, especially astrocytes have been implicated in the pathophysiology of mood disorder and the efficacy of antidepressants. The tricyclic antidepressant amitriptyline induces a G_{o/i} matrix metalloproteinase (MMP)/fibroblast growth factor receptor (FGFR)/ERK cascade, which is crucial for glial cell line-derived neurotrophic factor (GDNF) mRNA expression in rat C6 astroglial cells (C6 cells), primary astrocytes. However, the identity of the MMP involved has yet to be identified. The current study identified the mechanism of MMP activation induced by amitriptyline and the MMP subtypes involved.

Methods: C6 cells and primary astrocytes were used in the following experiments. The level of GDNF mRNA was measured by real-time PCR and the activity of MMP-2, -9 was measured by gelatin-zymography.

Results: Matrix metalloproteinase-2, -3 and -9 were expressed in C6 cells. Following amitriptyline treatment, MMP-9 activity in culture medium increased without any change in mRNA levels, whereas no change in MMP-2 activity was observed. A similar response with MMP-9 was observed with different classes of antidepressants, but not with drugs lacking antidepressant activity and monoamines. Amitriptyline-induced ERK activation/GDNF mRNA expression was blocked by MMP-3 and MMP-9 inhibition, but not by MMP-2 inhibition. Treatment with exogenous MMP-3 and MMP-9 increased GDNF mRNA expression. Amitriptyline-induced MMP-9 activation was suppressed with MMP-3 inhibition and exogenous MMP-3-induced GDNF mRNA expression suppression was blocked by MMP-9 inhibition, indicating that MMP-3 regulates MMP-9 activity. The FGFR-induced ERK/GDNF cascade was not blocked by MMP inhibition, indicating that MMP activation is upstream of FGFR activation. Furthermore, amitriptyline-induced MMP-9 activation is not direct but via intracellular signaling, as Go_{o/i} inhibition and Src family tyrosine kinases inhibition blocked amitriptyline-induced MMP-9 activation.

Conclusion: The current results elaborate a potential non-mono-amine mediated mechanism of antidepressant action involving MMP activation and intracellular signaling in astrocytes.

PS128
Ketamine R(-) and S(+) Pharmacological Actions; Together or Separate Rapid Acting Antidepressants
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Abstract
It is well documented that low dose racemic ketamine, a mixture of two enantiomers, has a rapid antidepressant effect within hours that may last for 1 to 2 weeks. Racemic ketamine probably will never become a major treatment of depression because it has significant schizophrenimimetic and drug abuse side effects. The mechanism of its rapid antidepressant actions continues to be a major target of current and future research. This presentation addresses the fact that the two enantiomers of racemic ketamine have overlapping but also different pharmacological actions that target either antidepressant, analgesic or anesthetic uses. Detailed pharmacological dose-effect studies for each enantiomer and their metabolites must involve classic basic science methodology. A wide range of ketamine concentrations vary from 50-10000 ng/ml (0.21 - 42.1 nmol/ml) in vitro and in vivo. After i.v administration, anesthetic concentrations are greater than 2,000 with peak levels as high as 10,000 ng/ml. Patients return to consciousness -1,100 ng/ml. Low dose ketamine has analgesic and antidepressant effects with venous plasma concentrations of ~150 ng/ml i.v. and as low as 40ng/ml oral administration. Given a preferential distribution of ketamine brain:plasma ratio of 6.5, its equivalent concentration is ~227 ng/ml in brain. In vitro NMDAR antagonism has a Ki value of ~190 ng/ml for S(+)-ketamine and ~360 ng/ml for R(-)-ketamine. These are similar to the low concentrations for antidepressant effects. In vivo, a low dose of racemic ketamine also
inhbits serotonin reuptake. Compared to S-(+)-ketamine, the
less potent anesthetic enantiomer R(-)-ketamine requires much
more additional concentration effect studies for its analgesic
and antidepressant effects.

PS129
Low TNFAIP3 was normalized with antidepressant in
patients with major depressive disorder
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Abstract
Rationale: Abnormalities in Toll-like receptors (TLRs) expres-
sion in depression have been inferred in part from observed
increases in TLR4 levels in peripheral blood mononuclear cells
(PBMCs) and postmortem brains of depressed and suicidal
patients. Activation of the TLR4 pathway partially explained the
inflammatory status in patients with major depressive disorder.
However, the negative regulators for TLR4 pathway have never
been investigated.

