

Research In Brief

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Regulation of Plasticity of Glutamate Synapses by Endocannabinoids and the Cyclic-AMP/Protein Kinase A Pathway in Midbrain Dopamine Neurons

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Dopamine neurons in the ventral tegmental area (VTA) of the brain play an important role in the regulation of reward-related learning and drug addiction. Most, if not all, addictive drugs induce a long-term modification of glutamate synaptic transmission to VTA dopamine (DA) neurons, a critical cellular mechanism of drug addiction. Recent evidence shows that endocannabinoid (eCB) signaling modulates glutamate transmission. This process is altered by exposure to drugs of abuse, which may be an important brain mechanism mediating addictive behaviors. In the present study, Drs. Samir Haj-Dahmane and Roh-Yu Shen identify a previously unsuspected role for eCB signaling in modulating the long-term plasticity of glutamate synapse of VTA DA neurons. This discovery may lead to better understanding of the cellular mechanisms of addiction in the brain.

Findings

- Long-term depression (LTD) at glutamate synapses of VTA DA neurons may be induced by low frequency stimulation (LFS) and moderate postsynaptic membrane depolarization. The induction of LFS-LTD requires an increase in intracellular calcium but not *N*-methyl-d-aspartate (NMDA) receptor activation.
- LTD induced by LFS is accompanied by a decrease in the probability of glutamate release.
- LTD induced by LFS requires retrograde eCB signaling and the activation of presynaptic CB₁ receptors (eCB-LTD).
- Inhibition of 2-arachidonoylglycerol biosynthesis abolishes eCB-LTD, strongly indicating that 2-arachidonoylglycerol is the retrograde eCB messenger responsible for eCB-LTD.

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Predicting Post-Treatment-Initiation Alcohol Use Among Patients with Severe Mental Illness and Alcohol Use Disorders

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Few researchers studying alcohol abuse among individuals with a severe mental illness have examined predictors of alcohol outcomes following substance abuse treatment. In this study, the above research team considered the relationship between psychosocial factors and initiation of drinking following treatment.

Findings

- The type of pretreatment residential setting was directly related to treatment, with participants who lived in supervised settings reporting significantly more days of treatment.
- The extent of psychiatric symptoms was also directly related to treatment, with participants reporting more psychiatric symptoms also reporting significantly fewer days of treatment.
- The number of days that participants attended treatment was indirectly associated with alcohol outcomes and was impacted by use of alcohol-specific coping skills (i.e., positive thinking, avoidance, seeking social support). More frequent use of alcohol-specific coping skills was associated with less initiation of drinking following treatment.
- Following treatment, many people who abuse alcohol and have a severe mental illness are able to stop using alcohol, offering a favorable prognosis for people with a dual diagnosis.
- Psychosocial interventions that improve alcohol-specific coping skills are particularly important for this population.

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Regulation of Plasticity (cont'd)

- Inhibition of the presynaptic cAMP/PKA pathway is required for reduced glutamate release and the expression of eCB-LTD.

Background

Endocannabinoids (eCBs) are lipid-signaling molecules which play a key role in the regulation of synaptic transmission and plasticity in the central nervous system. The eCB system is comprised of the endogenous eCBs, which are cannabis-like compounds and their receptors. The psychomotor effects of cannabis and eCBs are heavily mediated by the CB₁ receptors, which are extensively expressed. The synthesis and release of eCBs is initiated by the excitation of neurons (Di Marzo *et al.*, 1994), or the activation of G $\alpha_{q/11}$ -coupled receptors (Haj-Dahmane & Shen, 2005). Post-synaptically released eCBs (Anandamide and 2-arachidonoylglycerol) function as retrograde messengers of synapses and mediate both short- and long-term modulation of presynaptic neurotransmitter release (Wilson & Nicoll, 2001). Current evidence also shows that the eCB system is involved in addictive behaviors and some eCB receptor ligands might be effective in treating addiction.

Dopamine (DA) neurons in the VTA are the origin of the mesocortical/mesolimbic DA systems, brain circuits involved in regulation of motivation and reward-related learning. The activity of these neurons, which is believed to encode the salient aspect of reward (Schultz *et al.*, 1997), including the rewarding properties of drugs of abuse, is under the control of excitatory (glutamatergic) inputs arising from both cortical and subcortical areas. The long-term changes in the function of glutamatergic synapses on VTA DA neurons are thought to be an important cellular mechanism for addiction.

The glutamatergic synapses on VTA DA neurons are profoundly modulated by the eCB system. Previous studies have reported that eCBs are released 'on demand' in the VTA area. However, their role in modulating the long-term plasticity of glutamate synapses of VTA dopamine (DA) neurons remains unknown.

Results

Given that long-term change in the glutamate synapses on VTA DA neurons is a critical brain mechanism for addiction and the role of eCBs in modulating glutamate synapses and treating addiction, researchers investigated a possible role of eCB signaling in the long-term plasticity of glutamate synapses onto VTA DA neurons.

