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Effect of ibogaine on morphine: Induced modifications of palatability

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Effect of Ibogaine on Morphine-Induced Modifications of Palatability

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ABSTRACT

The ability of the potential anti-addictive agent ibogaine to modulate th . morphine-induced modification of quinine and sucrose palatability was assessed utilizing the taste reactivity test. Ibogaine (40 mg/kg) was administered 24 hr prior to an injection of morphine (2 mg/kg), followed 30 min later by a 5 min intraoral infusion of 0.05% quinine solution (Experiment 1) or 10% sucrose solution (Experiment 2). Treatment with morphine enhanced the palatability of both quinine and sucrose solution. Morphine reduced the aversiveness of quinine solution during the 5 min of testing and enhanced ingestive responding to sucrose solution, however, only during min 1 of the 5 min test. Pretreatment with ibogaine 24 hr earlier, regardless of treatment condition, also enhanced the palatability of quinine and sucrose solution. However, there was no evidence that ibogaine modulated the effect of morphine on quinine or sucrose palatability.

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INTRODUCTION

Recently, it has been suggested that ibogaine, an indole alkaloid found in the root bark of the Tabernanthe iboga shrub growing in West Central Africa, effectively attenuates the physiological and psychological properties of drug addiction. Howard Lotsof has been awarded 5 United States patents that propose that ibogaine treatment is effective as a pharmacotherapy to combat opiate addiction (U.S. patent 4,499,096), stimulant abuse (U.S. patent 4,587,243), alcohol abuse (U.S. patent 4,857,523), nicotine addiction (U.S. patent 5,026,697), and poly-drug dependency (U.S. patent 5,124,994) on the basis of anecdotal reports from drug users.

Although the anecdotal reports suggest that ibogaine may be an effective agent in reducing craving for addictive drugs, basic research on the efficacy of ibogaine to modify drug reward in animal models is lacking. The present study will investigate the ability of ibogaine to modify the palatability modulating effects of morphine in rats.

Ibogaine and Drug Reward

Studies directly examining the effects of ibogaine on drug-induced reward present compelling evidence that ibogaine reduces the rewarding properties of both cocaine and morphine. Several investigators have shown that administration of ibogaine disrupts intravenous and oral self-administration of morphine and cocaine. Glick, Rossman, Steindorf, Maisonneuve, and Carlson (1991)

demonstrated that pretreatment with ibogaine at doses greater than 10 mg/kg significantly reduced the intravenous self-administration of morphine 24 hr later, while at the same time, did not disrupt responding for water. In some animals, this suppression of responding for morphine persisted for several weeks. Similarly, Cappendijk and Dzoljic (1993) reported that pretreatment with 40 mg/kg ibogaine reduced the intravenous self-administration of cocaine. Additionally, ibogaine pretreatment also reduces the preference for and the oral consumption of cocaine (Sershen, Hashim, & Lajtha, 1994). Mice pretreated with 40 mg/kg ibogaine displayed reduced preference for and consumption of solution containing 200 mg/l cocaine.

The drug self-administration paradigm, however, has several disadvantages which may obscure the interpretation of ibogaine-induced changes in rates of responding for a drug. First, because testing occurs when animals are not drug free, a change in the rate of responding for the drug may be the result of direct motoric effects of the drug. Second, although it is possible that animals decrease their rate of responding for a drug because the drug is less rewarding, it is also possible that they decrease their rate of responding for a drug because the effective dose of the drug has increased rendering each infusion more potent. That is, as the animal attempts to maintain a constant level of drug reward, the rate of responding for a more potent drug infusion decreases.

Another measure of drug reward, known as the conditioned place preference paradigm, has been utilized to demonstrate that ibogaine reduces the rewarding properties of morphine. In the conditioned place preference paradigm, animals are injected with a particular drug and placed in a distinctive environment on one occasion, and injected with saline and placed in another, equally distinct, environment on a separate occasion. The animal is then given drug-free access to both places simultaneously. If the animal shows a preference for the drug-paired place, it is then assumed that the particular drug is rewarding. When morphine serves as the conditioning drug in the place preference paradigm, the strength of the place preference increases linearly with an increase in the dose of morphine, until reaching an asymptotic level (van der Kooy, 1987); that is, the strength of the place preference does not decline when higher doses of morphine are employed.

