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Antagonism of α3β4 nicotinic receptors as a strategy to reduce opioid and stimulant self-administration

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Abstract

The *iboga* alkaloid ibogaine and the novel *iboga* alkaloid congener 18-methoxycoronaridine are putative anti-addictive agents. Using patch–clamp methodology, the actions of ibogaine and 18-methoxycoronaridine at various neurotransmitter receptor ion-channel subtypes were determined. Both ibogaine and 18-methoxycoronaridine were antagonists at α3β4 nicotinic receptors and both agents were more potent at this site than at α4β2 nicotinic receptors or at NMDA or 5-HT_3 receptors; 18-methoxycoronaridine was more selective in this regard than ibogaine. In studies of morphine and methamphetamine self-administration, the effects of low dose combinations of 18-methoxycoronaridine with mecamylamine or dextromethorphan and of mecamylamine with dextromethorphan were assessed. Mecamylamine and dextromethorphan have also been shown to be antagonists at α3β4 nicotinic receptors. All three drug combinations decreased both morphine and methamphetamine self-administration at doses that were ineffective if administered alone. The data are consistent with the hypothesis that antagonism at α3β4 receptors is a potential mechanism to modulate drug seeking behavior. 18-Methoxycoronaridine apparently has greater selectivity for this site than other agents and may be the first of a new class of synthetic agents acting via this novel mechanism to produce a broad spectrum of anti-addictive activity.

Keywords: 18-Methoxycoronaridine; Dextromethorphan; Mecamylamine; Morphine; Methamphetamine; Nicotine receptor; Drug addiction

1. Introduction

18-Methoxycoronaridine is a novel *iboga* alkaloid congener that is being studied as a potential treatment for multiple forms of drug abuse. In rats, 18-methoxycoronaridine (10–40 mg/kg) decreases the self-administration of morphine (Glick et al., 1996; Maisonneuve and Glick, 1999), cocaine (Glick et al., 1996), methamphetamine (Glick et al., 2000a), nicotine (Glick et al., 2000a) and ethanol (Rezvani et al., 1997) but does not affect responding for a non-drug reinforcer (water; Glick et al., 1996). Like its parent compound ibogaine (Glick and Maisonneuve, 1998), the precise mechanism of action of 18-methoxycoronaridine has remained elusive despite evidence that it modulates dopamine release in the nucleus accumbens (Glick et al., 1996) and binds, with low affinity, to several types of receptors (Glick and Maisonneuve, 2000; Glick et al., 2000b). In functional assays ($^{86}Rb^+$ efflux from KXα3β4R2 cells) conducted by Dr. Kenneth Kellar (Georgetown University) as part of the NIMH Psychoactive Drug Screening Program, 18-methoxycoronaridine was found to be an antagonist at α3β4 nicotinic receptors. However, this work did not establish whether 18-methoxycoronaridine's nicotinic antagonist action was specific to the α3β4 subtype or whether other nicotinic subtypes were also affected, for example, the α4β2 subtype that is most prevalent in the brain (e.g., Flores et al., 1992). Hence, as part of the present study, the actions of both 18-methoxycoronaridine and ibogaine at both α3β4 and α4β2 receptors were determined using patch–clamp methodology.

Other studies from the Kellar laboratory have shown that the antitussive dextromethorphan and its metabolite dextrorphan are also antagonists at α3β4 receptors (Hernandez et al., 2000), and we have recently found that both dextromethorphan and dextrorphan reduce morphine, methamphetamine and nicotine self-administration in rats (Glick et al., 2001). Although dextromethorphan and dextrorphan are also antagonists at NMDA glutamate receptors (Murray and Leid, 1984; Ebert et al., 1998), the relative potencies of dextromethorphan and dextrorphan in our drug self-admin-

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istration studies (Glick et al., 2001) were more consistent with actions at α3β4 receptors than at NMDA receptors. Furthermore, mecamylamine, a nonspecific nicotinic antagonist, has been reported to decrease craving for cocaine in humans (Reid et al., 1999) and to reduce cocaine self-administration in rats (Levin et al., 2000); and Papke et al. (2001) recently reported that mecamylamine has preferential affinity for α3β4 receptors versus other nicotinic subtypes (e.g., α4β2). All of these findings together suggest that antagonism of acetylcholine's actions at α3β4 nicotinic receptors may constitute an important mechanism for reducing the rewarding effects of multiple drugs; α3β4 receptors are localized in brain areas that are well suited to modulate mesolimbic activity, both directly (e.g., Klink et al., 2001) and indirectly (e.g., Nishikawa et al., 1986; Quick et al., 1999).

