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Optogenetic control of alcohol-seeking behavior via the dorsomedial striatal circuit

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HIGHLIGHTS

- Operant alcohol self-administration increases NMDAR activity of DMS dSPNs.
- Inhibition of DMS dSPNs reversibly suppresses alcohol-seeking behavior and relapse.
- *In vivo* low-to-moderate frequency stimulation of mPFC inputs causes a D2R-dependent increase in alcohol-seeking.
- The same mPFC stimulation with blockade of D1Rs and D2Rs persistently decreases alcohol-seeking.

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ABSTRACT

Alcohol consumption alters glutamatergic transmission in many brain regions, including the dorsomedial striatum (DMS); this aberrant plasticity is thought to be responsible for alcohol-seeking behavior. Recent studies reported that alcohol induced such plasticity specifically in direct pathway spiny projection neurons (dSPNs) of the DMS. However, it is unknown how this specific change contributes to alcohol-seeking behavior and relapse. Here, we first demonstrated that operant alcohol self-administration increased NMDA receptor activity in DMS dSPNs. Next, we found that optogenetic inhibition of dSPNs reversibly decreased operant lever presses for alcohol and alcohol intake. Furthermore, optogenetic stimulation of corticostriatal inputs at low and moderate frequencies induced reliable LTD in DMS slices. Surprisingly, *in vivo* delivery of the LTD-inducing protocol increased operant alcohol self-administration; this effect was blocked by a D2R antagonist. Importantly, LTD induction in the presence of both D1 and D2 receptor antagonists produced a long-lasting decrease in operant alcohol self-administration. Our results suggest that suppressing DMS dSPNs activity and their cortical inputs represents a novel treatment mechanism for alcohol use disorder.

1. Introduction

The dorsal striatum is heavily involved in the development of alcohol addiction (Corbit et al., 2012; Lovinger and Alvarez, 2017; Wang et al., 2007). The medial part of the dorsal striatum (DMS) mediates goal-directed behaviors (Yin and Knowlton, 2006), which are altered by alcohol consumption (Corbit et al., 2012). Additionally, alcohol intake potentiates DMS glutamatergic transmission, inhibition of which attenuates alcohol-seeking behaviors; this suggests that alcohol-induced plasticity drives alcohol-drinking behavior (Ma et al., 2017; Wang et al., 2010, 2012).

The principal DMS cells are GABAergic spiny projection neurons (SPNs). Direct pathway SPNs (dSPNs) express dopamine D1 receptors (D1Rs) and project to the substantial nigra pars reticula (SNr), whereas indirect pathway SPNs (iSPNs) contain D2Rs and innervate the external segment of the globus pallidus (Gerfen and Surmeier, 2011). Our recent study using the two-bottle choice drinking procedure found that dSPNs positively regulated alcohol consumption, while iSPNs negatively regulated this behavior (Cheng et al., 2017; Cheng and Wang, 2019). Importantly, the cortical inputs onto the DMS control goal-directed behaviors involved in drug-seeking and taking (Balleine and O'Doherty, 2010; Everitt and Robbins, 2013; Lovinger, 2010; Yin and Knowlton,

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2006), and these inputs are linked to drug and alcohol addiction (Everitt and Robbins, 2016; Ma et al., 2017; Nam et al., 2013; Volkow et al., 2006). We recently reported that alcohol consumption induced long-lasting increases in glutamatergic activity in DMS dSPNs (Cheng et al., 2017; Wang et al., 2015) and their input from the medial prefrontal cortex (mPFC) (Ma et al., 2017). It is not known, however, whether inhibition of DMS dSPNs or of mPFC inputs reduces alcohol-seeking behavior.

The present study examined whether alcohol-seeking behavior was reduced by optogenetic inhibition of DMS dSPNs *in vivo* or by optogenetic induction of long-term depression (LTD), an approach that has been shown to suppress drug-seeking behaviors (Lee et al., 2013; Ma et al., 2014). The purpose of our work is to produce a long-lasting decrease in alcohol self-administration. Our data showed that dSPN NMDAR activity was potentiated after operant alcohol self-administration, and that inhibition of DMS dSPNs decreased alcohol-seeking and reinstatement behaviors. Surprisingly, *in vivo* delivery of an optogenetic LTD-inducing protocol to the mPFC input to the DMS caused a D2R-dependent increase in alcohol self-administration, while LTD induction in the presence of both D1R and D2R antagonists produced a long-lasting decrease in alcohol-seeking behavior.

2. Material and methods

2.1. Reagents

Vectors encoding adeno-associated virus (AAV)-Cre-dependent halorhodopsin (eNpHR)-enhanced green fluorescent protein (eGFP) (5.4×10^{12} vg/mL) and AAV-Chronos-GFP (5.8×10^{12} vg/mL) were obtained from the UNC Vector Core (Chapel Hill, NC). AAV2.5-Cre (4.9×10^{12} vg/mL) was purchased from the Upenn Vector Core (State College, PA). APV was obtained from Tocris. Raclopride, SCH 23390, and picrotoxin were purchased from Sigma Aldrich (St. Louis, MO).

2.2. Animals and housing

Male Long-Evans rats were obtained from Harlan Laboratories at three months old and weighed between 300 and 400 g at the start of the experiments. A full timeline of all manipulations and behavioral tests are provided in Fig. 2A. Animals were pair-housed in a standard-sized conventional rat cage (143 in^2) in a humidity (~30%) and temperature-controlled (22 °C) vivarium with a 12-h light/dark cycle (lights on at 7:00 a.m.). Animals had *ad libitum* access to food and water throughout experiment. All animal procedures were approved by the Texas A&M Institutional Animal Care and Use Committee and were conducted in agreement with the Guide for the Care and Use of Laboratory Animals, National Research Council, 1996.

2.3. Operant self-administration of alcohol

Long-Evans rats were trained to self-administer a 20% alcohol (v/v) in water in an operant self-administration chamber as previously described (Huang et al., 2017; Ma et al., 2018). Each chamber contains two levers: pressing the active lever results in the delivery of 0.1 ml of the alcohol solution; pressing of the inactive lever are recorded, but no programmed events occur. The alcohol solution was delivered by a stainless steel dipper that rested within an alcohol reservoir. When activated, the dipper was raised into the magazine port for 10 s, then returned to the reservoir. The weight of the alcohol reservoir was measured before and after the operant session to determine how much alcohol a rat consumed during the operant session. During alcohol delivery, the magazine port was illuminated, which served as a cue to signal reward availability. After a 48-h exposure to 20% alcohol in the home cage, and one overnight session in a chamber where pressing the active lever delivered 0.1 ml water in a fixed ratio 1 (FR1), operant sessions were conducted 5 d per week for two weeks, where a lever press resulted in the delivery of 20% alcohol (FR1); over this time-period, the sessions shortened from 3 h to 30 min. Subsequently, operant sessions were run 3 d per week for one week and the schedule requirement was increased to FR3. After 1 month of training, surgery was conducted as described below. Alcohol seeking was measured using the number of lever presses and alcohol intake was determined by subtracting the weight of the alcohol reservoir after the 30-min operant session from the weight prior to the session as previously described (Huang et al., 2017; Ma et al., 2018).