Parker, Siegel, and Luxton (submitted) have shown that administration of 40 mg/kg ibogaine prevents the establishment of a one-trial morphine-induced conditioned place preference. Given that ibogaine alone produced neither a place preference nor a place aversion, and that it did not interfere with place aversions produced by lithium or naloxone, it was concluded that ibogaine disrupted the rewarding effect of morphine rather than simply the formation of a drug-place association.

Ibogaine and Dopamine

The interaction between ibogaine and the dopamine system may be crucial in characterizing the effects of ibogaine on drug-taking behaviour. Investigations of intracranial electrical self-stimulation and intracranial drug self-administration have demonstrated the importance of dopaminergic systems in the reward process (e.g. Wise & Bozarth, 1982). Investigators have therefore attempted to identify and characterize the effect of ibogaine, if any, on the dopaminergic system..

In vivo microdialysis has been used to provide a measure of the effects of ibogaine pretreatment on extracellular dopamine (DA) levels in specific regions of the brain. In vivo microdialysis has revealed that both systemic and central injections of ibogaine reduce extracellular DA levels in the striatum and in the nucleus accumbens (nAcc) when measured 1 hr after the injection, but not when measured 19 hr after the injection (Maisonneuve, Keller, & Glick, 1991; Maisonneuve, Keller, & Glick, 1992; Glick, Rossman, Wang, Dong, & Keller, 1993).

While in vivo microdialysis yields a measure that represents extracellular DA levels, post-mortem tissue assay provides an indication of primarily intracellular DA levels. Results of experiments employing these two methods of assessment are generally in agreement concerning the effect of ibogaine on DA levels. When assessed 1-2 hr after administration of ibogaine,

but not 19 hr to 1 week after ibogaine treatment, post-mortem tissue levels of DA in the striatum (Maisonneuve, Rossman, Keller, & Glick, 1992; Sershen, Hashim, Harsing, & Lajtha, 1992) and nAcc (Maisonneuve et al., 1992) were nearly half of those in control animals.

The ability of ibogaine to modify the morphine-induced release of dopamine has also been assessed using in vivo microdialysis. Systemic injections of morphine (5 mg/kg) produced an increase in extracellular DA levels in the nAcc and striatum (Maisonneuve et al., 1991). However, when rats were pretreated with 40 mg/kg of ibogaine 19 hr prior to the morphine injections, the morphine induced rise in DA levels was no longer evident. This suggests that ibogaine interferes with the morphine-induced increase in DA even 19 hr after being administered.

Clearly, pretreatment with ibogaine modifies the dopaminergic system, and inhibits the morphine-induced rise in DA levels in the striatum and nucleus accumbens. A discrepancy exists, however, in the time course of ibogaine's effects on the DA system and its effects on drug-induced changes in the DA system and on drug reward. Nineteen hr after the administration of ibogaine, DA levels in the striatum and nAcc are unaffected when measured by in vivo microdialysis and post mortem tissue assay. However, at the same post-injection interval (19+ hr), ibogaine prevents the morphine-induced enhancement of DA release

at the nAcc and striatum, attenuates the intravenous self-administration of morphine, and prevents the establishment of a one trial morphine-induced conditioned place preference. At a time when ibogaine no longer has any measurable effect on DA levels, it disrupts other DA-dependent phenomena, such as the maintenance of morphine self-administration, and the establishment of a morphine-induced conditioned place preference.

Ibogaine and Motor Behaviour

It has consistently been observed that administration of ibogaine has a disruptive effect on locomotor activity in rats and mice. Systematic observations of ibogaine's effects on motor behaviour were conducted by O'Hearn, Long and Molliver (1993) and O'Hearn and Molliver (1993). Within 1 to 2 min following administration of a large dose of ibogaine (100 mg/kg), rats display a fine amplitude fast tremor of the head, as well as marked truncal ataxia when locomotion was attempted. The fine tremor and ataxia continues for 6 to 8 hr following injection, then gradually subsides as spontaneous activity resumes. Twenty-four hr following the ibogaine injection, rats display relatively normal motor activity. Utilizing automated photocell monitoring of ambulatory behaviour, it has been shown that ibogaine, at a dose of 40 mg/kg, inhibits motor activity during the first hr of measurement (Maisonneuve, Keller, & Glick, 1992; Maisonneuve et al., 1992; Sershen, Hashim, Harsing, & Lajtha, 1992). This effect was observed whether ibogaine was administered 19 hr or 1

hr before activity measurement.