Totally selective antagonists of α3β4 receptors are unavailable, and hence it is difficult to directly test our hypothesis that α3β4 receptor antagonists will reduce drug self-administration. However, we reasoned that if two agents had the common action of blocking this site but also had other actions that were unique to each agent, the combination of low doses of such agents (doses of each agent that would be ineffective if administered alone) might produce additive effects at the α3β4 site and reduce drug self-administration without the involvement of other actions contributing to side effects. In the present study, as an initial test of this idea, the effects of three such combined treatments (18-methoxycoronaridine plus dextromethorphan, 18-methoxycoronaridine plus mecamylamine, and dextromethorphan plus mecamylamine) on morphine and methamphetamine self-administration were assessed.

2. Materials and methods

2.1. Receptor functional analyses

Human embryonic kidney 293 (HEK293) fibroblasts (ATCC CRL1573) were cultured in minimal essential medium supplemented with 10% fetal bovine serum and 2 mM glutamine (Life Technologies, Rockville, MD). Cells were plated on poly-D-lysine-coated 35 mm nunc dishes, transfected by the LipofectaminePLUS method (Life Technologies), and examined functionally between 18 and 48 h post transfection. The following receptor subunit cDNAs were used (accession no.): nAChR-α3 (nicotinic acetylcholine receptor-α3; L31621), nAChR-α4 (L31620), nAChR-β2 (L31622), nAChR-β4 (U42976), 5-HT_3R-A (5-HT_{3A} receptor; M74425), NR1 (*N*-methyl-D-aspartate receptor 1; X63255), NR2A (X91561), and NR2B (M91562). The nAchR and NR clones were rats; the 5-HT_3R-A clone was mouse. Co-transfection of enhanced green fluorescent protein (EGFP, 10% of total cDNA) provided a marker to identify transfected cells. Transfected cells were selected for EGFP expression and examined by voltage-clamp recording in the whole-cell configuration using an Axopatch 200B patch–clamp amplifier (Axon Instruments, Foster City, CA). Thin-walled borosilicate glass microelectrodes (TW150F, World Precision Instruments, Sarasota, FL) had resistances of 3–5 MΩ when filled with an internal solution containing (in mM): 135 CsCl, 10 CsF, 10 HEPES (*N*-[2-hydroxyethyl]piperazine-*N*′-[2-ethanesulfonic acid]), 5 EGTA (ethylene glycol-bis[β-aminoethyl ether]-*N*,*N*,*N*′,*N*′-tetraacetic acid), 1 $MgCl_2$, 0.5 $CaCl_2$, pH 7.2. Whole-cell capacitance and series resistance were recorded and adequately compensated using the available circuitry of the amplifier. Current responses were filtered at 1 kHz with an 8-pole Bessel filter (Cygnus Technologies, Delaware Water Gap, PA), digitized at 3 kHz, and stored on a Macintosh PowerPC-G3 computer using an ITC-16 interface (Instrutech, Great Neck, NY) under control of the data acquisition and analysis program Synapse (Synergy Research, Gaithersburg, MD). Cells were continuously superfused with extracellular solution containing (in mM): 150 NaCl, 3 KCl, 5 HEPES, 1 $MgCl_2$, 1.8 $CaCl_2$, 10 glucose, and 0.1 mg/ml phenol red, pH 7.3 ($MgCl_2$ was omitted from all solutions used for the study of NMDA receptors). Drug stocks (10 mM) were made up in DMSO (dimethylsulfoxide) and diluted in extracellular solution immediately prior to use; final concentration of DMSO was 0.2% or lower. Control, agonist, and drug solutions were applied to individual cells by rapid perfusion. Solutions were driven by a syringe pump through a flowpipe having four inputs that converge at a single common output of approximately 100 μm diameter. Rapid switching between inputs was achieved using a set of upstream solenoid valves (Lee, Westbrook, CT) under computer control; the solution exchange rate was ~ 5 ms as measured from liquid junction currents.