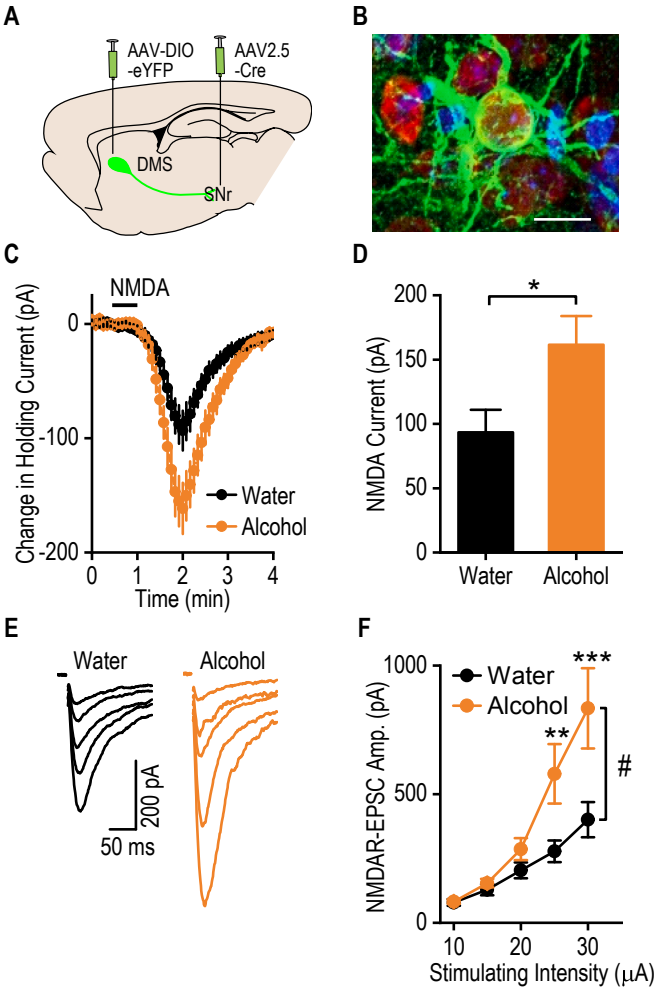

Fig. 1. Operant alcohol self-administration produces an increase in NMDAR activity of DMS dSPNs. A, Schematic of infusion of the indicated viral vectors into the SNr and DMS. Animals were then trained to self-administer alcohol for 8 weeks in operant chambers. **B**, Representative image of a fluorescently labeled dSPNs (green). The section was counter-stained with NeuroTrace Red (red) and DAPI (blue). Scale bar: 10 μm. **C-D**, NMDA-induced currents were significantly higher in the alcohol group than the age-matched control rats that drank water only and did not receive operant training. DMS slices were prepared and fluorescent neurons (dSPNs) were recorded 24 h after the last operant self-administration session. Changes in holding currents were measured after NMDA (10 μM, 30 s) was applied to the slices (C), and the peak NMDA current amplitudes were summarized for the indicated groups (D); $^{*}p < 0.05$, Mann-Whitney rank sum test; n = 12 neurons from 4 rats (Water) and 13 neurons from 4 rats (Alcohol). **E-F**, The NMDAR-EPSC amplitude in dSPNs was higher in the alcohol group than the age-matched water group. Representative NMDAR-EPSC traces evoked by a range of stimulation intensities in slices from the indicated groups (E), and input-output curves were compared (F); $^{\#}p < 0.05$; $^{**}p < 0.01$, $^{***}p < 0.001$ versus Water at the same stimulation intensity, two-way RM-ANOVA followed by SNK test; n = 13 neurons from 4 rats per group.

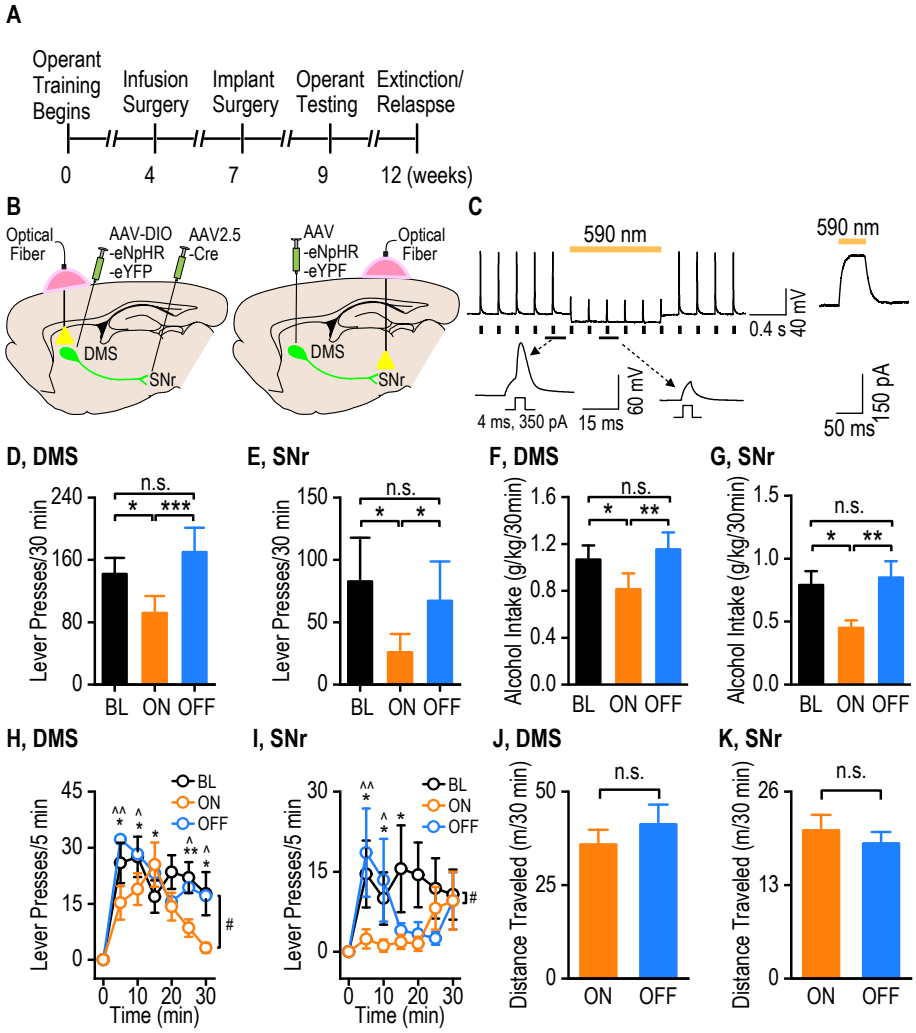

Fig. 2. Optogenetic inhibition of DMS dSPNs produces a reversible decrease in rat operant self-administration of alcohol. Animals were trained to self-administer alcohol in an operant setting, intracranially infused with the indicated viruses, and alcohol self-administration was measured with optical stimulation of dSPNs during operant sessions. **A**, Representation of the experimental protocol. **B**, Schematic representation of the infusion and fiber implant sites in the DMS (left) and SNr (right) of two separate groups of rats. **C,** A representative trace (left) showing the effects of pulsed current injection in DMS eNpHR-expressing neurons and of yellow laser (590 nm) stimulation. A sample trace on the right shows a hyperpolarization (voltage-clamp recording). **D-G,** Bar graphs depict the mean ± SEM of lever presses and alcohol intake (g/kg/30 min) during a 30-min session at baseline (BL), or during light stimulation (ON) of the DMS (D, F) or SNr (E, G), and 48 h after light stimulation (OFF). $^{*}p < 0.05$, $^{**}p < 0.01$, n.s. (not significant, $p > 0.05$); one-way RM ANOVA with SNK test; n = 13 (D, F) and 7 (E, G) rats. **H–I,** The profile of lever presses within the 30-min operant session under the indicated conditions (H, n = 13; I, n = 7); $^{\#}p < 0.05$ for BL versus ON and ON versus OFF; $^{*}p < 0.05$, $^{**}p < 0.01$ for ON versus BL at the same time; $\hat{p} < 0.05$, $\hat{\hat{p}} < 0.01$ for ON versus OFF at the same time. **J-K**, Bar graphs depict the mean ± SEM of distance traveled during a 30-min open field session with (ON) and without (OFF) light stimulation in the DMS (J) or SNr (K); n = 7 (J) and 5 (K) rats per group; n.s. (not significant, $p > 0.05$), paired *t*-test.

2.4. Surgery

2.4.1. Intracranial viral infusion

Intracranial Viral Infusion. Animals were anesthetized with isoflurane and mounted in a stereotaxic surgery frame. An incision was made, depending on the experiment, different small bilateral holes were drilled into the skull, and home-made injectors were lowered into the mPFC (3.2/2.6 mm AP, ± 0.65 mm ML, −4.0 mm DV; Figs. 4–6), DMS (1.2/0.36 mm AP, ± 1.8/2.3 mm ML, −4.6 mm DV), or SNr (−5.3 mm AP, ± 1.8 mm ML, −7.65 mm DV; Figs. 1 and 2). The home-made injectors were constructed with stainless steel tubing (McMaster-Carr, part # 8988K81, 0.01″ outer diameter, 0.005” inner diameter). The injector was inserted into plastic tubing that connected to a Hamilton syringe. We have used similar home-made injectors in our previous publications (Cheng et al., 2017; Huang et al., 2017; Ma et al., 2017, 2018; Lu et al., 2019). After the injector rested in the target place of the brain for 2 min, a Harvard Instruments micro-infusion pump delivered 1 µl viral vector via polyethylene tubing (PE20) connected to a 10-µl Hamilton syringe at a rate of 0.1 µl/min; the injector remained in place for 10 min following the infusion. After the infusions were completed, the incision was closed and the animals were monitored for one week or until they resumed normal activity. Meloxicam was administered post-operatively for pain management.

2.4.2. Intracranial implantation

Three weeks after the viral infusions, the animals were anesthetized with isoflurane and mounted in a stereotaxic frame. An incision was made, depending on the experiment, and bilateral optical fiber implants (300-nm core fiber secured to a 2.5-mm ceramic ferrule with a 5-mm fiber extending past the end of the ferrule) or bilateral guide cannulas (C252G-3.8; 21-gauge extending 4.5 mm past the pedestal; Plastics One) were lowered into the DMS (1.2 mm AP, ± 1.8 mm ML, −4.6 mm DV) or SNr (−5.3 mm AP, ± 1.8 mm ML, −7.65 mm DV). Implants were secured to the skull with metal screws and dental acrylic (Lang Dental). The incision was closed around the head cap and the skin was vet-bonded to the head cap. Meloxicam was administered post-operatively for pain management. Rats were monitored for one week or until they resumed normal activity.

