GluA1 and its PDZ-interaction: a role in experience-dependent behavioral plasticity in the forced swim test

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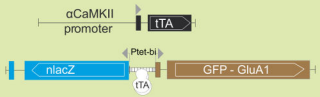
Background

Although current pharmacological treatment for depression mainly targets the monoaminergic system, recent studies indicate that glutamatergic neurotransmission is more principally involved in the neuropathology of depression [1]. Importantly, pharmacological blockade of NMDA receptors (e.g. with ketamine) has relatively long-lasting antidepressant effects with rapid onset [2]. The antidepressant properties of ketamine require activation and synaptic incorporation of GluA1-containing AMPA receptors [3]. Moreover, hippocampal samples from clinically depressed patients display reduced mRNA levels for GluA1 [4]. These findings argue that GluA1-dependent synaptic plasticity might be critically involved in the development of depression [1,2].

This study aimed to test how GluA1-dependent plasticity contributes to the experience-dependent expression of depression-related behaviors by making use of GluA1 transgenic mice and targeted deletion of GluA1 in hippocampus.

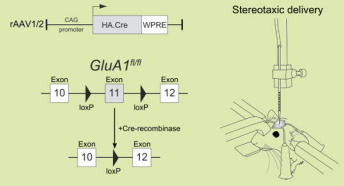
Methods

Transgenic expression of GluA1: In A1.1 mice a native GFP-GluA1 was expressed in the GluA1-knockout (*GluA1*$^{-/-}$) background, while in TG8.1 the most C-terminal leucine of GFP-GluA1 was lacking, thereby blocking PDZ-interaction.

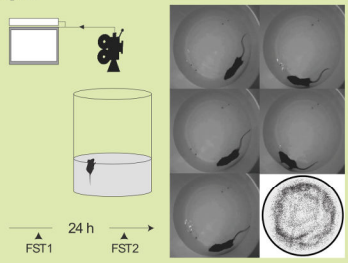


PDZ-Consensus: ---(T/S)XΦ
Native (A1.1): ---CMSHSSGMPLGATGL
Mutant (TG8.1): ---CMSHSSGMPLGATG

Virus-mediated deletion of GluA1: A Cre-recombinase expressing recombinant adeno-associated virus (rAAV) was stereotaxically injected into the dorsal or ventral hippocampus of mice with floxed GluA1-allels (GluA1$^{fl/fl}$), thereby deleting GluA1 from all infected cells.

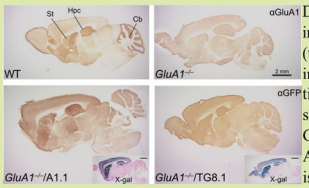


Forced swim test (FST): Mice were exposed to two sessions of forced swimming, 24-hours apart (FST1 [15 min] and FST2 [10 min] respectively). All behavioral assessment was fully computerized by custom-written software running in MATLAB. Latency to immobility and cumulative immobility were used as measures of behavioral despair.

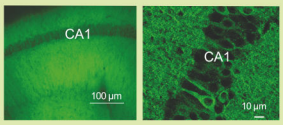


Results

Expression of GluA1 across the genotypes studied

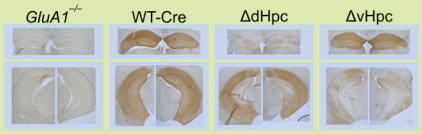


DAB - immunostainings against GluA1 (top) or GFP (bottom) in saggital brain sections of adult mice showing that the GFP-GluA1 transgene in A1.1 and TG8.1 mice is strongly expressed in most forebrain areas including striatum (St), hippocampus (Hpc) and Cerebellum (Cb). Insets illustrate β-galactosidase-activity (X-gal staining).

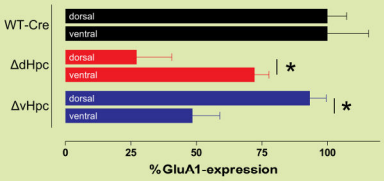


GFP-fluorescence in TG8.1 mice observed with a 20x (left) and 63x (right) objective. The GFP-GluA1 gene was preferentially localized to the dendrites indicating intact dendritic transport of receptors containing the transgenically expressed GFP-GluA1 subunits.