In the present study, LFS paired with moderate postsynaptic depolarization was shown to elicit a NMDA receptor-independent LTD at glutamate synapses of VTA DA neurons. This form of LTD was caused by a decrease in the probability of glutamate release. Examination of the mechanisms underlying this form of LTD revealed that it was mediated by retrograde eCB signaling. In addition,

inhibition of 2-arachidonoyl glycerol biosynthesis blocked LTD induction, suggesting that 2-arachidonoyl glycerol is the most likely retrograde eCB messenger mediating LTD. The eCB-LTD induced at glutamate synapses of VTA DA neurons also required the inhibition of the presynaptic cAMP/PKA pathway. Taken together, these results reveal a critical role and mechanisms of eCBs in controlling the long-term plasticity of glutamate synapses on VTA DA neurons.

Discussion

The results from this study provide new understanding regarding how long-term plasticity of glutamate synapses in VTA DA neurons are modulated. These findings also help to integrate the current understanding on eCB signaling and brain dopamine function.

Results show that the LFS-LTD induction requires increased intracellular calcium and is presynaptically expressed as reduced glutamate release via the inhibition of the presynaptic cAMP/PKA pathway. This conclusion is different from previous studies which posit that the calcium-dependent LFS-LTD is mediated by a postsynaptic mechanism and expressed as a decrease in AMPA receptor function/number.

Importantly, LFS-LTD appears to be mediated by retrograde eCB signaling. This is demonstrated in a series of pharmacological studies using CB₁ receptor ligands. Such a conclusion is consistent with ample previous research evidence showing that eCBs act as retrograde messengers at glutamate synapses in several brain areas. The involvement of eCBs in mediating LFS-induced LTD in the VTA is also in agreement with prior studies showing that eCB signaling plays a ubiquitous role in mediating presynaptic LTD at both the excitatory and inhibitory synapses in the brain. Finally, these results are the first demonstration of eCB-mediated long-term synaptic plasticity in VTA DA neurons.

Also similar to previous studies in other brain areas, this research demonstrated that LFS-LTD in VTA DA neurons does not require the activation of group I mGluRs and therefore is mechanistically different from the mGluR-LTD previously characterized in VTA DA neurons during development which is expressed by a switch of AMPA receptor subunits composition (Bellone & Lüscher, 2005).

Importantly, because increased glutamatergic synaptic transmission in VTA DA neurons has been proposed to be a critical cellular mechanism for addiction, it is possible that by reducing the strength of glutamate synapses, eCB signaling in VTA DA neurons may influence addiction-related behaviors and drugs of abuse-induced long-term functional alteration of the mesocortical/mesolimbic DA systems. This notion is consistent with the behavioral observation that disruption of eCB signaling by genetic deletion or pharmacological blockade

Predicting Post-Treatment-Initiation Alcohol Use (cont'd)

Background

Predictors of alcohol use outcomes were examined in 278 men (46%) and women (54%) diagnosed with both severe mental illness (SMI) (i.e., a current schizophrenia-spectrum or bipolar disorder) and an alcohol use disorder (AUD). Participants for this study were identified at a community mental health center where they were seeking outpatient dual-diagnosis treatment for mental health and substance abuse issues.

Individuals were recruited during the first two weeks following treatment admission to participate in a six-month, naturalistic, longitudinal study examining predictors of relapse to alcohol and drug use. Participants were interviewed at monthly intervals for six months. In-person assessments were conducted at baseline and at two-, four-, and six-month follow-ups. The one-, three-, and five-month interviews were conducted by phone. The six-month follow-up rate was 81%.

The average age of participants was 40 years. They had completed, on average, 12 years of education. The majority were black (65%), followed by white (27%), Latino (4%), Native American (3%) and other (1%). Participants were predominantly single, unemployed and low income (supported on less than \$10,000 annually). Forty-one percent lived in supervised settings (e.g., group home, halfway house) and 59% in unsupervised settings (e.g. apartment, private home). On average, they had been previously treated eight times for psychiatric problems and 14 times for substance use problems.

Participants were found eligible for the study if they met diagnostic criteria for current SMI (past 30 days) and current AUD (past 12 months) and had either lived at their current address for at least six months or could provide contact information for two individuals who could be used to reach them during the study. The majority (97%) met criteria for current alcohol dependence, 3% for current alcohol abuse, and 76% for abuse of or dependence on cocaine, marijuana, opiates, sedatives or amphetamines. Overall, 86% of participants met criteria for at least one drug abuse or dependence diagnosis. Regarding comorbid mental health disorders, 12% of participants met criteria for schizophrenia, 56% for bipolar disorder and 32% for schizoaffective disorder.

The research team assessed areas of participants' functioning related to substance abuse, psychiatric symptoms, alcohol-specific coping skills, self-efficacy to remain abstinent and treatment utilization.

Results

Researchers examined initiation of drinking six months after treatment on the basis of participants' mental disorder diagnosis. These diagnoses included 1) a schizophrenia-spectrum disorder with no bipolar disorder, 2) a bipolar disorder

with no schizophrenia-spectrum disorder, or 3) schizoaffective disorder. No statistical differences were found among the three groups in initiation of drinking following treatment.