2.5. Extinction and relapse of operant self-administration

At a stable baseline, extinction and reinstatement experiments were performed after optical stimulating during the operant sessions described below.

2.5.1. Extinction

During extinction sessions, animals were placed in the same chambers, with the same context. Lever press responses were recorded, but no alcohol was delivered, as previously described (Huang et al., 2017).

2.5.2. Alcohol-primed reinstatement

After a minimum of 9 extinction sessions, when the average lever presses across 3 consecutive sessions fell below 10, reinstatement was

conducted with and without continuous 5-mW laser stimulation for the session duration. Reinstatement was induced by the non-contingent delivery of 0.1 ml of 20% alcohol into the reward port at the start of the session, followed by the delivery of 0.1 ml of 20% alcohol after the first three presses of the active lever. This oral ethanol priming is not sufficient to produce a pharmacological effect (intake < 0.1 g/kg) (Wang et al., 2010). However, presentation of alcohol as the cue in this paradigm stimulates auditory (dipper activation sound), visual (magazine illumination), olfactory and gustatory (smell and taste of alcohol) senses. For the remainder of the 30-min session, subsequent presses of the active lever were recorded but no alcohol was delivered. Similar protocols have been used to induce reinstatement and studies have shown that following extinction, re-exposure to alcohol prior to testing reinstates alcohol-seeking behavior (Chiamulera et al., 1995; Huang et al., 2017; Wang et al., 2010). The experiment was conducted using a counterbalanced within-subject design, resulting in two tests per subject. Reinstatement tests were followed by 1 week of reacquisition of alcohol self-administration, before conducting another extinction session and reinstatement test.

2.6. Optogenetic stimulation and drug administration

2.6.1. dSPNs inhibition

Once a stable baseline had been attained after optical fiber implantation, operant sessions were conducted with 5-mW light stimulation of the DMS or SNr at 593.5 nm for the entire 30-min session, which has been employed in similar studies of operant alcohol self-administration (Seif et al., 2013).

2.6.2. LTD induction

Once a stable baseline had been attained after cannula implantation, the animals underwent optical LTD induction using low-frequency stimulation (LFS, 1 Hz for 10 min) or moderate-frequency stimulation (MFS; 12 Hz for 10 min). Each light pulse was 2 ms at 473 nm (5 mW). Thirty minutes later, operant sessions were conducted.

2.6.3. Drug administration

Rats were treated systemically with the D2R-antagonist, raclopride (0.01 mg/ml/kg), and/or the D1R-antagonist, SCH23390 (1 μg/ml/kg), 30 min before LTD induction. These doses were chosen because they did not change locomotion in open field testing or alter alcohol intake, when administered without LTD stimulation.

2.7. Open field test

As previously described (Cheng et al., 2017, 2018; Huang et al., 2017; Wei et al., 2018), the animals were placed in the center of an open field chamber and allowed to explore the apparatus for 30 min. The open field consisted of a square arena (16 in × 16 in) with 15 laser beam sensors on each x- and y-axis to track movement. The total distance traveled by each animal in 30 min was determined.

2.8. Electrophysiology

Long-Evans rats were anesthetized and perfused with an ice-cold cutting solution containing (in mM): 40 NaCl, 143.5 sucrose, 4 KCl, 1.25 NaH_2PO_4, 26 $NaHCO_3$, 0.5 $CaCl_2$, 7 $MgCl_2$, 10 glucose, 1 sodium ascorbate, and 3 sodium pyruvate; the solution was saturated with 95% O_2 and 5% CO_2. Coronal striatal slices (250 μm) were cut with a vibratome and incubated in a 1:1 mixture of cutting solution and external solution at 32 °C for 45 min. The external solution contained the following (in mM): 125 NaCl, 4.5 KCl, 2.0 $CaCl_2$, 1.0 $MgCl_2$, 1.25 NaH_2PO_4, 25 $NaHCO_3$, 15 sucrose, and 15 glucose, and was saturated with 95% O_2 and 5% CO_2. Slices were maintained in external solution at room temperature until use.

2.8.1. Whole-cell recordings

To measure NMDA-induced currents, NMDA (10 μM) was bath-applied for 30 s and holding currents were measured every 5 s (Cheng et al., 2017; Wang et al., 2010). NMDAR-EPSCs were recorded at −70 mV in the presence of 50 μM Mg^{2+}, 10 μM NBQX, and 100 μM picrotoxin in the bath solution, as previously described (Cheng et al., 2017; Wang et al., 2010). To measure AMPA-induced currents, AMPA (5 μM) was bath-applied for 15 s and holding currents were measured every 5s. To generate input-output curves for NMDAR-EPSCs, currents associated with increasing stimulation intensities were recorded. Raclopride (20 μM) was bath-applied and fEPSP/PS were compared before and after LFS.

2.8.2. Field potential recording

Extracellular field recordings were conducted using a patch pipette filled with 1 M NaCl and placed within the DMS as previously described (Wang et al., 2012). fEPSP/PS were evoked by optical stimulation through an objective lens at 0.05 Hz. Picrotoxin (100 μM) was bath-applied to block GABAergic transmission. Low to moderate frequency stimulation (1 Hz for 15 min or 12 Hz for 10 min) was applied to DMS slices to induce LTD.

2.9. Statistical analysis

Data were expressed as mean ± standard error of the mean (SEM). Data analyzed by one-way or two-way analysis of variance with repeated-measures (RM-AVOVA) or by *t*-test. Where significant ($p < 0.05$) ANOVA effects were detected, post-hoc Student-Newman-Keuls (SNK) were performed.

3. Results

3.1. Operant alcohol self-administration increases NMDAR activity in DMS dSPNs

Repeated cycles of alcohol consumption and withdrawal strengthened NMDAR activity of dSPNs in the DMS (Cheng et al., 2017; Wang et al., 2010). However, it is unclear whether such changes occur after operant alcohol self-administration. To measure NMDAR activity in dSPNs, we infused a retrograde Cre-expressing AAV (AAV2.5-Cre) (Ma et al., 2018; Walsh et al., 2014) into the SNr, and a Cre-dependent eYFP-expressing AAV (AAV-DIO-eYFP) into the DMS (Fig. 1A); this resulted in fluorescently labeled dSPNs (Fig. 1B, green). Rats were then trained to self-administer alcohol using the FR3 schedule (Huang et al., 2017; Ma et al., 2018). Twenty-four hours after the last alcohol-drinking session, whole-cell recording was conducted in DMS dSPNs of 4 rats which exhibited similar levels of lever presses and alcohol deliveries as those used in following behavioral tests. We found that the amplitude of NMDA-induced currents in dSPNs was dramatically greater in alcohol-exposed animals than in age-matched water controls, which did not receive operant training (Fig. 1C and D; $U = 32$, $p < 0.05$). Since the NMDA current was elicited by bath-application of NMDA, which activates both synaptic and extrasynaptic NMDARs (Carpenter-Hyland et al., 2004), the difference we observed may result from changes in synaptic and/or extrasynaptic NMDARs. To examine synaptic NMDAR activity, we measured the input-output relationship for NMDAR-EPSCs. As shown in Fig. 1E and F, EPSC amplitudes were significantly higher in dSPNs from alcohol-treated animals, as compared to age-matched water controls ($F_{(1,96)} = 5.41$, $p < 0.05$). These results suggest that operant self-administration of alcohol potentiates synaptic NMDAR activity in DMS dSPNs.

3.2. Optogenetic inhibition of DMS dSPNs transiently attenuates operant alcohol self-administration

The observed potentiation of NMDAR transmission is predicted to

excite DMS dSPNs and thus promote operant alcohol self-administration (Wang et al., 2010). We therefore examined whether selective optogenetic inhibition of DMS dSPNs decreased operant alcohol self-administration. Experimental protocol and timeline are illustrated in Fig. 2A. Two groups of rats (Fig. 2B) were trained to self-administer alcohol in an operant setting using the FR3 schedule. One group of rats was infused with AAV2.5-Cre into the SNr and AAV-DIO-eNpHR-eYFP into the DMS (Fig. 2B, left); a second group of animals was infused with an eNpHR-expressing virus (AAV-eNpHR-eYFP) into the DMS (Fig. 2B, right). The rats were then implanted with optical fibers into the DMS (first group, Fig. 2B, left) or the SNr (second group, Fig. 2B, right). With both of these approaches, dSPNs in the direct pathway of the basal ganglia were optically suppressed by yellow laser (590 nm) (Fig. 2C). Once lever presses for alcohol returned to baseline, alcohol intake and lever presses were tested 30 min after laser-stimulation in the DMS or SNr during the operant session. Optical inhibition of dSPNs (continuous 5-mW light) in the DMS significantly reduced lever presses and alcohol intake, relative to baseline [Fig 2D; $q = 3.90$, $p < 0.05$ and Fig. 2F; $q = 3.42$, $p < 0.05$]. When animals were tested again 48 h later without further light stimulation, they had returned to baseline behavior (Fig. 2D; $q = 2.19$, $p > 0.05$ and Fig. 2F; $q = 1.23$, $p > 0.05$). Optical inhibition of dSPNs axonal fibers (continuous 5-mW light) in the SNr produced the same effect as inhibition of dSPN cell bodies within the DMS. There were significant decreases in lever presses and alcohol intake when compared to baseline (Fig. 2E; $q = 4.70$, $p < 0.05$ and Fig. 2G; $q = 4.27$, $p < 0.05$). Again, the behavior returned to baseline within 48 h (Fig. 2E; $q = 1.29$, $p > 0.05$ and Fig. 2G; $q = 0.79$, $p > 0.05$).

