J Med Sci 2010;30(1):003-010
http://jms.ndmctsgh.edu.tw/3001003.pdf

Ethanol and NMDA Receptor Interactions: Implications for Pharmacotherapeutic Treatments

Michael Feyder[1], Marguerite C. Camp[1*], Andrew Holmes[1], and Yi-Chyan Chen[2]

[1]Section on Behavioral Science and Genetics, Laboratory for Integrative Neuroscience, National Institute on Alcoholism and Alcohol Abuse, NIH, Bethesda, MD, USA
[2]Department of Psychiatry, Tri-Service General Hospital, National Defense Medical Center, Taipei, Taiwan, Republic of China.

Alcohol abuse and dependence constitutes a significant societal and global burden; however, the basic scientific knowledge underlying the development and maintenance of alcohol dependence is limited, resulting in few pharmacotherapies and high rates of relapse following abstinence. A growing body of evidence supports an interaction between ethanol (EtOH) and N-methyl-D-aspartate receptors (NMDARs) in mediating the acute and chronic effects of EtOH. The aim of this review is to synthesize the current knowledge of this interaction on the molecular, cellular and behavioral levels and highlight possible avenues for pharmacotherapeutic treatments.

Key words: ethanol, NMDA receptor, neurotransmission, alcoholism

Alcohol abuse and dependence constitutes a significant societal and global burden as a result, in part, of its actions on the central nervous system (CNS)[1]. Despite this substantial impact, the basic scientific knowledge underlying the development and maintenance of alcohol dependence is limited, resulting in few pharmacotherapies and high rates of relapse following abstinence[2,3]. A growing body of evidence, however, supports an interaction between ethanol (EtOH) and N-methyl-D-aspartate receptors (NMDARs), a subclass of receptors activated by the major excitatory neurotransmitter glutamate, in mediating the acute and chronic effects of EtOH. The aim of this review is to synthesize the current knowledge of this interaction on the molecular, cellular and behavioral levels and highlight possible avenues for pharmacotherapeutic treatments.

Acute EtOH and NMDARs

Initial evidence for an acute interaction between EtOH and NMDARs was first reported when electrophysiological data revealed that EtOH dose-dependently inhibits NMDARs at concentrations that parallel intoxication in human subjects[4]. This finding was substantiated at the behavioral level by self-administration studies demonstrating that NMDAR antagonists substitute for EtOH in rats[5-7] while human subjects report EtOH-like effects following NMDAR antagonist administration[8,9]. Finally, pretreatment with an NMDAR antagonist following administration of EtOH prevents the development of acute tolerance[10,11].

Acute EtOH and NMDAR subunits

NMDARs are composed of two obligatory NMDA receptor 1 (NR1) subunits and at least one type of regulatory NR2A-D subunit or NR3A-B subunits[12]. The strongest subunit-specific evidence of EtOH inhibition focuses on the NR2A and NR2B subunits. Recombinant studies in Xenopus oocytes demonstrate an increased inhibitory sensitivity to EtOH in receptors expressing the NR2A and NR2B subunits, as compared to the NR2C and NR2D subunits, with equal sensitivity of the former two subunits in HEK 293 cells[13-15]. A more detailed analysis revealed that EtOH interacts with the fourth membrane-associated domain of the NR2A subunit in HEK 293 cells while no such data to date has been reported for the NR2B subunit[16,17].

In neurons, however, a prominent role for the NR2B subunit has emerged. Several studies demonstrated that select neurons sensitive to the NR2B antagonist ifenpro-

Received: October 29, 2009; Revised: November 9, 2009; Accepted: November 11, 2009
*Corresponding Author:Marguerite C. Camp, Section on Behavioral Science and Genetics, Laboratory for Integrative Neuroscience, National Institute on Alcohol Abuse and Alcoholism, 5625 Fisher's Lane Room 2N09, Bethesda, MD, USA 20892-9304. Tel: +301-443-4052; Fax: +301-480-1952; E-mail: campmc@mail.nih.gov

dil were similarly sensitive to EtOH, suggesting that the physiological consequence of EtOH inhibition is mediated by NR2B-expressing receptors in developmentally mature neurons[18-21].

Behavioral studies involving pharmacologically manipulated mice targeted at the NR2B subunit support its involvement in specific acute behavioral responses to EtOH; pretreatment with a NR2B antagonist increased the sedative/hypnotic effect of acute EtOH in mice[22,23].