2.2. Chemicals used in vivo

18-Methoxycoronaridine hydrochloride (1–2 mg/kg; Albany Molecular Research, Albany, NY) was dissolved in phosphate buffer and injected intraperitoneally 15 min before behavioral testing. Dextromethorphan hydrobromide (5 mg/kg; Sigma/RBI, St. Louis, MO) was dissolved in saline and injected subcutaneously 20 min before testing. Mecamylamine hydrochloride (1 mg/kg; Sigma/RBI) was dissolved in physiological saline and injected intraperitoneally 30 min before testing. All rats received two injections (for rats that received a single drug, half of them also received the appropriate saline/vehicle injection corresponding to each of the other two drugs).

2.3. Animals

Naïve female Long–Evans derived rats (250 g; Charles River, NY) were maintained on a normal 12 h light cycle (lights on at 7:00 a.m., lights off at 7:00 p.m.). For all experiments the "Principles of laboratory animal care" (NIH Publication No. 85-23, revised 1985) were followed.

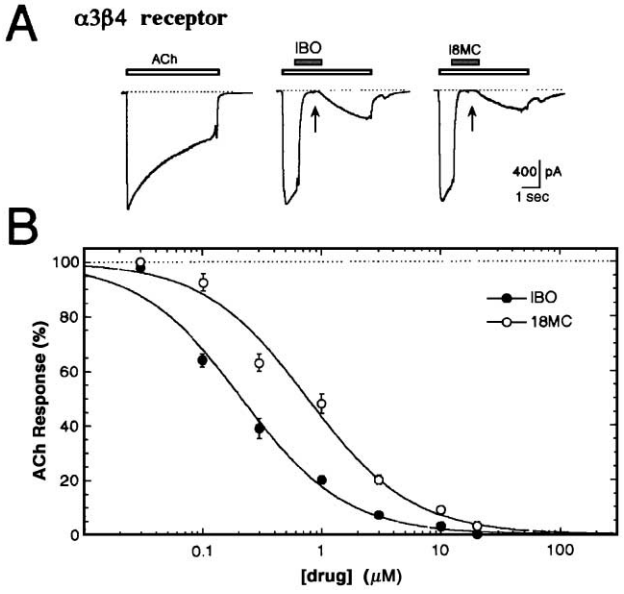

Fig. 1. Inhibition of α3β4 nicotinic receptors by ibogaine (IBO) and 18-methoxycoronaridine (18MC). Recombinant receptors were expressed in HEK293 cells and examined by whole-cell patch clamp recording with rapid application of agonist and drug solutions. (A) Whole-cell currents evoked by 1 mM ACh in transfected cells. Open bars depict the timing of ACh application. Filled bars depict the timing of co-application of 20 μM IBO or 20 μM 18MC. Inhibition was measured relative to control at the end of the drug application (arrows). (B) Inhibition of 1 mM ACh-evoked currents by various concentrations of IBO and 18MC. Data are mean ± SEM for 3–14 cells per point. Curve fits are given for the logistic equation: $I = I_{max}/(1+([drug]/IC_{50}))$. Best fitting IC_{50} values were 0.22 μM for IBO and 0.75 μM for 18MC.

2.4. Self-administration procedure

The intravenous self-administration procedure has been described previously (e.g., Glick et al., 1996, 2000a). Briefly, responses on either of two levers (mounted 15 cm apart on the front wall of each operant test cage) were recorded on an IBM compatible computer with a Med Associates interface. The intravenous self-administration system consisted of polyethylene-silicone cannulas constructed according to the design of Weeks (1972), Instech harnesses and swivels, and Harvard Apparatus infusion pumps (no. 55-2222). Shaping of the bar-press response was initially accomplished by training rats to bar-press for water. Cannulas were then implanted in the external jugular vein according to procedures described by Weeks (1972). Self-administration testing began with a 16-h nocturnal session followed by daily 1-h sessions, 5 days (Monday–Friday) a week. A lever-press response produced a 10-μl infusion of drug solution (0.01 mg of morphine sulfate) in about 0.2 s or a 50-μl infusion of drug solution (0.025 mg of methamphetamine sulfate) in about 1 s. Since all rats generally weighed 250 ± 20 g, each response delivered approximately 0.04 mg/kg of morphine or 0.1 mg/kg of methamphetamine. Experiments to assess the effects of experimental treatments were begun when baseline self-administration rates stabilized (< 10% variation from 1 day to the next across 5 days), usually after 2 weeks of testing. Each rat typically received two or three different treatments spaced at least 1 week apart. In order to provide an indication of the specificity of treatment effects on drug self-administration, all treatments were also administered to other rats bar-pressing for water (0.01 ml orally) on a comparable schedule (continuous reinforcement; 1-h sessions).