To better understand the effects of dSPN inhibition, we compared the profiles of lever presses with and without optical inhibition. Optical inhibition of dSPNs in the DMS and of dSPN fibers in the SNr again significantly reduced lever presses, as compared to baseline (Fig. 2H; $F_{(2,125)} = 8.05$, $p < 0.05$ and Fig. 2I; $F_{(2,67)} = 4.73$, $p < 0.05$). Importantly, in both groups of eNpHR-expressing rats, locomotor activity was not altered by this level of light intensity in the DMS (Fig. 2J; $t_{(6)} = -1.11$, $p > 0.05$) or SNr (Fig. 2K; $t_{(4)} = 1.25$, $p > 0.05$). Together, these data suggest that dSPN activity drives operant alcohol-seeking behavior.

3.3. In vivo optogenetic inhibition of DMS dSPNs transiently attenuates reinstatement of alcohol-seeking behavior

Another devastating effect of alcoholism is the increased likelihood of relapse, even after a prolonged period of abstinence. We investigated whether dSPN inhibition altered the likelihood of relapse using extinction and alcohol prime-induced reinstatement in both groups of eNpHR-expressing animals. After 9 sessions of operant extinction (Huang et al., 2017; Wang et al., 2010) (Fig. 3A and B), an alcohol-primed operant reinstatement protocol was conducted with or without continuous optical inhibition of dSPNs. We found that without laser stimulation, the alcohol prime induced the expected and significant increase in lever presses, as compared to the extinction baseline (Fig. 3C; $q = 5.21$, $p < 0.01$ and Fig. 3D; $q = 4.95$, $p < 0.01$). Optical inhibition of dSPNs at their soma in the DMS significantly abolished this alcohol prime-induced increase in lever presses (Fig. 3C; $q = 0.06$, $p > 0.05$). Similarly, optical inhibition of dSPN terminals in the SNr during reinstatement abolished this alcohol prime-induced increase in lever presses (Fig. 3D; $q = 0.57$, $p > 0.05$). These data suggest that dSPN activity plays an important role in the reinstatement of alcohol consumption.

3.4. Optogenetic induction of corticostriatal LTD in the DMS is associated with an increase in operant self-administration of alcohol

Optogenetic LTD induction of corticostriatal inputs in the brain has been shown to reverse drug-induced glutamatergic plasticity and thus

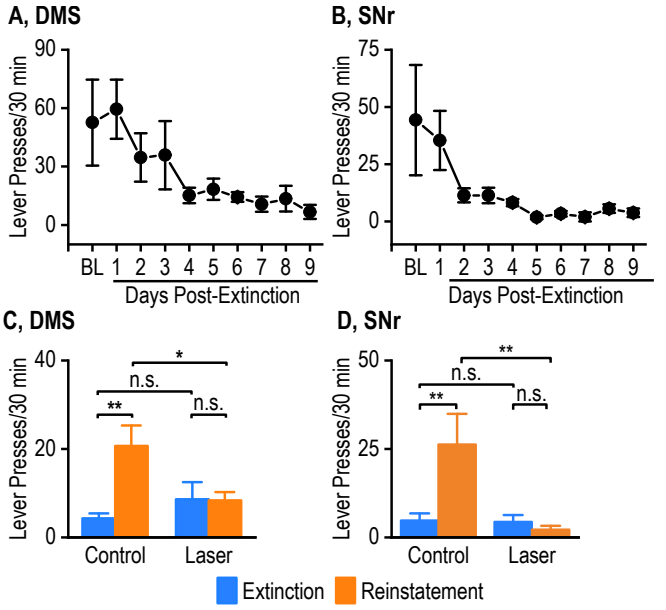

Fig. 3. *In vivo* optogenetic inhibition of DMS dSPNs prevents alcohol prime-induced reinstatement. After operant responding had been measured, the rats had 9 days of extinction operant sessions. An alcohol-primed reinstatement session was conducted on day 10 with (Laser) or without (Control) light stimulation. **A-B,** After operant responding had been measured, extinction operant sessions were conducted for 9 d. **A**, Line graph depicting the mean lever presses ± SEM over 9 d of extinction in animals after optogenetic stimulation in the DMS; n = 6 rats. **B,** Line graph depicting the mean lever presses ± SEM over 9 d of extinction in animals after optogenetic stimulation in the SNr; n = 5 rats. **C-D,** Bar graphs depict the mean lever presses ± SEM during extinction and alcohol-primed reinstatement, with the optical fiber implanted in the DMS (A) or SNr (B); *$p < 0.05$, **$p < 0.01$, n.s. (not significant, $p > 0.05$), two-way ANOVA followed by SNK test; n = 6 (DMS, Control), 5 (DMS, Laser), 4 (SNr, Control), and 5 (SNr, Laser).

persistently reduce addictive behaviors (Lee et al., 2013; Ma et al., 2014, 2018; Pascoli et al., 2012). To examine whether *in vivo* LTD induction changed operant alcohol self-administration, one group of animals was infused an AAV expressing a channelrhodopsin, Chronos, (AAV-Chronos-eGFP) into the rat mPFC; Chronos mediates greater photo currents than channelrhodopsin-2 (Klapoetke et al., 2014). DMS slices were prepared 8 weeks after the infusion. The mPFC input was stimulated by 470-nm light and this evoked field EPSP/population spikes (fEPSP/PS) (Huang et al., 2017). Optical low-frequency stimulation (LFS: 1 HZ for 15 min) of the mPFC input caused LTD of fEPSP/PS (Fig. 4A; 88.48 ± 3.07% of baseline and Fig. 4C, left; $t_{(8)} = 2.93$, $p < 0.05$) without altering the fiber volley amplitude, which represents presynaptic activity (Fig. 4B; 97.96 ± 2.71% of baseline and Fig. 4C right; $t_{(8)} = 0.28$, $p > 0.05$). Interestingly, optical moderate-frequency stimulation (MFS: 12 HZ for 10 min) of the mPFC input also caused robust LTD of fEPSP/PS (Fig. 4D; $t_{(7)} = 4.36$, $p < 0.01$) (Creed et al., 2015; Ronesi and Lovinger, 2005). These data suggest that optogenetic stimulation of the mPFC input at low-moderate frequencies produced reliable LTD in the DMS.

Another group of animals was trained to self-administer 20% alcohol in an operant setting; AAV-Chronos-eGFP was infused into the mPFC and optical fibers were implanted into the DMS. Once the animals returned to baseline after surgery, 900 pluses of 1-Hz light stimulation were delivered into the DMS to induce corticostriatal LTD, 30 min before the next operant session. Surprisingly, this resulted in an increase in lever presses and alcohol intake over baseline (Fig. 4E; $q = 8.57$, $p < 0.001$ and Fig. 4F; $q = 8.08$, $p < 0.001$), with a return to baseline within 2 days (Fig. 4E; $q = 1.23$, $p > 0.05$ and Fig. 4F; $q = 2.59$, $p > 0.05$). Using 12-Hz stimulation of the DMS also resulted