Morphine and Feeding

The role of endogenous opioids in feeding behaviour has been clearly demonstrated. Beginning with Holtzman (1974), who reported that naloxone, an opiate antagonist, markedly reduced consumption in food deprived rats, researchers have shown that excitation of the opioid system with agonists such as morphine, enhances feeding (e.g. Capuano, Leibowitz, & Barr, 1990; Morley, Levine, Kneip, Grace, Zeugner, & Sherman, 1985; and Sanger & McCarthy, 1980).

There is evidence that morphine-induced enhancement of feeding is centrally mediated and that it potentially involves reward mechanisms. Microinjections of morphine directly into the ventral tegmental area (Mucha & Iversen, 1986) and nAcc (Evans & Vaccarino, 1990; Mucha & Iversen, 1986), structures critical for DA-mediated reward, produce an increase in consumption. Furthermore, pretreatment with the dopamine receptor blocker, α-flupenthixol, prevents the morphine-induced enhancement of feeding (Evans & Vaccarino, 1990).

Opiate-Induced Modification of Palatability

Considerable experimental evidence suggests that opiate-induced modification of food intake is mediated by a shift in the relative palatability of the particular food or solution being consumed. Convincing evidence for the palatability hypothesis

has been provided by investigations that used the sham-feeding paradigm and two-bottle preference test. In the sham-feeding paradigm, animals are implanted with gastric fistulae, which allow the stomach contents to drain prior to absorption, thereby preventing the process of satiation. If rats suppress intake in a sham feeding preparation, it is assumed that the suppression is mediated by a preabsorptive mechanism such as a shift in the palatability of the food. Rats pretreated with naloxone, an opiate antagonist, display a significant decrease in consumption of sham-fed sucrose (Rockwood & Reid, 1982).

In the two-bottle preference test animals are presented with one bottle containing a flavoured solution and one containing water. The ratio of the amount of flavoured solution ingested relative to the total amount of fluid consumed is calculated. Since the dependent measure is relative preference rather than total amount consumed (as in a one choice test), the effect of a manipulation of preference for the flavoured solution cannot be merely interpreted as hypodipsia or hyperdipsia; instead a modification in relative preference for the flavoured solution is interpreted as a palatability shift. Two-bottle preference tests reveal that animals pretreated with morphine will demonstrate an increased preference for sweetened food or water compared to unadulterated water (e.g. Evans & Vaccarino, 1990; Milano, Wild, Hui, Hubbell, & Reid, 1989).

The potential confound with the sham-feeding and two bottle

preference test paradigms is that interpretation may be obscured by motivational factors other than palatability. Since the animal must voluntarily approach and consume the food or solution, other influences, such as the activity and satiety level of the animal, could modulate consumption. A more direct measure of palatability, the taste reactivity (TR) test, was developed in 1978 by Grill and Norgren. In the taste reactivity paradigm, rats are surgically implanted with an intraoral cannula allowing the investigator to deliver a particular tastant directly into the animal's mouth. Depending on the palatability of the solution being infused, rats will display a characteristic pattern of orofacial reactions. For example, an infusion of highly palatable sucrose solution typically elicits ingestive reactions such as tongue protrusions and mouth movements, whereas, an infusion of bitter quinine solution elicits aversive reactions such as gapes, chin rubs, and paw pushes. This test permits the direct examination of the effects of both rewarding and non-rewarding agents on the palatability of various substances.

Utilizing the taste reactivity test, investigators have shown that morphine enhances the palatability of both sucrose and quinine solutions. When morphine was systemically administered 30-120 min prior to an infusion of sucrose (Rideout & Parker, in press) or a sucrose/quinine mixture (Doyle, Berridge, & Gosnell,

1993), rats spent more time displaying ingestive reactions. Additionally, rats pretreated with morphine displayed significantly fewer aversive reactions during an infusion of 0.05% quinine solution (Parker, Maier, Rennie, & Crebolder, 1992; Clarke & Parker, in press). Therefore, morphine pretreatment enhanced the palatability of both sucrose and quinine solutions.

To assess whether changes in sucrose and quinine palatability are DA-mediated, the effect of the dopamine receptor blocking agent, pimozide, and the indirect dopamine agonist, amphetamine, on taste reactivity have been assessed. Pimozide enhanced the aversive reactions elicited by quinine solution (Parker & Lopez, 1990), and suppressed the ingestive reaction of tongue protrusions elicited by sucrose solution, suggesting that dopamine blockade reduced the palatability of these tastants (Leeb, Parker, & Eikelboom, 1991). Conversely, it would be expected that excitation of the DA system would result in an enhancement of palatability. Rats pretreated with the indirect DA agonist amphetamine, displayed suppressed aversive reactions in response to an intraoral infusion of quinine solution (Parker, and Leeb, 1994), but did not display a modification of ingestive reactions during a sucrose infusion.