3. Results

3.1. Drug actions at neurotransmitter receptor ion-channels

Transfected HEK293 cells expressing various receptor subunit cDNAs were examined by whole-cell patch–clamp recording with fast perfusion of agonist and drug solutions. We began with α3β4 nicotinic acetylcholine receptors as these are the principal ganglionic nicotinic acetylcholine receptor subtype for which inhibition by ibogaine had previously been demonstrated (Badio et al., 1997; Mah et al., 1998; Fryer and Lukas, 1999). Transfected HEK293 cells expressing α3β4 nicotinic acetylcholine receptors were voltage-clamped to − 70 mV and stimulated with 1 mM acetylcholine at 30 s intervals. Acetylcholine alone evoked a large inward current not seen in untransfected cells. Application of 20 μM ibogaine or 20 μM 18-methoxycoronaridine alone did not produce any response. Co-application of either 20 μM ibogaine or 20 μM 18-methoxycoronaridine nearly abolished the ACh-evoked responses in all cells tested ($N = 15$) (Fig. 1A). The inhibition developed rapidly in the presence of acetylcholine and reversed more slowly following the removal of drug. The inhibition was concen-

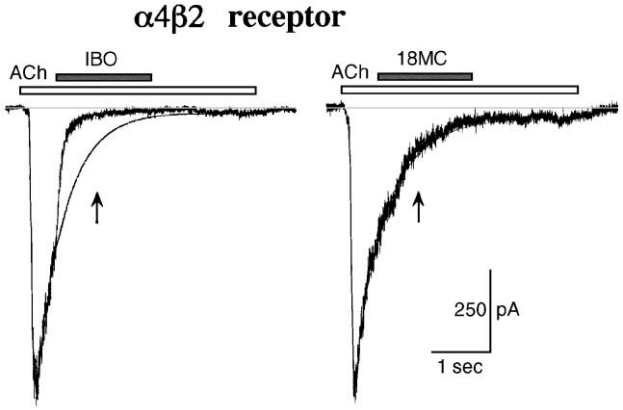

Fig. 2. Inhibition of α4β2 nicotinic receptors by ibogaine (IBO) but not by 18-methoxycoronaridine (18MC). Whole-cell currents in cells expressing recombinant α4β2 receptors were evoked by 300 μM ACh. Open bars depict the timing of ACh application. Filled bars depict the timing of co-application of 20 μM IBO or 20 μM 18MC. Because of the relatively rapid desensitization of the α4β2 response, inhibition was measured relative to control (superimposed curve fits) during drug application (arrows).

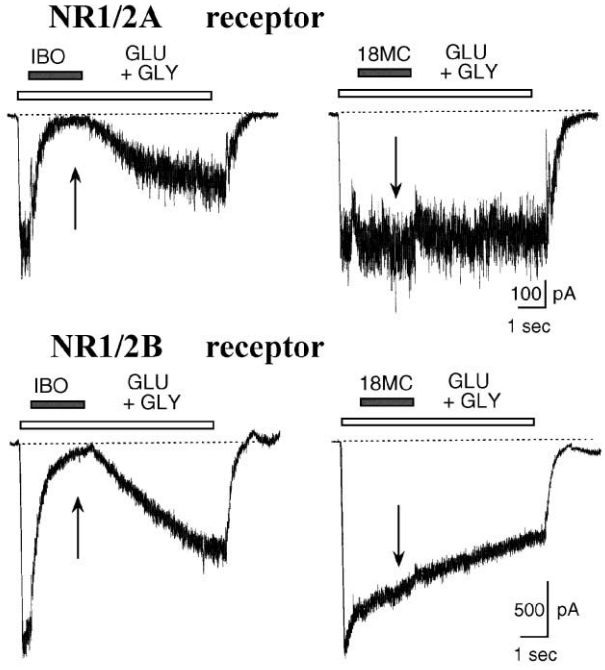

Fig. 3. Inhibition of NMDA receptors by ibogaine (IBO) but not by 18-methoxycoronaridine (18MC). Whole-cell currents in cells expressing recombinant NR1/2A and NR1/2B receptors were evoked by 100 μM glutamate plus 10 μM glycine. Open bars depict the timing of agonist application. Filled bars depict the timing of co-application of 20 μM or 20 μM 18MC. Inhibition was measured relative to control at the end of the drug application (arrows).