Acute EtOH and NMDAR downstream signaling pathways

NMDARs are ligand-gated ionotropic receptors that, upon glutamate binding, are permeable to calcium ions and initiate downstream signal transduction pathways[12]. Expectedly, EtOH inhibition reduces calcium ion influx and calcium-dependent synthesis of cyclic GMP[24,25]. Further research, however, has detailed specific downstream responses to EtOH.

NMDARs interact with RhoGTPase-activating proteins (RhoGAPs) to regulate dendritic spine morphology[26], and various RhoGAP splice variants in Drosophila mediate the stimulating or sedating effects of acute EtOH at low or high doses repectively[27]. Furthermore, mice lacking Eps8, a protein that interacts with NMDARs and regulates actin dynamics, show a diminished behavioral response and corresponding resistance to actin remodeling following acute EtOH administration[28]. Taken together, these results suggest that EtOH inhibition of NMDARs influence cytoskeletal remodeling and may contribute to the structural adaptations associated with chronic EtOH administration[29].

Quantitative trait locus and gene array analyses of mice selectively bred for behavioral sensitivity to acute EtOH reveal an influential role for, among other genes, NMDARs and its downstream modulators. Studies report alterations of the NR1 subunit, zinc finger protein 179, a member of the RING finger protein family induced following NMDAR activation[30-32], and peroxidredoxin, a protein with antioxidant function in response to NMDAR activation[33]. Collectively, these data suggest NMDARs and various downstream molecules influence the acute behavioral response to EtOH.

EtOH has been shown to inhibit NR2A- and NR2B-expressing receptors, in part, by dephosphorylation via tyrosine phosphatases[34]. Compensatory phosphorylation of NMDARs, which increases open channel probability by reducing the voltage-dependent magnesium ion blockage[35], has emerged as a common cellular mechanism, an effect termed homeostatic phosphorylation. Following acute EtOH administration, dopamine receptor 1 (D1) activation of dopamine and cAMP-regulated phosphoprotein-32 kD (DARPP-32) phosphorylates the NR1 subunit in the nucleus accumbens of mice[36], while transient phosphorylation of NR1 by the cAMP-dependent protein kinase A (PKA) has been reported in the ventrolateral medulla in the rat[37]. Interestingly, mice lacking various isoforms of adenylate cyclase, the enzyme responsible for cAMP synthesis, display abnormal PKA substrate phosphorylation, increased sedation following EtOH administration, and decreased voluntary EtOH consumption[38]. These studies demonstrate an influential role of adenylate cyclase, cAMP, DARPP-32, and NR1 homeostatic phosphorylation in response to acute EtOH administration.

EtOH also induces regionally specific homeostatic phosphorylation of the NR2B subunit via activation of Fyn, a tyrosine kinase associated with EtOH dependence in human populations[39]. Acute EtOH administration increases NR2B tyrosine phosphorylation in the hippocampus, an effect not seen in Fyn knock-out (KO) mice[40,41]. The homeostatic phosphorylation by Fyn is manifested behaviorally; Fyn KO mice show an enhanced sensitivity to the sedative/hypnotic effects of ethanol[41] while an opposite effect is observed following overexpression of Fyn in the forebrain[42].

Acute EtOH administration also increases NR2B phosphorylation and Fyn activation in the dorsal striatum[43]. Given that this region is implicated in habit-based learning and sensitive to drugs of abuse[44], injection of a Fyn inhibitor into this region reduces EtOH self-administration in rats[43]. Interestingly, following the removal of EtOH from brain slice preparations, the electrophysiological response of NR2B-expressing receptors in the dorsal striatum is enhanced (termed ‘long term facilitation’), providing a plausible mechanism for sustained cellular alterations[43].

Finally, acute EtOH activates the GTP-binding protein H-Ras and inhibits the tyrosine kinase Src to promote the endocytosis of NR2A-expressing receptors[45]. This selective endocytosis results in an increase in the proportion of NMDARs solely expressing the NR2B subunit as compared to those expressing both the NR2A and NR2B subunits[45]. This shift highlights the recruitment of cellular machinery responsible for receptor trafficking in response to EtOH and may be the initial manifestation of a more stable shift in subunit ratio observed after chronic ethanol administration.

Chronic EtOH and NMDARs

Behavioral tolerance following chronic EtOH treatment (CET) is dependent on repeated NMDAR inhibition and is blocked following pre-treatment of an NMDAR antagonist[46]. CET results in an up-regulation of NMDARS as measured by an increase in NMDAR radioligand binding[47,48] (but see[49,50]); however, further research suggests that this up-regulation may be subunit specific.