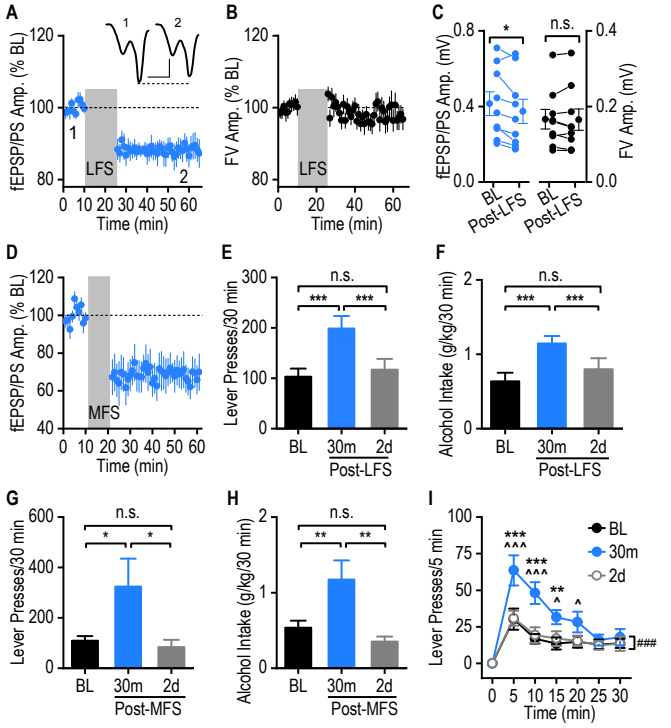

Fig. 4. In vivo delivery of corticostriatal LTD-inducing protocols to the DMS transiently potentiates operant self-administration of alcohol. Rats were infused with AAV-Chronos-eYFP into the mPFC and LTD was measured in DMS slices (A–D) or LTD-inducing protocols were delivered *in vivo* to test their effects on operant alcohol self-administration (E–I). **A,** fEPSP/PS was activated by light stimulation (473 nm) and LTD was induced by optical low frequency stimulation (LFS, 1 Hz for 15 min) of cortical inputs within the DMS. Scale bars: 3 ms, 0.2 mV; n = 9 slices from 6 rats. **B,** Optogenetic stimulation of cortical inputs in DMS slices did not alter fiber volley (FV). **C**, Scatter plots of optogenetically induced changes in fEPSP/PS and FV amplitudes; $*p < 0.05$; n.s., not significant, paired *t*-test. **D,** LTD was induced by optical moderate frequency stimulation (MFS, 12 Hz for 10 min) of cortical inputs within the DMS. A significant long-lasting depression (67.79 ± 7.38% of baseline) of fEPSP/PS was observed following optogenetic stimulation of DMS slices; n = 8 slices from 5 rats. **E-F**, Bar graphs (mean ± SEM) depicting lever presses (E) or alcohol intake (F) at baseline (BL), 30 min (30m), or 2 days (2d) after *in vivo* delivery of the 1-Hz LFS protocol. Light was delivered to the DMS once, 30 min before the testing session. n.s., not significant, $p > 0.05$; $***p < 0.001$; One-way RM ANOVA followed by SNK test. n = 12 rats/group. **G-H**, Bar graphs depicting the mean ± SEM of lever presses and alcohol intake during a 30-min operant session at BL, 30 m, and 2d after the 12-Hz MFS protocol. $*p < 0.05$; $**p < 0.05$; one-way RM ANOVA with SNK test. n = 7 rats/group. **I**, The profile of lever presses during 30-min operant sessions conducted at the indicated time-points after the 1-Hz LFS protocol; $^{\#\#\#}p < 0.001$ for BL versus 30 m and for 30 m versus 2d; $**p < 0.01$ and $***p < 0.001$ for 30m versus BL at the same time-point; $\hat{}p < 0.05$ and $\tilde{}p < 0.001$ for 30m versus 2d at the same time-point; two-way RM ANOVA followed by SNK test. n = 12 rats/group.

in similar significant increases in lever presses and alcohol intake (Fig. 4G; $q = 3.63$, $p < 0.05$ and Fig. 4H; $q = 4.65$, $p < 0.01$). Again, these effects of *in vivo* MFS were transient and returned to baseline at day 2 (Fig. 4G; $q = 0.46$, $p > 0.05$ and Fig. 4H; $q = 1.32$, $p > 0.05$).

We next examined the lever press profiles of each session conducted before and after optical LFS. Corticostriatal stimulation at 1 Hz consistently increased the lever presses observed 30 min post-stimulation (Fig 4I; $F_{(2,127)} = 21.62$, $p < 0.001$). These results showed that after receiving the LTD protocol, the animals pressed levers to obtain alcohol for 20 min of the 30-min session, instead of the normal pattern of frequent lever presses at the beginning of the session, followed by a decline over time. These results are unexpected and given our recent finding that inhibition of DMS iSPNs transiently increased alcohol intake (Cheng et al., 2017), this suggests that LTD induced by this protocol may preferentially suppress iSPNs, resulting in increased alcohol consumption.

3.5. D2Rs are required for optogenetic LTD-Mediated enhancement of alcohol self-administration

Since previous studies demonstrated that D2Rs contribute to LTD induction in dorsostriatal slices (Augustin et al., 2018; Kreitzer and Malenka, 2007), we next examined whether D2R inhibition altered the LFS-induced change in operant alcohol self-administration. Animals received an intraperitoneal injection of vehicle or the D2R antagonist, raclopride (0.01 mg/kg), 30 min before *in vivo* optical LFS. As expected, while LFS with vehicle pretreatment caused significant increases in lever presses (Fig. 5A; BL versus LFS + Veh, $q = 3.34$, $p < 0.05$) and alcohol intake (Fig. 5B; BL versus LFS + Veh, $q = 5.04$, $p < 0.01$), administration of raclopride blocked the LFS-induced increases in lever presses (Fig. 5A; BL versus LFS + Rac, $q = 0.75$, $p > 0.05$ and Fig. 5C; $F_{(1,12)} = 12.55$, $p < 0.05$) and alcohol intake (Fig. 5B; BL versus LFS + Rac, $q = 1.25$, $p > 0.05$). To confirm the D2R role in LTD induction, we bath-applied raclopride (20 μM) and measured mPFC-derived fEPSP/PS before and after LFS. We found that while suppressed immediately following LFS, the fEPSP/PS amplitude was gradually returned to the baseline level 30 min after LFS (Fig. 5D and E; 97.31 ± 1.44% of baseline for 31–40 min post-LFS, $t_{(4)} = 1.87$, $p > 0.05$) and that the fEPSP/PS amplitude was significantly greater in raclopride-treated slices than in controls without LFS (Fig. 5D and E; $t_{(11)} = -2.72$, $p < 0.05$). Additionally, raclopride treatment did not affect locomotion (Fig. 5F; $t_{(15)} = 0.16$, $p > 0.05$). Furthermore, raclopride treatment alone (without LFS induction) did not alter lever presses (Fig. 5G; $t_{(6)} = -0.04$, $p > 0.05$) or alcohol intake (Fig. 5H; $t_{(6)} = -0.77$, $p > 0.05$). These findings reinforce our idea that D2Rs mediated the LFS-induced increase in lever pressing for alcohol.

3.6. Pre-treatment with D1R- and D2R-Antagonists before optical LFS of mPFC inputs produces a long-lasting decrease in alcohol consumption

Given that D1R inhibition promoted LTD induction in dSPNs (Shen et al., 2008) and that dSPN inhibition suppressed alcohol-seeking behavior (Fig. 2), we next examined whether additional D1R blockade altered operant alcohol self-administration. We administered a cocktail of raclopride plus a D1R antagonist, SCH 23390, 30 min before LFS induction. Excitingly, we observed a significant decrease in lever presses and alcohol intake, and this effect lasted for 9 days after optical stimulation (Fig. 6A; $F_{(5,39)} = 5.24$, $p < 0.001$ and Fig. 6B; $F_{(5,29)} = 4.15$, $p < 0.01$). Furthermore, analysis of the profile of the lever presses 30 min post-LFS revealed that while rats treated with vehicle showed significantly more lever presses, as compared to baseline (Fig. 6C; $F_{(2,40)} = 11.19$, $p < 0.001$), those treated with D1R and D2R antagonists exhibited significantly fewer lever presses. Importantly, drug administration did not affect locomotion (Fig. 6D; $t_{(16)} = 1.26$, $p > 0.05$). Lastly, without LFS induction, the administration of raclopride and SCH 23390 did not alter the number of lever presses (Fig. 6E; $t_{(10)} = 0.76$, $p > 0.05$) or alcohol intake (Fig. 6F; $t_{(10)} = -0.12$, $p > 0.05$). We then explored how synaptic transmission changed following pretreatment with raclopride and SCH 23390 prior to LFS induction. One day 2, there was a marginally significant decrease in AMPA-induced currents between rats pretreated with raclopride and SCH 23390, as compared to those in rats pretreated with vehicle and LFS (Fig. 6G and H; $p = 0.07$), indicating that *in vivo* delivery of LFS with pretreatment of raclopride and SCH 23390 caused a long-lasting synaptic suppression in DMS. Taken together, these results suggest that a combination of *in vivo* DMS LFS stimulation and inhibition of both D1Rs and D2R provides a long-lasting decrease in alcohol consumption.