tration-dependent; IC_{50} values were 0.22 μM for ibogaine versus 0.75 μM for 18-methoxycoronaridine, and the concentration–response relationship had a Hill slope of unity, which is consistent with a single site of action (Fig. 1B). These data confirm previous reports of the actions of ibogaine and indicate that 18-methoxycoronaridine has similar actions at ganglionic nicotinic acetylcholine receptors. Results were somewhat different, however, at neuronal nicotinic acetylcholine receptors. In this case, transfected HEK293 cells expressing α4β2 nicotinic acetylcholine receptors were voltage-clamped to −70 mV and stimulated with 300 μM acetylcholine at 30-s intervals. Acetylcholine alone evoked an inward current whereas application of 20 μM ibogaine or 20 μM 18-methoxycoronaridine alone did not produce any response. Co-application of 5 μM ibogaine inhibited the ACh-evoked response by 61 ± 4% ($N=5$) whereas 5 μM 18-methoxycoronaridine produced no apparent inhibition ($N=6$). At higher drug concentrations (20 μM) (Fig. 2), ibogaine inhibition reached 93 ± 3% ($N=7$) versus only 8 ± 4% by 18-methoxycoronaridine ($N=6$). These data suggest the IC_{50} for ibogaine at α4β2 nicotinic acetylcholine receptors is on the order of 1–5 μM and that 18-methoxycoronaridine is considerably less potent ($IC_{50}>20$ μM) at this neuronal nicotinic acetylcholine receptor subtype.

Another known action of ibogaine involves the inhibition of NMDA-type glutamate receptors (Popik et al., 1995; Chen et al., 1996) presumably by interaction with the PCP/MK-801 (phencyclidine/dizocilpine) binding site (Sweetnam et al., 1995; Chen et al., 1996). Indeed, it has been suggested that the NMDA receptor-mediated actions of ibogaine may be central to its putative anti-addictive properties (Popik et al., 1995). To study these, transfected HEK293 cells expressing NR1/2A or NR1/2B receptors were voltage-clamped to −70 mV and stimulated with 100 μM glutamate plus 10 μM glycine at 30 s intervals. Agonist application evoked a large inward current not seen in untransfected cells. Application of 20 μM ibogaine or 20 μM 18-methoxycoronaridine alone did not produce any response. Co-application of 20 μM ibogaine reduced the agonist-evoked response of NR1/2A receptors by 98 ± 3% ($N=3$) and of NR1/2B receptors by 95 ± 2% ($N=3$); inhibition by 10 μM ibogaine was 51 ± 9% ($N\pm3$) and 82 ± 3% ($N\pm3$), respectively (Fig. 3). These data are consistent with IC_{50} values of 3–5 μM obtained in hippocampal neurons (Popik et al., 1995; Chen et al., 1996). However, we also found that co-application of 18-methoxycoronaridine failed to inhibit either NR1/2A ($N=3$) or NR1/2B receptors ($N=5$) at concentrations up to 20 μM (Fig. 3). This result necessarily calls into question any role of NMDA receptors in the putative anti-addictive actions of these drugs.

There are no published studies examining the effects of ibogaine on 5-HT_3 receptor function. Although some action may be expected based on competition binding studies (Sweetnam et al., 1995), it remains to be seen whether ibogaine binding to these receptors activates, inhibits, or otherwise alters channel function. Likewise, the effects of 18-methoxycoronaridine on 5-HT_3 receptor function have