Supporting this hypothesis, CET increases NMDA-mediated neurotoxicity in neurons found in the cortex[51], the hippocampus[52], and the granule cell layer of the cerebellum[53]. NMDAR-mediated neurotoxicity is thought to be the result of excessive activation of NR2B-expressing receptors[54] suggesting a subunit-specific up-regulation of these receptors. Although the NR1 and NR2A subunits have been reported to be up-regulated following CET, NR2B up-regulation has been the most consistently reported and replicated. The subunit is up-regulated in the cortex[55-58], the hippocampus[55,59], and the amygdala[60,61]. In addition, the NR1 subunit is up-regulated in the ventral tegmental area (VTA)[62]. Importantly, glutamatergic inputs from the prefrontal cortex, hippocampus and amygdala, along with modulating dopaminergic input from the VTA, converge and are integrated in the nucleus accumbens, a region important for reinforced behavior[63,64]. Thus, it is hypothesized that NR2B up-regulation following CET may underlie the adaptive neurocircuitry driving persistent EtOH-seeking behavior[65].

Supporting this hypothesis, CET alters the electrophysiological and morphological properties of neurons. Following CET, ifenprodil occludes the inhibitory effects of EtOH on excitatory postsynaptic currents in the central nucleus of the amygdala, an effect that was less in naïve rats, and suggests an up-regulation of functional NR2B-expressing receptors[66]. Similar results were observed in the lateral/basolateral amygdala[61]. Structurally, CET results in an enlargement of dendritic spines as measured by increased immunoreactivity of the cytoskeletal protein F-actin and the postsynaptic scaffolding protein PSD-95, which serves to stabilize synaptic NMDARs and organize downstream signaling molecules[29]. Taken together, CET alters the functional and structural properties of neurons possibly by up-regulation of NR2B-expressing receptors.

CET-induced up-regulation of NR2B subunits involves a variety of mechanisms. An extracellular physical interaction between tissue plasminogen activator (tPA) and the NR2B subunit is required for up-regulation following CET[67]. Intracellularly, CET increases DNA binding and promoter activity of the transcription factor activator protein 1 (AP-1) at its NR2B-specific site[68,69]. AP-1 complexes, heterodimers consisting of members from the Fos, Jun and activating transcription factor (ATF) protein families[70], differ between cortical and cerebellar granule cells following CET but may involve these various protein family[68,69].

It is therefore hypothesized that repeated inhibition of NMDARs following CET results in more structural homeostatic response involving the up-regulation of NMDARs. Disruption of this inhibition or in receptor up-regulation results in abnormal EtOH-related behavior. Pre-treatment with some, but not all, NMDAR antagonists prior to EtOH administration prevents the acquisition of a conditioned place preference[71,72], and has been reported in mice lacking the NR2A subunit[73]. Furthermore, mice in which the NR1 subunit was engineered for a reduced affinity for glycine, a co-agonist that potentiates NMDAR channel properties, show reduced anxiety-like behavior and motor incoordination following CET[74].

Once NMDAR up-regulation is established, it is hypothesized that elimination of EtOH results in a hyperglutamatergic state that perpetuates EtOH-seeking behavior[75]. Global administration[76,77] or specific injection into the nucleus accumbens[78,79] of NMDAR antagonists reduces EtOH self-administration in rodents. Interestingly, administration of an NMDAR antagonist to rats exposed to stimuli associated with CET reduces EtOH-seeking behavior when re-presented with EtOH-cues[80]. Together, these studies suggest that modification of excessive glutamatergic transmission may reduce EtOH-seeking behavior.

Alcohol dependence and development of pharmacotherapies

Clinical research on alcohol dependence supports the involvement of excessive glutamatergic transmission mediated by NMDARs. Recovering alcohol-dependent subjects report less subjective effects after administration the NMDAR antagonist ketamine, suggestive of a pre-disposed tolerance due to EtOH-induced NMDAR upregulation[81]. Non-dependent subjects with a family history of alcoholism also showed attenuated responses to ketamine compared to those with no family history and suggest that heightened glutamatergic transmission may be a pre-disposing trait to be exacerbated by chronic alcohol consumption[82]. Recovering alcohol-dependent subjects also have increased levels of glutamate, glycine and the resulting markers of oxidative stress in their cerebral spinal fluid[83]. Finally, the NMDAR antagonist memantine reduces cue-induced craving in recovering alcohol-dependent subjects[84].