Objectives: In vivo, the mRNA expression levels of negative regu-
lation genes including SOCS1, TOLLIP, SIGIRR, MyD88, NOD2, and
TNFAIP3 in PBMCs were examined in 56 patients with MDD and
35 health controls. The mRNA expression levels were assessed in parallel with a housekeeping gene using qRT-PCR before and after treatment with antidepressants. We also investigated the in vitro effects of fluoxetine on the TNFAIP3 expression. First, TNFAIP3 in human monocytes (THP-1 cell line) was measured by qRT-PCR after treated with fluoxetine (10⁻⁶–10⁻⁷ M). Second, we pretreated monocytes, which had TNFAIP3 gene knockdown, with fluoxetine before LPS-stimulation. Then, interleukin 6 (IL-6) and tumor necrotic factor alpha (TNFα) were measured by qRT-PCR.

Results: In vivo, TOLLIP, MyD88, NOD2 and TNFAIP3 were
expressed at lower levels in patients with MDD. Only TNFAIP3
was significantly increased and normalized by treatment with
antidepressants for 4 weeks. In vitro, fluoxetine could signifi-
cantly increase TNFAIP3 mRNA expression in human mono-
cyte. The suppressive effects of fluoxetine on IL-6 and TNFα decreased partially after knockdown of TNFAIP3 gene.

Conclusions: These findings suggest that antidepressant treat-
ment exerts anti-inflammatory effects in patients with MDD
partially through increasing expression of TNFAIP3 gene. Further
studies investigating the effects of manipulating TNFAIP3 gene
on depression is needed to fully elucidate the underlying mechanism.

Keywords: negative regulation, toll-like receptor, innate immu-
nity, major depressive disorder, inflammation

PS130
Maternal fluoxetine treatment influences
anxiety- and depressive-like behaviours in
adolescent offspring: a rodent model
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Abstract
Objective: Approximately 10% of pregnant women are pre-
scribed antidepressant drugs, most commonly the selective
serotonin reuptake inhibitor, Fluoxetine. Fluoxetine crosses the
placenta and is excreted in breast milk raising concerns regard-
ing the consequences of infant exposure. The aim of this study
was to evaluate the effects of maternal Fluoxetine treatment on
offspring behaviours of relevance to neurodevelopmental and
psychiatric disorders, using a rodent model of depression.

Methods: Sprague-Dawley (SD; healthy model) and Wistar-
Kyoto (WKY; depression model) pregnant rats were treated with
Fluoxetine (10mg/kg/day) or vehicle, from gestational
day 0 to postnatal day 14 (~5 weeks in total). Once offspring
reached adolescence (~5 weeks of age), locomotor activity, anxiety-
like and depressive-like behaviours were assessed using the
open field test (OFT), elevated plus maze (EPM) and forced
swim test (FST).

Results: Fluoxetine exposed offspring displayed an increase in
distance travelled in the OFT, an effect that was independent of
rat strain and sex. Fluoxetine exposure also caused an increase
(up to 50%) in time spent in the corners of the OFT in SD male
and WKY female rats compared to their respective vehicle
controls. Similarly, in the EPM, maternal Fluoxetine treatment
resulted in a significant increase in time spent in the closed
arms in SD males and WKY males (29–52%). A similar trend
was observed in females, but this did not reach significance.
In the FST, maternal Fluoxetine treatment increased immobility
time in exposed offspring (28%), an effect that was independent of
strain or sex.

Conclusion: Maternal Fluoxetine treatment resulted in signifi-
cant increases in anxiety-like and depressive-like behaviours in
exposed offspring, largely independent of the rat model used.
However, further studies in various models of maternal depres-
sion are required to confirm these preliminary findings and
establish the effects of Fluoxetine exposure on the developing
brain.

PS131
Transcriptomic evidence for dematuration of the
mouse frontal cortex and hippocampus by chronic
antidepressant treatment
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Abstract
The selective serotonin reuptake inhibitor, fluoxetine (FLX),
is widely used to treat depression and anxiety disorders, but
mechanisms underlying its antidepressant effect remain largely
unknown. Previous studies that evaluated several molecular
and/or electrophysiological features of the maturation stages of
each neuron type have demonstrated that FLX treatment can
reverse the established maturation of certain types of neurons
in the hippocampus and frontal cortex (FC). However, this dema-
turation effect of FLX in the adult brain has not been assessed
with regard to genome-wide gene expression patterns.

In this study, we compared gene expression patterns in the
FLX-treated FC and hippocampus of adult mice with those of
the corresponding brain regions of normal infant mice. The gene
expression patterns of FLX-treated mice significantly resembled
those of normal infant mice in the FC and, to a large extent, in
the hippocampus. In addition, time-course analyses of the ages
of infant mice used in the comparisons with FLX-treated mice
indicated that the gene expression patterns of FLX-treated mice
were most similar to those of the youngest infants examined
(1-week-old hippocampus and 2-week-old FC).