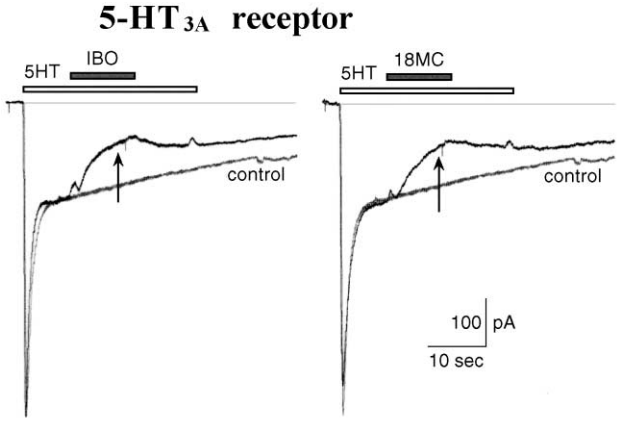

Fig. 4. Inhibition of 5-HT_3 receptors by ibogaine (IBO) and 18-methoxycoronaridine (18MC). Whole-cell currents in cells expressing recombinant 5-HT_{3A} receptors were evoked by 100 μM serotonin (5HT). Open bars depict the timing of agonist application. Filled bars depict the timing of co-application of 20 μM IBO or 20 μM 18MC. Inhibition was measured relative to control at the end of the drug application (arrows).

not been examined. Transfected HEK293 cells expressing 5-HT_{3A} receptors were voltage-clamped to −70 mV and stimulated with 100 μM serotonin at 30-s intervals. Serotonin alone evoked an inward current not seen in untransfected cells. Application of 20 μM ibogaine or 20 μM 18-methoxycoronaridine alone did not produce any response, indicating that neither is an agonist at 5-HT_3 receptors. Co-application of 20 μM ibogaine or 20 μM 18-methoxycoronaridine inhibited serotonin-evoked responses by 53 ± 3% (N=8) versus 50 ± 3% (N=4), respectively (Fig. 4). Thus, it appears that these drugs have similar potencies at 5-HT_3 receptors with IC_{50} values of approximately 20 μM.

3.2. In vivo treatment effects

Figs. 5–7 show the effects of drug treatments on morphine and methamphetamine self-administration and on responding for water. All three drug combinations, but none of the drugs administered alone, significantly decreased morphine and methamphetamine self-administration while having no effect on responding for water. The particular doses of 18-methoxycoronaridine, dextromethorphan and mecamylamine selected for study were, in each instance, based on knowledge of the respective dose–response functions. The doses of 18-methoxycoronaridine (1 and 2 mg/kg) were approximately one-fifth of those required to decrease morphine (Glick et al., 1996) and methamphetamine (Glick et al., 2000a) self-administration, respectively, when administered alone. The dose of dextromethorphan (5 mg/kg) was one-half to one-fourth of that necessary to decrease morphine and methamphetamine self-administration (Glick et al., 2001), respectively, when administered alone. The dose of mecamylamine (1 mg/kg) was one third of that required to decrease either morphine or methamphetamine self-administration, and at a dose of 3 mg/kg, mecamylamine also decreases responding for water (data not shown). Lastly, although Fig. 7 only shows results with the 2 mg/kg dosage of 18-methoxycoronaridine, virtually identical results were found with 1 mg/kg.

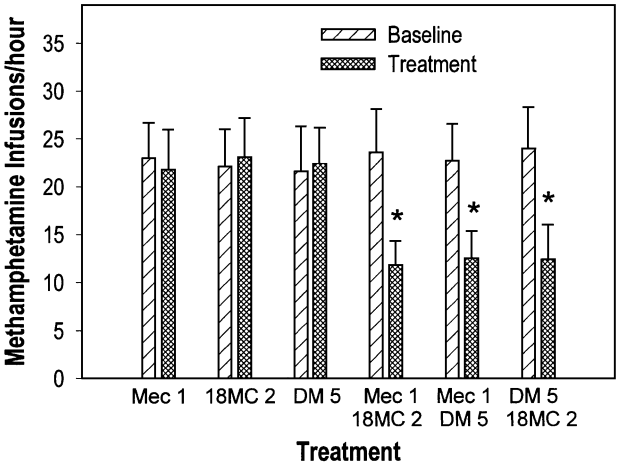

Fig. 6. Effects of drug combinations on methamphetamine self-administration. Rats were administered two of the following treatments before testing: mecamylamine (MEC; 1 mg/kg i.p., 30 min), 18-methoxycoronardine (18MC; 2 mg/kg i.p., 15 min), dextromethorphan (DM; 5 mg/kg s.c., 20 min), or vehicle (saline for MEC and DM; phosphate buffer for 18MC). Each data point represents the mean (± SEM) responses per hour of 6–7 rats. * Significant differences between baseline and treatment (paired t-test, $P<0.01$).