Give the evidence for altered glutamatergic transmission, preventative and corrective modulation may serve as a potential pharmacological target in the treatment of alcohol dependence. Preventative targets include NMDARs and various downstream signaling molecules activated following NMDAR inhibition. Prevention of NMDAR up-regulation by inhibition of transcription factors, trafficking proteins, synaptic incorporation and stabilizing scaffolding proteins may prevent excessive glutamatergic transmission. Once receptor up-regulation has occurred, targets that restore proper glutamatergic tone by antagonizing or down-regulating NMDARs, possibly targeted at NR2B-expressing receptors[85], or inhibiting downstream signaling molecules may provide corrective therapy.

CONCLUSION

EtOH inhibits NMDARs within the CNS, with possible neural functioning and specific behavioral responses mediated by an interaction with the NR2B subunit. As a result, regionally specific homeostatic responses and downstream signaling molecules are engaged affecting channel function, cytoskeletal remodeling and behavior. Persistent inhibition by EtOH results in up-regulation of NMDARs, consequential alterations in the functional and morphological properties of neurons, and activation of transcription factors to create an excessive glutamatergic state. Attenuation of this enhanced state reduces EtOH-seeking behavior and may provide a pharmacological target for the treatment of alcohol dependence.

REFERENCES

1. Eckardt, MJ, Harford TC, Kaelber CT, Parker ES, Rosenthal LS, Ryback RS, Salmoiraghi GC, Vanderveen E, Warren KR. Health Hazards Associated With Alcohol Consumption. JAMA 1981;246:648-666.
2. Garbutt JC, West SL, Carey TS, Lohr KN & Crews FT. Pharmacological Treatment of Alcohol Dependence: A Review of the Evidence. JAMA 1999;281:1318-1325.
3. Kosten TR & O'Connor PG. Management of Drug and Alcohol Withdrawal. N Engl J Med 2003;348:1786-1795.
4. Lovinger DM, White G & Weight FF. Ethanol inhibits NMDA-activated ion current in hippocampal neurons. Science 1989;243:1721-1724 .
5. Bienkowski P, Stefanski R & Kostowski W. Competitive NMDA receptor antagonist, CGP 40116, substitutes for the discriminative stimulus effects of ethanol. European Journal of Pharmacology 1996;314: 277 .
6. Kotlinska J & Liljequist S. The NMDA/glycine receptor antagonist, L-701,324, produces discriminative stimuli similar to those of ethanol. European Journal of Pharmacology 1997;332:1.
7. Hodge CW, Cox AA, Bratt AM, Camarini R, Iller K, Kelley SP, Mehmert KK, Nannini MA, Olive MF. The discriminative stimulus properties of self-administered ethanol are mediated by GABA(A) and NMDA receptors in rats. Psychopharmacology 2001;154:13-22.
8. Krystal JH, Petrakis IL, Webb Elizabeth, Cooney NL, Karper LP, Namanworth Sheila ; Stetson Philip, Trevisan LA, Charney DS. Dose-Related Ethanol-like Effects of the NMDA Antagonist, Ketamine, in Recently Detoxified Alcoholics. Arch Gen Psychiatry 1998;55:354-360.
9. Dickerson D, Pittman B, Ralevski E, Perrino A, Limoncelli D, Edgecombe J, Acampora G, Krystal JH, and Petrakis I. Ethanol-like effects of thiopental and ketamine in healthy humans. J Psychopharmacol, 0269881108098612 (2008).
10. Khanna JM, Shah G, Weiner J, Wu PH. & Kalant H. Effect of NMDA receptor antagonists on rapid tolerance to ethanol. European Journal of Pharmacology 1993;230:23.
11. Karcz-Kubicha M. & Liljequist S. Effects of post-ethanol administration of NMDA and non-NMDA receptor antagonists on the development of ethanol tolerance in C57BI mice. Psychopharmacology 1995;120:49.
12. Cull-Candy S, Brickley S & Farrant M. NMDA receptor subunits: diversity, development and disease. Current Opinion in Neurobiology 2001;11:327 .
13. Mirshahi T & Woodward JJ. Ethanol sensitivity of heteromeric NMDA receptors: Effects of subunit assembly, glycine and NMDAR1 Mg^{2+}-insensitive mutants. Neuropharmacology 1995;34:347.
14. Benson Chu & Vellareddy Anantharam. Ethanol Inhibition of Recombinant Heteromeric NMDA Channels in the Presence and Absence of Modulators. Journal of Neurochemistry 1995;65:140-148.
15. Blevins T, Mirshahi T & Woodward JJ. Increased agonist and antagonist sensitivity of N-methyl--aspartate stimulated calcium flux in cultured neurons following chronic ethanol exposure. Neuroscience Letters 1995;200:214.
16. Ren H, Honse Y & Peoples RW. A Site of Alcohol