On the other hand, Berridge and colleagues have reported that dopamine blockade by 6-hydroxydopamine (6-OHDA) lesions of the midbrain DA systems (Berridge, Venier, & Robinson, 1989) and pretreatment with haloperidol, a DA receptor blocker (Treit &

Berridge, 1990), modified neither sucrose nor quinine palatability. A primary difference in methodology between the studies conducted by Parker and colleagues and Berridge and colleagues is the duration of the taste reactivity (TR) test; Parker and colleagues employ a 5-10 min TR test, whereas Berridge and colleagues employ a 1 min TR test. In fact, the effects of pimozide and amphetamine on palatability are not apparent during the first min of the longer tests (Lopez & Parker, 1990; Leeb et al., 1991; Parker & Leeb, 1994). Current investigations are underway between the laboratories of Berridge and Parker to determine the conditions under which pimozide and amphetamine modify palatability.

Present Experiment

The present series of experiments examine whether or not morphine-induced modulation of quinine and sucrose palatability can be modified by pretreatment with ibogaine. It is predicted then, that pretreatment with ibogaine should prevent the expected morphine-induced reduction of quinine aversiveness and enhancement of sucrose palatability.

EXPERIMENT 1

Experiment 1 investigated the effect of ibogaine on morphine-modulated taste reactions elicited by quinine solution. Rats were pretreated with 40 mg/kg, ip, of ibogaine 24 hr prior to receiving a systemic injection of morphine (2 mg/kg, sc) or saline. Thirty min later they were intraorally infused with 0.05% quinine solution for a 5 min taste reactivity (TR) test.

METHOD

Subjects

The subjects were 26 naive male Sprague-Dawley rats weighing 320-393 g on the day of the test. They were housed individually in suspended stainless steel cages and were maintained, ad libitum, on Purina rat chow. The housing room was illuminated on a 12 hr Light/12 hr Dark schedule.

Drugs

Ibogaine HCl (donated by The National Institute on Drug Abuse) was prepared daily at a concentration of 10 mg/ml of distilled water and was administered intraperitoneally (ip). Morphine sulphate (donated by The National Institute on Drug Abuse) was prepared at a concentration of 1 mg/ml of saline and was administered subcutaneously (sc).

Apparatus

The taste reactivity test was conducted in a glass test chamber measuring 22.5 x 26 x 20 cm. The chamber was illuminated by four 100W light bulbs, with two on either side of the chamber

and two aimed at a mirror hung below the floor of the chamber. Each rat's cannula was attached to the infusion pump (Harvard Apparatus, Model 22) by a 35 cm length of polyethylene (PE 90) tubing. To facilitate scoring of the orofacial and somatic reactions of each rat, a Panasonic videocamera was focussed at the mirror which was hung at an angle, providing a view of the ventral surface of each rat, below the floor of the test chamber.

Procedure

Surgery. One week after arriving in the laboratory, all rats were implanted with intraoral cannulae. After being deprived of water for 24 hr, each rat was anaesthetized with a mixture of ketamine (100 mg/kg, ip) and rompun (10 mg/kg, ip) 15 min after being injected with atropine (0.5 ml, ip). A 15 gauge thin-walled stainless steel needle was inserted through the skin in the dorsal mid-neck region. The needle was advanced subcutaneously behind the ear along the inside of the cheek where it exited into the mouth behind the first molar through the soft part of the cheek. A 10 cm length of polyethylene (PE 90) tubing was inserted through the barrel of the needle; which was then removed, leaving the tubing in its place. The tubing was then cut to the proper length and secured at the neck by a 20 gauge intramedic adapter, and in the mouth by a 5 mm rubber O-ring. To prevent infection, the skin at both punctures sites was swabbed with iodine during the surgery, and throughout the experiment as required.

TR Testing. After a one week recovery period, each rat was given three 5 min adaptation trials, each separated by 24 hr. On each of the adaptation trials, the rat was removed from its home cage and placed in the TR test chamber and its cannula was attached to the infusion pump. After a 1 min habituation period in the chamber, each rat received a 5 min intraoral infusion of water at a rate of 1 ml/min.