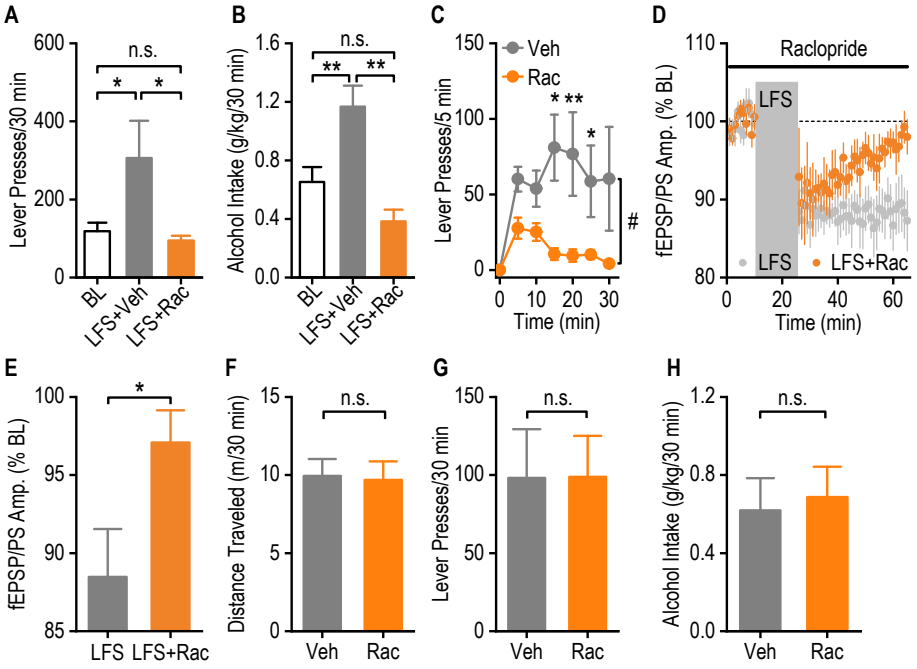

Fig. 5. D2R inhibition prevents optogenetic LTD-mediated enhancement of alcohol self-administration, Animals were systemically administered a D2R antagonist (raclopride [Rac], 0.01 mg/kg), or vehicle (Veh; saline), 30 min before LTD induction by light stimulation at 1 Hz for 15 min. The animals started a 30-min operant session 30 min after light stimulation. **A-B**, Bar graph depicting the mean ± SEM of alcohol lever presses or alcohol intake at baseline (BL; n = 7), in Veh-treated rats with LFS (n = 7), and in Rac-treated rats with LFS (n = 6); n.s. (not significant): $p > 0.05$, $^{*}p < 0.05$, $^{**}p < 0.01$; one-way RM ANOVA with SNK test. **C,** The profile of lever presses during the 30-min operant session for rats treated as indicated (Veh, n = 7; Rac, n = 6) before the 1-H z LFS protocol; $^{\#}p < 0.05$; $^{*}p < 0.05$, $^{**}p < 0.01$ for the comparison of Veh and Rac groups; two-way RM ANOVA with SNK test. **D-E**, D2R inhibition by raclopride suppressed LFS-induced LTD in DMS slices. Raclopride (20 μM) was bath-applied during the entire recording period. Time course (D) and bar graphs (E) show a greater amplitude (i.e., smaller LTD) of post-LFS fEPSP/PS in raclopride-treated slices than in controls. The control data are the same as Fig. 4A for comparison. n = 5 slices from 2 rats (LFS + Rac) and 9 slices from 6 rats (LFS). $^{*}p < 0.05$ by *t*-test. **F**, Bar graph of the distance traveled by animals in an open field test conducted 30 min after Veh (n = 8) or Rac (n = 9) injection. **G-H**, Bar graphs depicting the mean ± SEM of lever presses (G) and alcohol intake (H) during an operant session conducted 30 min after an intraperitoneal injection of raclopride (Rac) or vehicle (Veh). Paired *t*-test, n = 7 per group.

4. Discussion

4.1. Conclusion

In this study, we demonstrated that operant alcohol self-administration potentiated NMDAR activity in DMS dSPNs. We also found that optogenetic inhibition of this neuronal population reversibly inhibited alcohol-seeking behavior. Interestingly, *in vivo* delivery of an LTD-inducing protocol to the corticostriatal input within the DMS transiently increased alcohol seeking, which could be abolished by a D2R antagonist. Importantly, LTD induction in the presence of both D1R and D2R antagonists caused a persistent decrease in operant alcohol self-administration that lasted for 9 days. Thus, our findings may provide a new therapy for alcohol and/or drug use disorders.

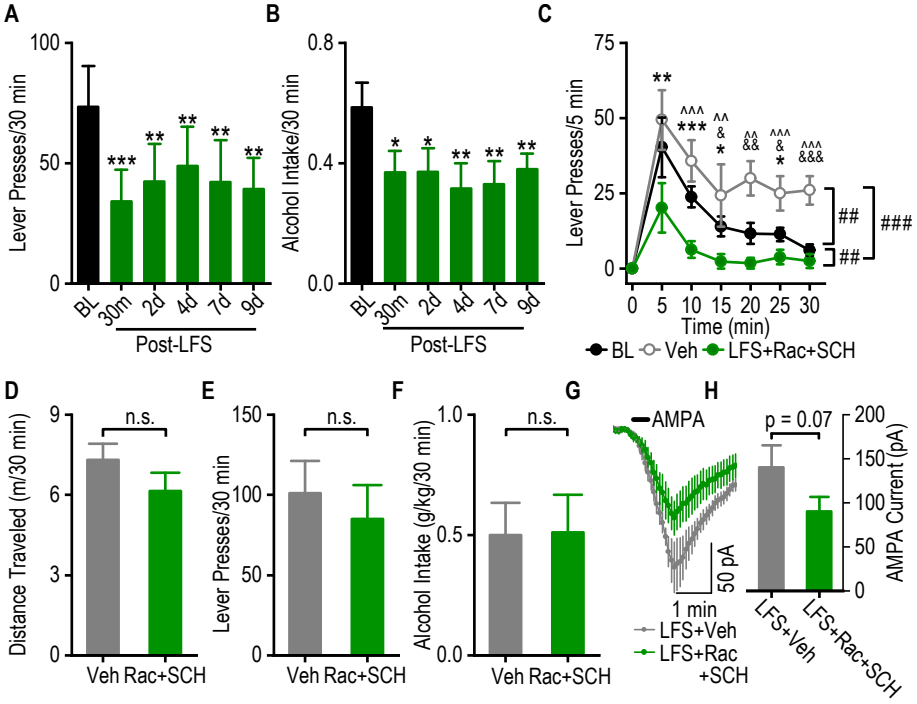

Fig. 6. Inhibition of both D1Rs and D2Rs during light stimulation of mPFC inputs produces a long-lasting decrease in operant self-administration of alcohol. Animals were systemically administered a cocktail of a D1R antagonist (SCH 23390 [SCH], 1 μg/kg) and Rac (0.01 mg/kg), or vehicle (Veh; saline), 30 min before receiving the 1-Hz LFS of mPFC inputs. **A-B,** Bar graphs of the lever presses or alcohol intake of rats before (BL) and after the treatment of LFS + Rac + SCH (green bars); $^{*}p < 0.05$, $^{**}p < 0.01$ and $^{***}p < 0.001$ versus BL, n = 9. **C**, The profile of lever presses at BL and 30 min after rats were treated with Veh or the cocktail. * or $^{\&}$ $p < 0.05$; $^{\#\#}$, **, $^{\&\&}$, or $^{\wedge\wedge}p < 0.01$; $^{\#\#\#}$, ***, $^{\&\&\&}$, or $^{\wedge\wedge\wedge}p < 0.001$; *: BL versus LFS + Rac + SCH; ^: Veh versus LFS + Rac + SCH; $^{\&}$: Veh versus BL. One-way (A, B) or two-way (C) RM ANOVA followed by SNK test. n = 9 for all groups. **D**, Bar graph of the distance traveled by animals in an open field test conducted 30 min after the indicated treatments (n = 9 for both groups). **E-F**, Bar graphs depicting the mean ± SEM of lever presses (E) and alcohol intake (F) during an operant session conducted 30 min after an intraperitoneal injection of Rac plus SCH23390 (Rac + SCH) or Veh; n = 11 rats per group; For D–F: n.s. (not significant, $p > 0.05$), paired *t*-test. **G-H**, Changes in holding currents were measured after AMPA (5 μM, 15 s) was applied to the slices (G), and the peak AMPA current amplitudes were calculated (H); $p = 0.07$, paired *t*-test. n = 11 neurons from 4 rats (LFS + Veh) and 9 neurons from 2 rats (LFS + Rac + SCH).

4.2. Contribution of dSPNs to drug-taking and relapse

The present study showed that operant alcohol self-administration, like the two-bottle choice drinking paradigm (Cheng et al., 2017), resulted in a robust increase in NMDAR activity in DMS dSPNs, as compared to water-drinking rats. Previous findings indicate that operant alcohol self-administration induced significantly higher NMDAR-mediated EPSCs than did operant sucrose self-administration, suggesting an effect of alcohol exposure, rather than that of the training procedure, on NMDAR activity (Ma et al., 2018). Given that NMDAR inhibition in the DMS reduced operant alcohol seeking and that DMS dSPNs inhibition reduced home-cage alcohol intake (Cheng et al., 2017; Wang et al., 2010), it is no surprise that optical inhibition of this neuronal population decreased operant alcohol self-administration in the current study. Previous studies have used intra-DMS infusion of a D1R-inhibitor, SCH23390, or chemogenetic inhibition of dSPNs to decrease alcohol intake during the two-bottle choice drinking paradigm (Cheng et al., 2017; Wang et al., 2015). In contrast to these previous findings, we were able to decrease operant alcohol seeking and intake by optical inhibition of dSPNs at either the cell soma or terminals. dSPN inhibition has also been shown to decrease cocaine-induced locomotor sensitization and self-administration (Ambroggi et al., 2009; Chandra et al., 2013).