Action in the Fourth Membrane-associated Domain of the N-Methyl-D-aspartate Receptor. J. Biol. Chem. 2003;278:48815-48820.
17. Honse Y, Ren H, Lipsky RH. & Peoples RW. Sites in the fourth membrane-associated domain regulate alcohol sensitivity of the NMDA receptor. Neuropharmacology 2004;46:647.
18. Yang X., Criswell HE, Simson P, Moy S. & Breese GR. Evidence for a selective effect of ethanol on N-methyl-d-aspartate responses: ethanol affects a subtype of the ifenprodil-sensitive N- methyl-d-aspartate receptors. J Pharmacol Exp Ther 1996;278:114-124.
19. Michael J. Courtney, Jyrki P. Kukkonen & Karl EO. Åkerman. Ethanol Specifically Inhibits NMDA Receptors with Affinity for Ifenprodil in the Low Micromolar Range in Cultured Cerebellar Granule Cells. Journal of Neurochemistry 1997;69:2162-2168.
20. Lovinger DM. Developmental decrease in ethanol inhibition of N-methyl-D-aspartate receptors in rat neocortical neurons: relation to the actions of ifenprodil. J Pharmacol Exp Ther 1995;274:164-172.
21. Mameli M, Zamudio PA, Carta M. & Valenzuela CF. Developmentally Regulated Actions of Alcohol on Hippocampal Glutamatergic Transmission. J. Neurosci. 2005;25:8027-8036.
22. Janel M B-R & Andrew H. Functional roles of NMDA receptor NR2A and NR2B subunits in the acute intoxicating effects of ethanol in mice. Synapse 2005;56:222-225.
23. Benjamin P, Chen Y-C, Abigail JE , Rose-Marie K, Masayoshi M, Andrew H. Role of Major NMDA or AMPA Receptor Subunits in MK-801 Potentiation of Ethanol Intoxication. Alcoholism: Clinical and Experimental Research 2008;32:1479-1492.
24. Paula LH, Carolyn SR, Frances M. & Boris T. V-methyl-D-aspartate Receptors and Ethanol: Inhibition of Calcium Flux and Cyclic GMP Production. Journal of Neurochemistry 1989;52:1937-1940.
25. Dildy JE & Leslie SW. Ethanol inhibits NMDA-induced increases in free intracellular Ca^{2+} in dissociated brain cells. Brain Research 1989;499: 383.
26. Nakazawa T, Kuriu T, Tezuka T, Umemori H, Okabe S, Yamamoto T. Regulation of dendritic spine morphology by an NMDA receptor-associated Rho GTPase-activating protein, p250GAP. Journal of Neurochemistry 2008;105:1384-1393.
27. Rothenfluh A, Threlkeld RJ, Bainton RJ, Tsai LT, Lasek AW, Heberlein U. Distinct Behavioral Responses to Ethanol Are Regulated by Alternate RhoGAP18B Isoforms. Cell 2006;127:199.
28. Offenhäuser N, Castelletti D, Mapelli L, Soppo BE, Regondi MC, Rossi P, D’Angelo E, Frassoni C, Amadeo A, Tocchetti A, Pozzi B, Disanza A, Guarnieri D, Betsholtz C, Scita G, Heberlein U, Di Fiore PP. Increased Ethanol Resistance and Consumption in Eps8 Knockout Mice Correlates with Altered Actin Dynamics. Cell 2006;127:213.
29. Ezekiel PC-H. & Chandler LJ. Homeostatic plasticity during alcohol exposure promotes enlargement of dendritic spines. European Journal of Neuroscience 2006;24:3496-3506.
30. Tabakoff B, Bhave SV. & Hoffman PL. Selective Breeding, Quantitative Trait Locus Analysis, and Gene Arrays Identify Candidate Genes for Complex Drug-Related Behaviors. J. Neurosci. 2003;23:4491-4498.
31. Zhao Q, Chen K-S, Bejjani BA. & Lupski JR. Cloning, Genomic Structure, and Expression of Mouse Ring Finger Protein GeneZnf179. Genomics 1998;49:394.
32. Ohkawa Noriaki, Kokura Kenji, Matsu-ura Toru, Obinata Takashi, Konishi Yoshiyuki, Tamura Takaaki. Molecular cloning and characterization of neural activity-related RING finger protein (NARF): a new member of the RBCC family is a candidate for the partner of myosin V. Journal of Neurochemistry 2001;78:75-87.
33. Papadia S, Soriano FX, LéveilléF, Martel MA, Dakin KA, Hansen HH, Kaindl A, Sifringer M, Fowler J, Stefovska V, McKenzie G, Craigon M, Corriveau R, Ghazal P, Horsburgh K, Yankner BA, Wyllie DJ, Ikonomidou C, Hardingham GE. Synaptic NMDA receptor activity boosts intrinsic antioxidant defenses. Nat Neurosci 2008;11:476-487.
34. Alvestad RM, Grosshans DR, Coultrap SJ, Nakazawa T, Yamamoto T, Browning MD. Tyrosine Dephosphorylation and Ethanol Inhibition of N-Methyl-D-aspartate Receptor Function. J. Biol. Chem. 2003;278:11020-11025.
35. Chen L & Mae Huang L-Y. Protein kinase C reduces Mg^{2+} block of NMDA-receptor channels as a mechanism of modulation. Nature 1992;356: 521-523.
36. Maldve RE, Zhang TA, Ferrani-Kile K, Schreiber SS, Lippmann MJ, Snyder GL, Fienberg AA, Leslie SW, Gonzales RA, Morrisett RA. DARPP-32 and regulation of the ethanol sensitivity of NMDA receptors in the nucleus accumbens. Nat Neurosci 2002;5:641.
37. Lin H-H, Chang S-J, Shie H-J & Lai C-C. Ethanol inhibition of NMDA-induced responses and acute tolerance to the inhibition in rat rostral ventrolateral medulla in vivo: Involvement of cAMP-dependent