The design of the experimental manipulations is presented in Table 1. Separate groups of rats were randomly assigned to pretreatment conditions (Ibogaine, Saline) and treatment conditions (Morphine, Saline), with 6-7 rats per group. On the day following the last adaptation trial, the rats were given the pretreatment injection. Each rat was injected with 40 mg/kg of ibogaine or saline (at a volume of 4 ml/kg) and returned to its home cage. Twenty-four hr later, the rats received injections of 2 mg/kg of morphine or saline (at a volume of 2 ml/kg) 30 min prior to receiving a 5 min intraoral infusion of 0.05% quinine sulfate solution at a rate of 1 ml/min in the TR test chamber.

Behavioural Categories. The videotapes of the TR test were scored in real time by observers blind to the experimental assignments by means of an event recorder for the IBM personal computer (The Observer, Noldus Inc., NL). The behavioural categories were: aversive reactions, comprised of chin rubbing (CR: forward movement of the head with the chin rubbing against the floor or a wall), gaping (G: triangular, wide opening of the

Table 1. Design of Experiment 1.

Pretreatment	24 hr ------> Treatment	30 min --------> TR Trial
Ibogaine	Morphine	Quinine (n=7)
Ibogaine	Saline	Quinine (n=7)
Saline	Morphine	Quinine (n=6)
Saline	Saline	Quinine (n=6)

mouth), paw pushing (PP: rhythmic treading of the front paws against the floor or a wall), head shaking (HS: rapid shaking of the head from side to side), and limb flicking (LF: rapid flailing of the front paws). The 5 aversive behaviours were also combined to produce a composite score of total aversive reactions, which was included in the analyses in addition to the 5 individual reactions. The measures of activity included rearing (R: lifting both front paws off of the floor without engaging in grooming), and active locomotion (AL: forward or lateral movement along the floor of the chamber with both front paws on the floor). In addition, the neutral/mildly aversive reaction of passive dripping (PD: allowing the solution to fall from the mouth without actively attempting to expel it) was also included in the analysis. Each of the TR reactions described above were collected as frequency scores.

Data Analysis

The frequency of the individual taste reactions as well as the composite scores elicited during the 5 min intraoral infusion of quinine solution were each entered into a 2 by 2 ANOVA with the factors of pretreatment condition (Ibogaine or Saline) and treatment condition (Morphine or Saline). The same analysis of each behaviour was also conducted for the first min of testing only, because 1 min is the duration most commonly employed in TR testing (e.g. Berridge & Grill, 1983).

RESULTS

5 min Taste Reactions

Figure 1 presents the mean frequency of each taste reaction elicited by 0.05% quinine solution during the 5 min TR test in Experiment 1. As indicated in the figure, rats injected with morphine 30 min prior to the quinine infusion displayed fewer of the active aversive reactions of chin rubs, head shakes, and limb flicks, but more passive dripping than rats injected with saline. The 2 by 2 ANOVAs of each of the taste reactions revealed a significant main effect of treatment for chin rubs [$F(1,22)=5.36$, $p<.05$], head shakes [$F(1,22)=32.29$, $p<.001$], limb flicks [$F(1,22)=6.69$, $p<.025$], and passive drips [$F(1,22)=16.38$, $p<.001$].

Additionally, pretreatment with ibogaine also modified taste reactions elicited by quinine solution, independently of the treatment condition. Rats pretreated with ibogaine 24 hr prior to the second injection of either morphine or saline displayed fewer of the active aversive reaction of paw pushes, but more passive dripping than did rats pretreated with saline. The analysis revealed a significant main effect of pretreatment for paw pushes [$F(1,22)=6.59$, $p<.025$], passive drips [$F(1,22)=7.90$, $p<.01$]. There is no evidence, however, that pretreatment with ibogaine modified the effect of morphine treatment. The pretreatment by treatment interaction was not significant for any behaviour.

Figure 1. Mean (±SEM) frequency of aversive taste reactions and passive drips elicited by a 5 min intraoral infusion of 0.05% quinine solution in Experiment 1.