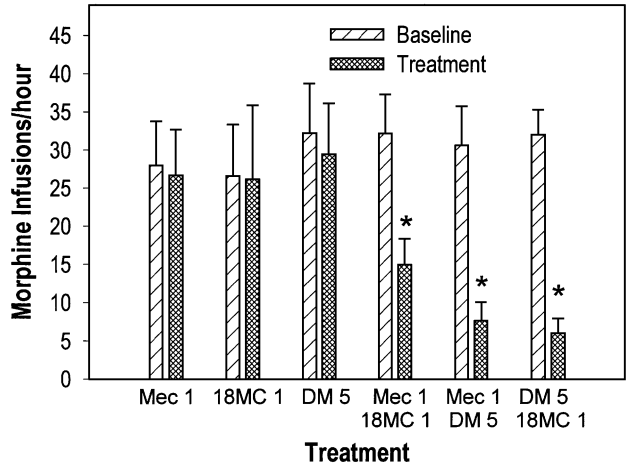

Fig. 5. Effects of drug combinations on morphine self-administration. Rats were administered two of the following treatments before testing: mecamylamine (MEC; 1 mg/kg i.p., 30 min), 18-methoxycoronardine (18MC; 1 mg/kg i.p., 15 min), dextromethorphan (DM; 5 mg/kg s.c., 20 min), or vehicle (saline for MEC and DM; phosphate buffer for 18MC). Each data point represents the mean (± SEM) responses per hour of 6–8 rats. * Significant differences between baseline and treatment (paired t-test, $P<0.01$–0.001).

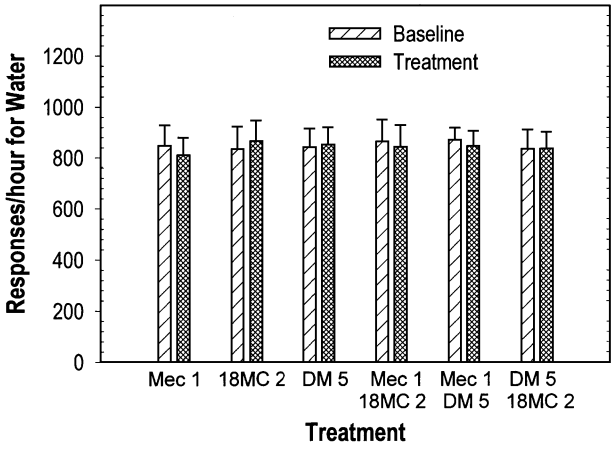

Fig. 7. Effects of drug combinations on responding for water. Rats were administered two of the following treatments before testing: mecamylamine (MEC; 1 mg/kg i.p., 30 min), 18-methoxycoronardine (18MC; 2 mg/kg i.p., 15 min), dextromethorphan (DM; 5 mg/kg s.c., 20 min), or vehicle (saline for MEC and DM; phosphate buffer for 18MC). Each data point represents the mean (± SEM) responses per hour of six rats.

4. Discussion

Previous studies have reported inhibition by ibogaine of both nicotinic acetylcholine receptors (Badio et al., 1997; Mah et al., 1998; Fryer and Lukas, 1999) and NMDA receptors (Popik et al., 1995; Chen et al., 1996). Competitive binding to both NMDA receptors and 5-HT_3 receptors has also been demonstrated (Sweetnam et al., 1995). However, the selectivity of ibogaine for the various neurotransmitter receptor ion-channel subtypes has been little explored and the activity of 18-methoxycoronaridine at these receptors has not been tested. Therefore, we sought to compare the actions of ibogaine and 18-methoxycoronaridine at nACh, NMDA, and 5-HT_3 receptors.

