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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 glutamate mechanisms in major depressive disorder have attracted much attention in recent years as a promising target for developing 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 depression-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-NR2B$_{CT}$ 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

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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$\alpha_{i/o}$/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 was suppressed 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 G$\alpha_{i/o}$ inhibition and Src family tyrosine kinases inhibition blocked amitriptyline-induced MMP-9 activation.

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

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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 schizophrenomimetic 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 40 ng/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