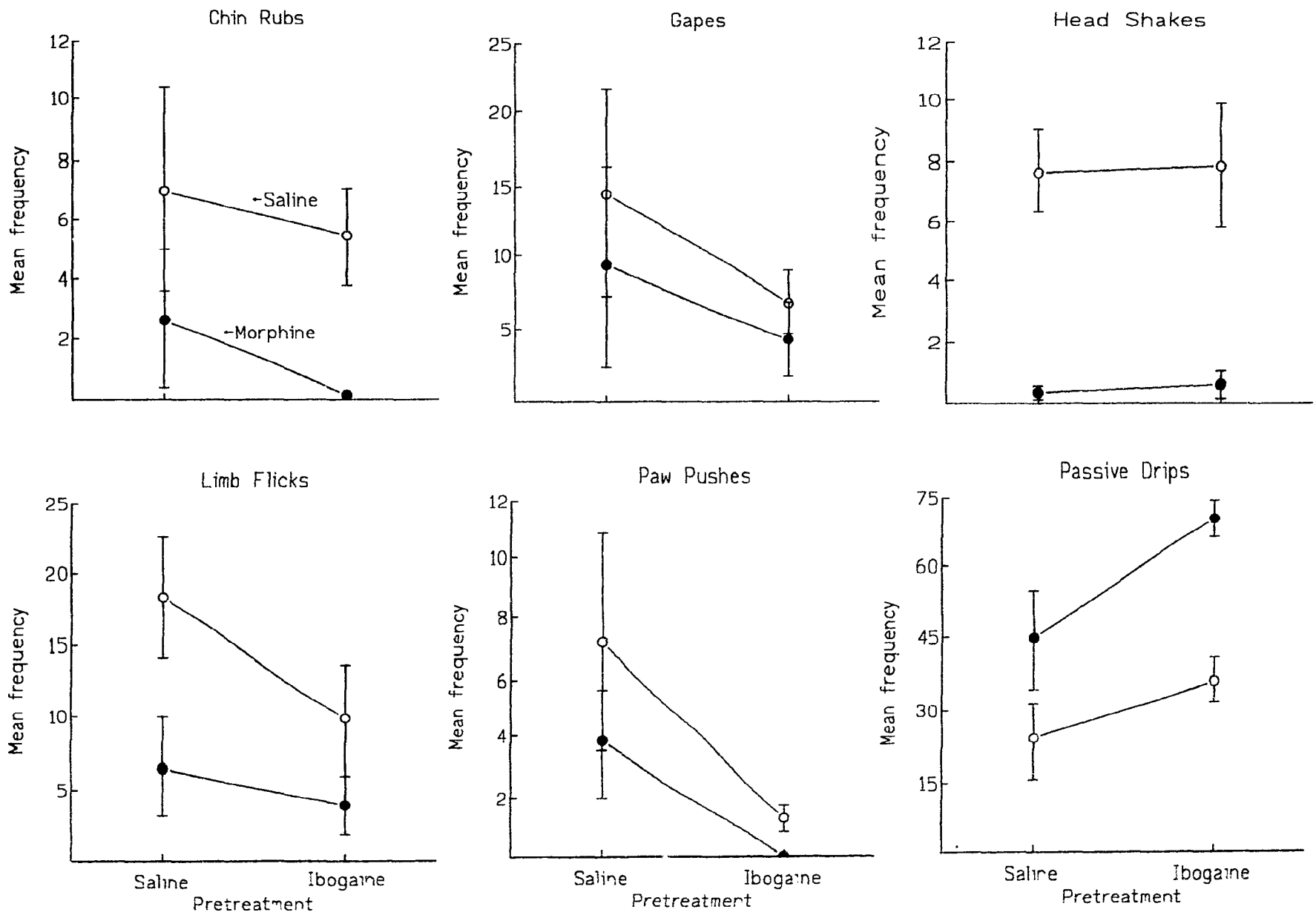

Chin Rubs
Gapes
Head Shakes
Limb Flicks
Paw Pushes
Passive Drips
Mean frequency
←Saline
←Morphine
Saline
Ibogaine
Pretreatment

Figure 2 presents the composite score of total aversive reactions summed across the 5 individual aversive reactions (CR, G, HS, LF, PP) during the 5 min test of Experiment 1. A 2 by 2 ANOVA revealed a significant main effect of treatment for total aversive reactions, $F(1,22)=6.20$, $p<.025$; morphine suppressed aversive reactions elicited by quinine. Additionally, the main effect of pretreatment approached significance, $F(1,22)=2.90$, $p=.10$; ibogaine tended to suppress quinine aversiveness. However, there is no evidence that ibogaine modulates the effect of morphine on palatability. The pretreatment by treatment interaction was not significant.