Hippocampal expression of GluA1 in virus injected mice

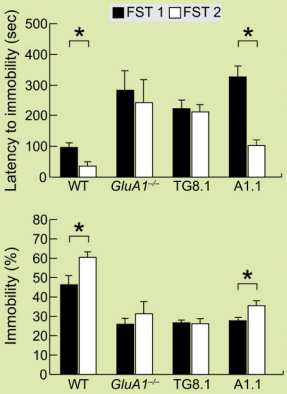


Representative images of GluA1-stained sections from dorsal (top) and ventral (bottom) hippocampus



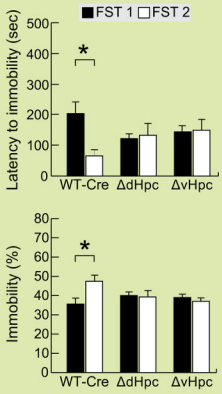
Staining intensity was quantified relative to Cre-injected wild-type mice (WT-Cre). Quantification showed symmetrical removal of GluA1, with significantly more GluA1 lacking in targeted areas (*p<0.05).

Behavioral despair in GluA1 transgenic mice



Mice globally lacking GluA1 (*GluA1*$^{-/-}$, N=7) and mice with blocked PDZ-interaction of GluA1 (TG8.1 mice, N=9) are impaired in the experience dependent expression of behavioral despair in the forced swim test (FST), as shown by comparable latency to (left) and cumulative immobility (right) across sessions (FST1 vs FST2). Transgenic GFP-GluA1 expression in A1.1 mice (N=7) rescues this effect, as latency to immobility and cumulative immobility is reduced across sessions in these mice, as it is in wild type (WT, N=10) controls (*p<0.05).

Behavioral despair in mice lacking hippocampal GluA1



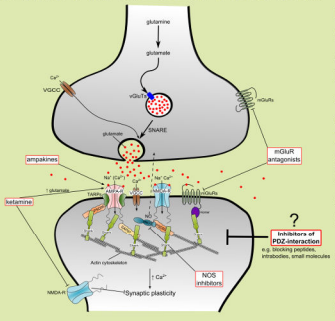
Mice with selective deletion of GluA1 in dorsal (ΔdHpc, N=6) or ventral (ΔvHpc, N=13) are strongly impaired in the experience-dependent expression of behavioral despair, as shown by comparable levels of latency to immobility (left) and cumulative immobility (right) across sessions (FST1 vs FST2). In contrast, wild type mice injected with a Cre-expressing virus (WT-Cre, N=7) show normal reduction in latency to immobility and increased overall immobility (*p<0.05).

Conclusions & Outlook

- Experience-dependent expression of behavioral despair in the forced swim test (FST) is dependent on GluA1-containing AMPA receptors and their interaction with PDZ-domain containing proteins (e.g. SAP97, Shank3).

- Hippocampal GluA1-containing AMPA receptors are required for experience-dependent expression of behavioral despair in the FST.

- These results link GluA1-dependent synaptic plasticity with the pathophysiology of depression and might provide a new target for antidepressant treatment.



Emerging drug targets at the glutamatergic synapse in the treatment of depression. Inhibition of NMDA-Rs and mGluRs and activation of AMPA-Rs has antidepressant-like effects. Similarly, blocking the PDZ-interaction of proteins (e.g. GluA1, nNOS) in the glutamatergic postsynaptic density might exert comparable antidepressant-like effects.

Abbreviations: AMPA-R = AMPA receptor; mGluRs = metabotropic glutamate receptors; NMDA-R = NMDA receptor; nNOS = neuronal nitric oxide synthase; TARPs = transmembrane AMPA receptor regulatory proteins; VGCC = voltage gated calcium channel; vGluTs = vesicular glutamate transporters

References:

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[3] Li, N., Lee, B., Liu, R.-J., Banasr, M., Dwyer, J. M., Iwata, M., Li, X.-Y., et al. (2010). mTOR-dependent synapse formation underlies the rapid antidepressant effects of NMDA antagonists. Science, 329(5994), 959–64.

[4] Duric, V., Banasr, M., Stockmeier, C. A., Simen, A. A., Newton, S. S., Overholser, J. C., Jurjus, G. J., et al. (2012). Altered expression of synapse and glutamate related genes in post-mortem hippocampus of depressed subjects. International Journal of Neuropsychopharmacology, i, 1–14.

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