protein kinases. Neuropharmacology 2006;51:747.
38. Maas JW Jr, Vogt SK, Chan GC, Pineda VV, Storm DR, Muglia LJ. Calcium-Stimulated Adenylyl Cyclases Are Critical Modulators of Neuronal Ethanol Sensitivity. J. Neurosci. 2005;25:4118-4126.
39. Pastor IJ, Laso FJ, Inés S, Marcos M & González-Sarmiento R. Genetic association between -93A/G polymorphism in the Fyn kinase gene and alcohol dependence in Spanish men. European Psychiatry 2009;24:191.
40. Kalluri HSG. & Ticku MK. Effect of ethanol on phosphorylation of the NMDAR2B subunit in mouse cortical neurons. Molecular Brain Research 1999;68:159-168.
41. Miyakawa T, Yagi T, Kitazawa H, Yasuda M, Kawai N, Tsuboi K, Niki H. Fyn-Kinase as a Determinant of Ethanol Sensitivity: Relation to NMDA-Receptor Function. Science 1997;278:698-701.
42. Boehm SL 2nd, Peden L, Jennings AW, Kojima N, Harris RA, Blednov YA. Over-expression of the fyn-kinase gene reduces hypnotic sensitivity to ethanol in mice. Neuroscience Letters 2004;372:6.
43. Wang J, Carnicella S, Phamluong K, Jeanblanc J, Ronesi JA, Chaudhri N, Janak PH, Lovinger DM, Ron D. Ethanol Induces Long-Term Facilitation of NR2B-NMDA Receptor Activity in the Dorsal Striatum: Implications for Alcohol Drinking Behavior. J. Neurosci. 2007;27:3593-3602.
44. Gerdeman GL, Partridge JG, Lupica CR & Lovinger DM. It could be habit forming: drugs of abuse and striatal synaptic plasticity. Trends in Neurosciences 2003;26:184.
45. Suvarna N, Borgland SL, Wang J, Phamluong K, Auberson YP, Bonci A, Ron D. Ethanol Alters Trafficking and Functional N-Methyl-D-aspartate Receptor NR2 Subunit Ratio via H-Ras. J. Biol. Chem. 2005;280:31450-31459.
46. Wu PH, John Mihic S, Liu J-F, Le AD & Kalant H. Blockade of chronic tolerance to ethanol by the NMDA antagonist, (+)-MK-801. European Journal of Pharmacology 1993;231:157.
47. Hu X-J & Ticku MK. Chronic ethanol treatment upregulates the NMDA receptor function and binding in mammalian cortical neurons. Molecular Brain Research 1995;30:347.
48. Follesa P & Ticku MK. Chronic Ethanol-mediated Up-regulation of the N-Methyl-D-aspartate Receptor Polypeptide Subunits in Mouse Cortical Neurons in Culture. J. Biol. Chem. 1996;271:13297-13299.
49. Margaret FT, Kevin JA. & Bruce EH. Stability of MK-801 Binding Sites Following Chronic Ethanol Consumption. Alcoholism: Clinical and Experimental Research 1994;18:1004-1008.
50. Snell LD, Tabakoff B. & Hoffman PL. Radioligand binding to the N-methyl-d-aspartate receptor/ionophore complex: alterations by ethanol in vitro and by chronic in vivo ethanol ingestion. Brain Research 1993;602:91.
51. Chandler LJ, Hunter N, Colin S & Fulton C. Chronic Ethanol Exposure Potentiates NMDA Excitotoxicity in Cerebral Cortical Neurons. Journal of Neurochemistry 1993;60:1578-1581.
52. Smothers CT, Mrotek JJ. & Lovinger DM. Chronic Ethanol Exposure Leads to a Selective Enhancement of N-Methyl-D-aspartate Receptor Function in Cultured Hippocampal Neurons. J Pharmacol Exp Ther 1997;283:1214-1222.
53. Iorio KR, Reinlib L, Tabakoff B & Hoffman PL. Chronic exposure of cerebellar granule cells to ethanol results in increased N-methyl-D-aspartate receptor function. Mol Pharmacol 1992;41:1142-1148.
54. Zhou M. & Baudry M. Developmental Changes in NMDA Neurotoxicity Reflect Developmental Changes in Subunit Composition of NMDA Receptors. J. Neurosci. 2006;26:2956-2963.
55. Kalluri HSG, Mehta AK & Ticku MK. Up-regulation of NMDA receptor subunits in rat brain following chronic ethanol treatment. Molecular Brain Research 1998;58:221-224.
56. Chandler LJ, Dean N & Greg S. Chronic Ethanol Upregulates NMDA and AMPA, but Not Kainate Receptor Subunit Proteins in Rat Primary Cortical Cultures. Alcoholism: Clinical and Experimental Research 1999;23: 363-370.
57. Henniger MSH, Wotjak CT & Hölter SM. Long-term voluntary ethanol drinking increases expression of NMDA receptor 2B subunits in rat frontal cortex. European Journal of Pharmacology 2003;470:33.
58. Qiang M, Denny AD & Ticku MK. Chronic Intermittent Ethanol Treatment Selectively Alters N-Methyl-D-aspartate Receptor Subunit Surface Expression in Cultured Cortical Neurons. Mol Pharmacol 2007;72: 95-102.
59. Nelson TE, Ur CL & Gruol DL. Chronic intermittent ethanol exposure enhances NMDA-receptor-mediated synaptic responses and NMDA receptor expression in hippocampal CA1 region. Brain Research 2005;1048: 69.
60. Roberto M, Bajo M, Crawford E, Madamba SG & Siggins GR. Chronic Ethanol Exposure and Pro-