Figure 3 presents the mean frequency of each of the general activity measures, rearing and active locomotion, during the 5 min of testing. Morphine suppressed both rearing and active locomotion. The 2 by 2 ANOVAs for each measure revealed a significant main effect of treatment for rearing [$F(1,22)=10.70$, $p<.01$] and active locomotion [$F(1,22)=4.51$, $p<.05$]. Furthermore, ibogaine suppressed both rearing and active locomotion. The analysis revealed a significant main effect of pretreatment for rearing [$F(1,22)=11.40$, $p<.01$] and active locomotion [$F(1,22)=5.02$, $p<.05$]. Ibogaine pretreatment, however, did not modify the effect of morphine on activity; the pretreatment by treatment interaction for neither rearing nor active locomotion was significant.

Figure 2. Mean (±SEM) frequency of total aversive reactions displayed during the 5 min intraoral infusion of 0.05% quinine solution in Experiment 1.

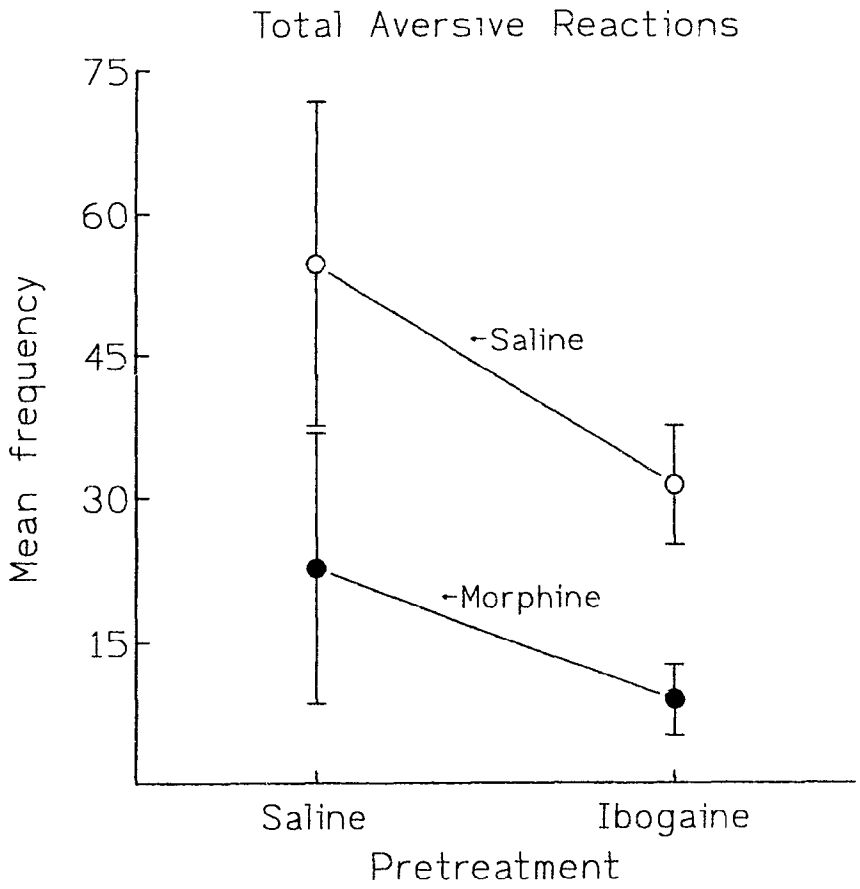
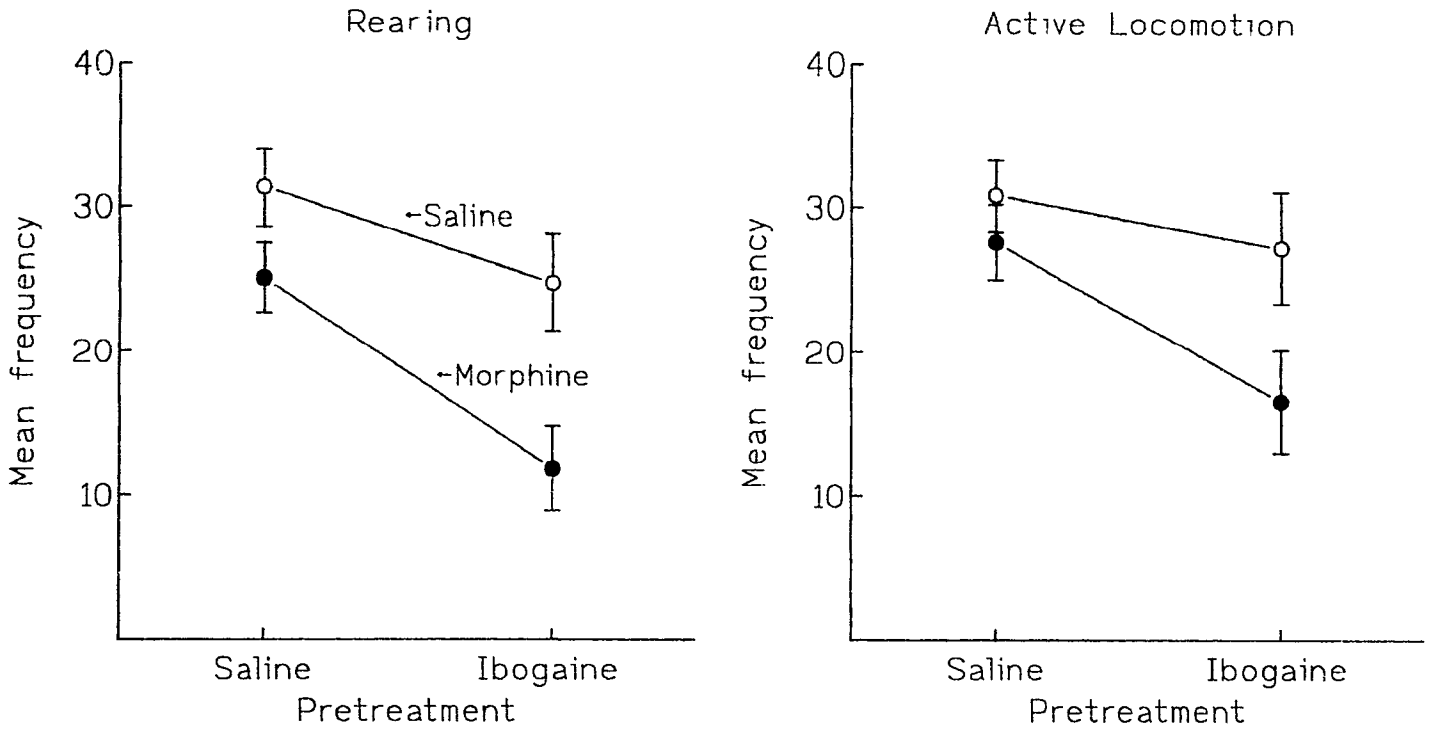