At the six month follow-up, 63% of the participants had good clinical outcomes (i.e., were completely abstinent or moderate drinkers with no/infrequent problems). Thirty-seven percent had poor clinical outcomes (i.e., engaged in heavy drinking or encountered problems or both).

Individuals who lived in supervised residential settings and those with fewer psychiatric symptoms reported attending significantly more treatment, compared to those in unsupervised settings and those with greater psychiatric symptoms.

Surprisingly, baseline alcohol use was not directly associated with subsequent treatment. In addition, neither baseline psychiatric symptoms nor treatment utilization were directly associated with alcohol use six months after treatment. In fact, participants with fewer initial psychiatric symptoms reported more use of treatment which, in turn, was related to more frequent coping.

Finally, more frequent use of coping skills and greater self-efficacy to remain abstinent were directly associated with less initiation of drinking following treatment. The effect of treatment on initiation of drinking after treatment was mediated by the use of positive coping skills but not by self-efficacy. It appears that participation in treatment resulted in better alcohol use outcomes because of the participants' use of coping skills.

Discussion/Clinical Implications

The high incidence of good clinical outcomes among participants (63%) highlights the prospects for good alcohol outcomes after treatment among individuals with dual diagnoses.

More treatment days resulted in better alcohol outcomes. Dual-diagnosis treatment clinics should provide interventions that increase engagement and retention of treated individuals with SMI-AUD.

Fewer psychiatric symptoms were associated with attending more treatment sessions, which was in turn, associated with better alcohol outcomes. This highlights the importance of treatment providers working to reduce psychiatric symptoms quickly, in order to reduce the risk of treatment dropout.

Alcohol-specific coping skills mediated the relationship between number of days of treatment and alcohol outcomes. It appears that the relationship of the number of days of treatment and positive alcohol outcomes was due, at least in part, to improvement in alcohol-related coping skills.

Regulation of Plasticity (cont'd)

of CB₁ receptors profoundly reduces the reward properties of nearly all drugs of abuse (Maldonado *et al.*, 2006). On the other hand, exposure to drugs of abuse modulates synaptic eCB signaling in the VTA and in the target areas of VTA DA neurons. Taken together, these studies along with the characterization of eCB-LTD suggest that eCB signaling in VTA DA neurons could be an important cellular mechanism for addiction.

Conclusion

The results from the present study firmly establish eCB-LTD at glutamate synapses as an important form of long-term synaptic plasticity in VTA DA neurons. Future studies examining the impact of selective disruption of eCB signalling in VTA DA neurons may help determine the role of eCB-LTD in addictive behaviors. These results contribute to the understanding of cellular molecular mechanisms mediating the potential therapeutic effects of eCB receptor ligands for addiction. This knowledge sheds additional light on the cellular mechanisms of addiction and may facilitate the development of better therapeutic agents to treat addiction.

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References

- Bellone, C., & Lüscher, C. (2005). mGluRs induce a long-term depression in the ventral tegmental area that involves a switch of the subunit composition of AMPA receptors. *European Journal of Neuroscience*, *21*, 1280-1288.
- Di Marzo, V., Fontana, A., Cadas, H., Schinelli, S., Cimino, G., Schwartz, J. C., & Piomelli, D. (1994). Formation and inactivation of endogenous cannabinoid anandamide in central neurons. *Nature*, *372*, 686-691.
- Haj-Dahmane, S., & Shen, R.-Y. (2005). The wake-promoting peptide orexin-B inhibits glutamatergic transmission to dorsal raphe nucleus serotonin neurons through retrograde endocannabinoid signaling. *Journal of Neuroscience*, *25*, 896-905.
- Maldonado, R., Valverde, O., & Berrendero, F. (2006). Involvement of the endocannabinoid system in drug addiction. *Trends in Neuroscience*, *29*, 225-232.
- Schultz, W., Dayan, P., & Montague, P. R. (1997). A neuronal substrate of prediction and reward. *Science*, *275*, 1593-1599.
- Wilson, R. I., & Nicoll, R. A. (2001). Endogenous cannabinoids mediate retrograde signaling at hippocampal synapses. *Nature*, *410*, 588-592.

Predicting Post-Treatment-Initiation Alcohol Use (cont'd)

Conclusion

The results of this study make a substantial contribution to understanding the influence of multiple factors on alcohol use outcomes. They emphasize the importance of psychosocial interventions with SMI individuals and particularly, those interventions that increase the use of alcohol-specific coping skills as a means of enhancing alcohol outcomes following treatment.

Further research is needed to identify the types of alcohol-specific coping skills that positively impact alcohol outcomes as well as to better understand the influence of the quality of treatment and psychiatric and addictions pharmacotherapy adherence on outcomes. In addition, more research is needed on the features of supervised residential settings for those with SMI-AUD that lead to better alcohol outcomes (e.g. requirements for abstinence or treatment, reduced access to alcohol and drugs, supportive peer group).

Study limitations include a reliance on self-report data which may have resulted in inflated correlations among study variables and a focus on relatively short-term (i.e., six months) as opposed to longer term (e.g., more than one year) outcomes.

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