Interestingly, animals that received optical stimulation of the cell soma exhibited drinking patterns that were similar to baseline up until the last 10 min of the session, during which they showed reduced lever pressing. However, optical stimulation of the cell terminals produced a different pattern consisting of fewer lever presses in the first 20 min, with a return to baseline levels during the last 10 min of the session. It is possible that delivery of the optical protocol to the cell terminals resulted in the inhibition of a larger number of cells, as compared to dSPN inhibition using cell soma stimulation, which is dependent on the retrograde transport of Cre. Additionally, prior studies indicate that optical somatic inhibition and terminal inhibition may vary depending on the local ionic composition of the cytosol (Wiegert et al., 2017). Another possibility is that inhibition of dSPN cell bodies in the DMS may also affect collateral transmission in the internal segment of globus pallidus (GPi). The GPi contains neurons that send excitatory projections to the lateral habenula, which can then drive inhibition dopamine neurons and produce aversion (Shabel et al., 2012). While optical inhibition of dSPN cell bodies in the DMS can activate multiple pathways, inhibition of dSPN axonal fibers in the SNr is more circuit-specific, which may account for the behavioral differences observed.

In addition to suppressing alcohol behaviors, our results show that inhibition of dSPNs blocked relapse. Several recent studies corroborate with this finding, indicating that systemic or intra-striatal administration of D1R antagonists reduced alcohol reinstatement (Chaudhri et al., 2009; Marchant and Kaganovsky, 2015; Sciascia et al., 2014). Unlike these previous studies, we were able to specifically inhibit dSPNs only during alcohol drinking and relapse sessions. Importantly, we were able to show that inhibition of dSPNs did not elicit any decrease in locomotion. These data show that activation of the dSPN circuit is sufficient to modify animal behaviors (Kravitz et al., 2012; Prast et al., 2014), implying that inhibition of this circuit should also be sufficient to reduce alcohol intake.

4.3. Contribution of plasticity to drug taking

While optogenetic/pharmacological dSPNs inhibition certainly did reduce alcohol-seeking and -taking, this change was only transient; a long-term behavioral change that can be elicited by one treatment session would be more appropriate for translation to a human therapy. In order to achieve long-term behavioral changes, we used low- and moderate-frequency stimulation-induced corticostriatal LTD. This demonstrated that LTD can be induced in the DMS by stimulation of corticostriatal synapses at 1 or 12 Hz. Several previous studies have utilized low-frequency stimulation-induced LTD to depotentiate drug-induced plasticity in the ventral striatum (Creed et al., 2015; Lee et al., 2013; Ma et al., 2014; Pascoli et al., 2012).

Although we were reliably able to induce LTD *ex* vivo, delivery of the corticostriatal LTD protocol *in vivo* resulted in a transient increase, rather than decrease, in lever pressing and alcohol intake. For the following two reasons, we believe that LFS induced LTD preferentially in iSPNs and this LTD drove the increases in alcohol-associated behavior. First, the increases were abolished by a D2R antagonist, which is required for LTD induction in iSPNs (Augustin and Lovinger, 2018; Kreitzer and Malenka, 2007; Shen et al., 2008; Surmeier et al., 2014). Second, our recent study found that chemogenetic inhibition of DMS iSPNs also transiently increased alcohol intake (Cheng et al., 2017). Importantly, when administering inhibitors of both D1Rs and D2Rs prior to delivery of the LTD protocol, we observed a long-lasting decrease in alcohol self-administration. This *in vivo* optogenetic and pharmacological stimulation caused a long-lasting depression of AMPA receptor activity. The current study reveals that induction of corticostriatal LTD produced a sustained decrease in alcohol-seeking and -taking behaviors.

The critical findings of this study show that selective inhibition of DMS dSPNs suppresses alcohol-seeking behavior and relapse, and that administration of D1Rs and D2Rs antagonist followed by LTD induction produces a long-lasting decrease in alcohol self-administration. These data suggest that a combination of D1R/D2R antagonists and *in vivo* DMS LFS stimulation has the potential for a novel treatment for alcohol and/or drug use disorders in humans. While optogenetics has not yet been used in the treatment of human alcohol consumption disorders, it provides an effective means of dissecting the mechanisms underlying this disease. Recent studies have demonstrated -that deep brain stimulation provides a viable tool to reduce alcohol use disorders in human patients (Luscher et al., 2015). Taken together with our present findings, novel deep brain stimulation protocols may be developed to produce long-lasting treatments for alcohol consumption disorders.

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Declarations of interest

The authors declare no conflict of interest.

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References

Ambroggi, F., Turiault, M., Milet, A., Deroche-Gamonet, V., Parnaudeau, S., Balado, E., Barik, J., van der Veen, R., Maroteaux, G., Lemberger, T., Schutz, G., Lazar, M., Marinelli, M., Piazza, P.V., Tronche, F., 2009. Stress and addiction: glucocorticoid receptor in dopaminoceptive neurons facilitates cocaine seeking. Nat. Neurosci. 12, 247–249.

Augustin, S.M., Chancey, J.H., Lovinger, D.M., 2018. Dual dopaminergic regulation of corticostriatal plasticity by cholinergic interneurons and indirect pathway medium spiny neurons. Cell Rep. 24, 2883–2893.

Augustin, S.M., Lovinger, D.M., 2018. Functional relevance of endocannabinoid-dependent synaptic plasticity in the central nervous system. ACS Chem. Neurosci. 9, 2146–2161.

Balleine, B.W., O'Doherty, J.P., 2010. Human and rodent homologies in action control: corticostriatal determinants of goal-directed and habitual action. Neuropsychopharmacology 35, 48–69.

Carpenter-Hyland, E.P., Woodward, J.J., Chandler, L.J., 2004. Chronic ethanol induces synaptic but not extrasynaptic targeting of NMDA receptors. J. Neurosci 24, 7859–7868.

Chandra, R., Lenz, J.D., Gancarz, A.M., Chaudhury, D., Schroeder, G.L., Han, M.H., Cheer,