The present results are consistent with previous reports of the antagonist action of ibogaine at $\alpha3\beta4$ nicotinic receptors; and the data also indicate that 18-methoxycoronaridine has a similar action. Furthermore, both ibogaine and 18-methoxycoronaridine show some selectivity for this site in that much higher drug concentrations were required to block other sites. Ibogaine was at least five times less potent at $\alpha4\beta2$ than at $\alpha3\beta4$ sites, and even more times less potent at NMDA and 5-HT_3 receptors. 18-Methoxycoronaridine was approximately 25 times less potent at 5-HT_3 than at $\alpha3\beta4$ receptors and, up to at least 20 μM, was inactive at $\alpha4\beta2$ and NMDA receptors. Hence, the data suggest that antagonism at $\alpha3\beta4$ receptors is a potentially important mechanism of action for both ibogaine and 18-methoxycoronaridine.

Low dose combinations of 18-methoxycoronaridine with either mecamylamine or dextromethorphan or of mecamylamine with dextromethorphan all significantly reduced morphine and methamphetamine self-administration without affecting responding for water. It should be noted that although baseline rates of responding for water were much higher than baseline rates of drug self-administration, previous work (e.g., Glick et al., 1991) has established that higher rates are more rather than less sensitive to nonspecific treatment effects. Due to the inverted U shape that is characteristic of drug self-administration dose-effect functions, treatment-induced decreases in drug self-administration at a single infusion dosage could conceivably reflect either potentiation (leftward shift of dose-effect function) or antagonism (rightward shift of dose-effect function) of the self-administered drug. Although this issue was not directly addressed here, previous work involving the effects of 18-methoxycoronardine on morphine self-administration (Maisonneuve and Glick, 1999) and of dextromethorphan on methamphetamine self-administration (Jun and Schindler, 2000) indicated that, in both instances, the infusion dose–effect function was shifted downward without displacement to the left or right. This suggests that these treatments reduced the reinforcing efficacy of the self-administered drugs. Because the effects of the combination treatments assessed in the present study closely mimicked the effects produced by higher doses of each component of the treatments (e.g., Glick et al., 2000a,b, 2001), it is likely that the reinforcing efficacies of the self-administered drugs were similarly reduced here.

Antagonism at $\alpha3\beta4$ sites is the only known action that 18-methoxycoronaridine, mecamylamine and dextromethorphan have in common. The self-administration results are therefore consistent with our original hypothesis implicating antagonism at $\alpha3\beta4$ receptors as a potential mechanism to modulate drug seeking behavior. However, this antagonism at $\alpha3\beta4$ receptors may not directly involve the dopaminergic mesolimbic pathway presumed to mediate drug reward. Although low densities of $\alpha3\beta4$ receptors reside in the dopaminergic nuclei of the ventral tegmental area, $\alpha3\beta4$ nicotinic receptors are mainly located in the medial habenula and the interpeduncular nucleus (e.g., Klink et al., 2001; Quick et al., 1999). While the interpeduncular nucleus receives its main input from the medial habenula, forming the habenulointerpeduncular pathway, there are multiple avenues for interaction between this pathway and the mesolimbic pathway. Thus, for example, the medial habenula receives input from the nucleus accumbens and has efferents to the ventral tegmental area; and the interpeduncular nucleus has efferent connections to the brainstem raphe nuclei and the medial dorsal thalamic nucleus, both of which directly or indirectly (e.g., via the prefrontal cortex) connect to the ventral tegmental area. Functional interactions between the habenulointerpeduncular and mesolimbic pathways have in fact been demonstrated (Nishikawa et al., 1986).

Aside from accounting for other preclinical effects of dextromethorphan (Pulvirenti et al., 1997; Jun and Schindler, 2000; Glick et al., 2001) and mecamylamine (Levin et al., 2000) on drug self-administration, the mechanism proposed here may also underlie effects already reported in the human literature. Administration of mecamylamine was found to reduce cue-induced craving in patients addicted to cocaine (Reid et al., 1999), and in short-term clinical trials, dextromethorphan reduced heroin intake (Koyuncuoglu, 1995), craving (Koyuncuoglu and Saydam, 1990), and signs of opioid withdrawal (Koyuncuoglu and Saydam, 1990; Koyuncuoglu, 1995). 18-Methoxycoronaridine, having apparently greater selectivity for $\alpha3\beta4$ sites than either mecamylamine or dextromethorphan, or the prototypical *iboga* alkaloid ibogaine, may represent the first of a new class of synthetic agents acting via a novel mechanism and having a broad spectrum of activity to diminish multiple forms of addictive behavior. This hypothesis will be pursued in further studies.

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