tracted Abstinence Alter NMDA Receptors in Central Amygdala. Neuropsychopharmacology 2005;31:988.

61. Floyd DW, Jung KY. & McCool BA. Chronic Ethanol Ingestion Facilitates N-Methyl-D-aspartate Receptor Function and Expression in Rat Lateral/Basolateral Amygdala Neurons. J Pharmacol Exp Ther 2003;307: 1020-1029.
62. Ortiz J, Fitzgerald LW, Charlton M, Lane S, Trevisan L, Guitart X, Shoemaker W, Duman RS, Nestler EJ. Biochemical actions of chronic ethanol exposure in the mesolimbic dopamine system. Synapse 1995;21:289-298.
63. Grace AA, Floresco SB, Goto Y & Lodge DJ. Regulation of firing of dopaminergic neurons and control of goal-directed behaviors. Trends in Neurosciences 2007;30:220.
64. Carlezon Jr, W. A. & Thomas MJ. Biological substrates of reward and aversion: A nucleus accumbens activity hypothesis. Neuropharmacology 2009;56:122.
65. Gonzales RA, Job MO & Doyon WM. The role of mesolimbic dopamine in the development and maintenance of ethanol reinforcement. Pharmacology & Therapeutics 2004;103:121.
66. Roberto M, Schweitzer P, Madamba SG, Stouffer DG, Parsons LH, Siggins GR. Acute and Chronic Ethanol Alter Glutamatergic Transmission in Rat Central Amygdala: an In Vitro and In Vivo Analysis. J. Neurosci. 2004;24:1594-1603.
67. Pawlak R, Melchor JP, Matys T, Skrzypiec AE & Strickland S. Ethanol-withdrawal seizures are controlled by tissue plasminogen activator via modulation of NR2B-containing NMDA receptors. Proceedings of the National Academy of Sciences of the United States of America 2005;102:443-448.
68. Cebers G, Hou Y-N, Cebere A, Terenius L & Liljequist S. Chronic ethanol enhances NMDA-induced AP-1 activity in cultured rat cerebellar granule cells. NeuroReport 1996;8:217-220.
69. Qiang M, Rani CSS & Ticku MK. Neuron-Restrictive Silencer Factor Regulates the N-Methyl-D-aspartate Receptor 2B Subunit Gene in Basal and Ethanol-Induced Gene Expression in Fetal Cortical Neurons. Mol Pharmacol 2005;67:2115-2125.
70. Karin M, Liu Z-G & Zandi E. AP-1 function and regulation. Current Opinion in Cell Biology1997;9:240.
71. Biala G & Kotlinska J. Blockade of the acquisition of ethanol-induced conditioned place preference by N-methyl-D-aspartate receptor antagonists. Alcohol Alcohol. 1999;34:175-182.
72. Boyce-Rustay JM. & Cunningham CL. The Role of NMDA Receptor Binding Sites in Ethanol Place Conditioning. Behavioral Neuroscience 2004;118:822.
73. Boyce-Rustay J. & Holmes A. Ethanol-related behaviors in mice lacking the NMDA receptor NR2A subunit. Psychopharmacology 2006;187:455.