J.F., Dietz, D.M., Lobo, M.K., 2013. Optogenetic inhibition of D1R containing nucleus accumbens neurons alters cocaine-mediated regulation of Tiam1. Front. Mol. Neurosci. 6, 13.
Chaudhri, N., Sahuque, L.L., Janak, P.H., 2009. Ethanol seeking triggered by environmental context is attenuated by blocking dopamine D1 receptors in the nucleus accumbens core and shell in rats. Psychopharmacology (Berlin) 207, 303–314.
Cheng, Y., Huang, C.C.Y., Ma, T., Wei, X., Wang, X., Lu, J., Wang, J., 2017. Distinct synaptic strengthening of the striatal direct and indirect pathways drives alcohol consumption. Biol. Psychiatry 81, 918–929.
Cheng, Y., Wang, J., 2019. The use of chemogenetic approaches in alcohol use disorder research and treatment. Alcohol 74, 39–45.
Cheng, Y., Wang, X., Wei, X., Xie, X., Melo, S., Miranda, R.C., Wang, J., 2018. Prenatal exposure to alcohol induces functional and structural plasticity in dopamine D1 receptor-expressing neurons of the dorsomedial striatum. Alcohol Clin. Exp. Res. 42, 1493–1502.
Chiamulera, C., Valerio, E., Tessari, M., 1995. Resumption of ethanol-seeking behaviour in rats. Behav. Pharmacol. 6, 32–39.
Corbit, L.H., Nie, H., Janak, P.H., 2012. Habitual alcohol seeking: time course and the contribution of subregions of the dorsal striatum. Biol. Psychiatry 72, 389–395.
Creed, M., Pascoli, V.J., Luscher, C., 2015. Addiction therapy. Refining deep brain stimulation to emulate optogenetic treatment of synaptic pathology. Science 347, 659–664.
Everitt, B.J., Robbins, T.W., 2013. From the ventral to the dorsal striatum: devolving views of their roles in drug addiction. Neurosci. Biobehav. Rev. 37, 1946–1954.
Everitt, B.J., Robbins, T.W., 2016. Drug addiction: updating actions to habits to compulsions ten years on. Annu. Rev. Psychol. 67, 23–50.
Gerfen, C.R., Surmeier, D.J., 2011. Modulation of striatal projection systems by dopamine. Annu. Rev. Neurosci. 34, 441–466.
Huang, C.C.Y., Ma, T., Roltsch Hellard, E.A., Wang, X., Selvamani, A., Lu, J., Sohrabji, F., Wang, J., 2017. Stroke triggers nigrostriatal plasticity and increases alcohol consumption in rats. Sci. Rep. 7, 2501.
Klapoetke, N.C., Murata, Y., Kim, S.S., Pulver, S.R., Birdsey-Benson, A., Cho, Y.K., Morimoto, T.K., Chuong, A.S., Carpenter, E.J., Tian, Z., Wang, J., Xie, Y., Yan, Z., Zhang, Y., Chow, B.Y., Surek, B., Melkonian, M., Jayaraman, V., Constantine-Paton, M., Wong, G.K., Boyden, E.S., 2014. Independent optical excitation of distinct neural populations. Nat. Methods 11, 338–346.
Kravitz, A.V., Tye, L.D., Kreitzer, A.C., 2012. Distinct roles for direct and indirect pathway striatal neurons in reinforcement. Nat. Neurosci. 15, 816–818.
Kreitzer, A.C., Malenka, R.C., 2007. Endocannabinoid-mediated rescue of striatal LTD and motor deficits in Parkinson's disease models. Nature 445, 643–647.
Lee, B.R., Ma, Y.Y., Huang, Y.H., Wang, X., Otaka, M., Ishikawa, M., Neumann, P.A., Graziane, N.M., Brown, T.E., Suska, A., Guo, C., Lobo, M.K., Sesack, S.R., Wolf, M.E., Nestler, E.J., Shaham, Y., Schluter, O.M., Dong, Y., 2013. Maturation of silent synapses in amygdala-accumbens projection contributes to incubation of cocaine craving. Nat. Neurosci. 16, 1644–1651.
Lovinger, D.M., 2010. Neurotransmitter roles in synaptic modulation, plasticity and learning in the dorsal striatum. Neuropharmacology 58, 951–961.
Lovinger, D.M., Alvarez, V.A., 2017. Alcohol and basal ganglia circuitry: animal models. Neuropharmacology 122, 46–55.
Lu, J., Cheng, Y., Wang, X., Woodson, K., Kemper, C., Disney, E., Wang, J., 2019. Alcohol intake enhances glutamatergic transmission from D2 receptor-expressing afferents onto D1 receptor-expressing medium spiny neurons in the dorsomedial striatum. Neuropsychopharmacology 44, 1123–1131.
Luscher, C., Pascoli, V., Creed, M., 2015. Optogenetic dissection of neural circuitry: from synaptic causalities to blue prints for novel treatments of behavioral diseases. Curr. Opin. Neurobiol. 35, 95–100.
Ma, T., Barbee, B., Wang, X., Wang, J., 2017. Alcohol induces input-specific aberrant synaptic plasticity in the rat dorsomedial striatum. Neuropharmacology 123, 46–54.
Ma, T., Cheng, Y., Roltsch Hellard, E., Wang, X., Lu, J., Gao, X., Huang, C.C.Y., Wei, X., Ji, J., Wang, J., 2018. Bidirectional and long-lasting control of alcohol-seeking behavior by corticostriatal LTP and LTD. Nat. Neurosci. 21, 373–383.
Ma, Y.Y., Lee, B.R., Wang, X., Guo, C., Liu, L., Cui, R., Lan, Y., Balcita-Pedicino, J.J., Wolf, M.E., Sesack, S.R., Shaham, Y., Schluter, O.M., Huang, Y.H., Dong, Y., 2014. Bidirectional modulation of incubation of cocaine craving by silent synapse-based remodeling of prefrontal cortex to accumbens projections. Neuron 83, 1453–1467.
Marchant, N.J., Kaganovsky, K., 2015. A critical role of nucleus accumbens dopamine D1-family receptors in renewal of alcohol seeking after punishment-imposed abstinence. Behav. Neurosci. 129, 281–291.
Nam, H.W., Hinton, D.J., Kang, N.Y., Kim, T., Lee, M.R., Oliveros, A., Adams, C., Ruby, C.L., Choi, D.S., 2013. Adenosine transporter ENT1 regulates the acquisition of goal-directed behavior and ethanol drinking through A2A receptor in the dorsomedial striatum. J. Neurosci. 33, 4329–4338.
Pascoli, V., Turiault, M., Luscher, C., 2012. Reversal of cocaine-evoked synaptic potentiation resets drug-induced adaptive behaviour. Nature 481, 71–75.
Prast, J.M., Schardl, A., Schwarzer, C., Dechant, G., Saria, A., Zernig, G., 2014. Reacquisition of cocaine conditioned place preference and its inhibition by previous social interaction preferentially affect D1-medium spiny neurons in the accumbens corridor. Front. Behav. Neurosci. 8, 317.
Ronesi, J., Lovinger, D.M., 2005. Induction of striatal long-term synaptic depression by moderate frequency activation of cortical afferents in rat. J. Physiol. 562, 245–256.
Sciascia, J.M., Mendoza, J., Chaudhri, N., 2014. Blocking dopamine D1-like receptors attenuates context-induced renewal of pavlovian-conditioned alcohol-seeking in rats. Alcohol Clin. Exp. Res. 38, 418–427.
Seif, T., Chang, S.J., Simms, J.A., Gibb, S.L., Dadgar, J., Chen, B.T., Harvey, B.K., Ron, D., Messing, R.O., Bonci, A., Hopf, F.W., 2013. Cortical activation of accumbens hyperpolarization-active NMDARs mediates aversion-resistant alcohol intake. Nat. Neurosci 16, 1094–1100.
Shabel, S.J., Proulx, C.D., Trias, A., Murphy, R.T., Malinow, R., 2012. Input to the lateral habenula from the basal ganglia is excitatory, aversive, and suppressed by serotonin. Neuron 74, 475–481.
Shen, W., Flajolet, M., Greengard, P., Surmeier, D.J., 2008. Dichotomous dopaminergic control of striatal synaptic plasticity. Science 321, 848–851.
Surmeier, D.J., Graves, S.M., Shen, W., 2014. Dopaminergic modulation of striatal networks in health and Parkinson's disease. Curr. Opin. Neurobiol. 29C, 109–117.
Volkow, N.D., Wang, G.J., Telang, F., Fowler, J.S., Logan, J., Childress, A.R., Jayne, M., Ma, Y., Wong, C., 2006. Cocaine cues and dopamine in dorsal striatum: mechanism of craving in cocaine addiction. J. Neurosci. 26, 6583–6588.
Walsh, J.J., Friedman, A.K., Sun, H., Heller, E.A., Ku, S.M., Juarez, B., Burnham, V.L., Mazei-Robison, M.S., Ferguson, D., Golden, S.A., Koo, J.W., Chaudhury, D., Christoffel, D.J., Pomeranz, L., Friedman, J.M., Russo, S.J., Nestler, E.J., Han, M.H., 2014. Stress and CRF gate neural activation of BDNF in the mesolimbic reward pathway. Nat. Neurosci. 17, 27–29.
Wang, J., Ben Hamida, S., Darcq, E., Zhu, W., Gibb, S.L., Lanfranco, M.F., Carnicella, S., Ron, D., 2012. Ethanol-mediated facilitation of AMPA receptor function in the dorsomedial striatum: implications for alcohol drinking behavior. J. Neurosci. 32, 15124–15132.
Wang, J., Carnicella, S., Phamluong, K., Jeanblanc, J., Ronesi, J.A., Chaudhri, N., Janak, P.H., Lovinger, D.M., Ron, D., 2007. Ethanol induces long-term facilitation of NR2B-NMDA receptor activity in the dorsal striatum: implications for alcohol drinking behavior. J. Neurosci. 27, 3593–3602.
Wang, J., Cheng, Y., Wang, X., Roltsch Hellard, E., Ma, T., Gil, H., Ben Hamida, S., Ron, D., 2015. Alcohol elicits functional and structural plasticity selectively in dopamine D1 receptor-expressing neurons of the dorsomedial striatum. J. Neurosci. 35, 11634–11643.
Wang, J., Lanfranco, M.F., Gibb, S.L., Yowell, Q.V., Carnicella, S., Ron, D., 2010. Long-lasting adaptations of the NR2B-containing NMDA receptors in the dorsomedial striatum play a crucial role in alcohol consumption and relapse. J. Neurosci. 30, 10187–10198.
Wei, X., Ma, T., Cheng, Y., Huang, C.C.Y., Wang, X., Lu, J., Wang, J., 2018. Dopamine D1 or D2 receptor-expressing neurons in the central nervous system. Addict. Biol. 23, 569–584.
Wiegert, J.S., Mahn, M., Prigge, M., Printz, Y., Yizhar, O., 2017. Silencing neurons: tools, applications, and experimental constraints. Neuron 95, 504–529.
Yin, H.H., Knowlton, B.J., 2006. The role of the basal ganglia in habit formation. Nat. Rev. Neurosci. 7, 464–476.