74. Kiefer F, Jahn H, Koester A, Montkowski A, Reinscheid RK, Wiedemann K. Involvement of NMDA receptors in alcohol-mediated behavior: mice with reduced affinity of the NMDA R1 glycine binding site display an attenuated sensitivity to ethanol. Biological Psychiatry 2003;53:345.
75. Gass JT & Olive MF. Glutamatergic substrates of drug addiction and alcoholism. Biochemical Pharmacology 2008;75:218-265.
76. McMillen BA, Joyner PW, Parmar CA, Tyer WE & Williams HL. Effects of NMDA glutamate receptor antagonist drugs on the volitional consumption of ethanol by a genetic drinking rat. Brain Research Bulletin 2004;64:279.
77. Holter SM, Danysz W & Spanagel R. Novel Uncompetitive N-Methyl-D-Aspartate (NMDA)-Receptor Antagonist MRZ 2/579 Suppresses Ethanol Intake in Long-Term Ethanol-Experienced Rats and Generalizes to Ethanol Cue in Drug Discrimination Procedure. J Pharmacol Exp Ther 2000;292:545-552.
78. Rassnick S, Pulvirenti L & Koob G. Oral ethanol self-administration in rats is reduced by the administration of dopamine and glutamate receptor antagonists into the nucleus accumbens. Psychopharmacology 1992;109:92.
79. Gremel CM. & Cunningham CL. Involvement of Amygdala Dopamine and Nucleus Accumbens NMDA Receptors in Ethanol-Seeking Behavior in Mice. Neuropsychopharmacology 2008;34:1443.
80. von der Goltz C, Vengeliene V, Bilbao A, Perreau-Lenz S, Pawlak CR, Kiefer F, Spanagel R. Cue-induced alcohol-seeking behaviour is reduced by disrupting the reconsolidation of alcohol-related memories. Psychopharmacology 2009;205:389.
81. Krystal JH, Petrakis IL, Limoncelli D, Webb E, Gueorgueva R, D’Souza DC, Boutros NN, Trevisan L, Charney DS. Altered NMDA Glutamate Receptor Antagonist Response in Recovering Ethanol-Dependent Patients. Neuropsychopharmacology 2003;28:2020.
82. Petrakis IL, Limoncelli D, Gueorguieva R, Jatlow P, Boutros NN, Trevisan L, Gelernter J, Krystal JH. Altered NMDA Glutamate Receptor Antagonist Response in Individuals With a Family Vulnerability to Alcoholism. Am J Psychiatry 2004;161:1776-1782.
83. Tsai GE, Ragan P, Chang R, Chen S, Linnoila VM,

Coyle JT. Increased Glutamatergic Neurotransmission and Oxidative Stress After Alcohol Withdrawal. Am J Psychiatry 1998;155:726-732.
84. Krupitsky EM, Neznanova O, Masalov D, Burakov AM, Didenko T, Romanova T, Tsoy M, Bespalov A, Slavina TY, Grinenko AA, Petrakis IL, Pittman B, Gueorguieva R, Zvartau EE, Krystal JH. Effect of Memantine on Cue-Induced Alcohol Craving in Recovering Alcohol-Dependent Patients. Am J Psychiatry 2007;164:519-523.
85. Nagy J. Alcohol Related Changes in Regulation of NMDA Receptor Functions. Curr Neuropharmacol 2008;6:39-54.