Total Aversive Reactions
75
60
45
30
15
Mean frequency
←Saline
←Morphine
Saline
Ibogaine
Pretreatment

Figure 3. Mean (±SEM) frequency of rearing and active locomotion displayed during the 5 min intraoral infusion of 0.05% quinine solution in Experiment 1.


Rearing
Active Locomotion
Mean frequency
40
30
20
10
←Saline
←Morphine
Saline
Ibogaine
Pretreatment

Minute 1 Taste Reactions

The taste reactions elicited by 0.05% quinine solution during min 1 only of the 5 min test were also analyzed. As in the total 5 min test, morphine reduced the aversiveness of quinine solution. The 2 x 2 ANOVAs revealed a significant treatment effect for the reactions of head shakes [$F(1,22)=28.45$, $p<.001$], total aversive reactions [$F(1,22)=6.82$, $p<.025$], passive drips [$F(1,22)=14.86$, $p<.001$], and rearing [$F(1,22)=4.79$, $p<.05$] during min 1. Morphine treatment suppressed head shakes, total aversive reactions, and rearing, but enhanced the frequency of passive drips during min 1 of testing. A significant pretreatment effect was also obtained for the reactions of passive drips [$F(1,22)=5.78$, $p<0.025$] and rearing [$F(1,22)=4.44$, $p<.05$]. Ibogaine pretreatment, independent of treatment condition, enhanced the frequency of passive drips elicited by quinine solution during the first min of testing, but reduced the frequency of rearing. Ibogaine pretreatment, however, did not modulate the effect of morphine on quinine palatability during min 1 of the 5 min infusion.

Pretreatment-Treatment Body Weight Change

In order to determine whether the enhancement of palatability produced by ibogaine was the result of a difference in deprivation states between the ibogaine and saline pretreated groups, the difference between each rats body weight at treatment and at pretreatment was determined. The resulting change scores

were entered into a 2 by 2 Analysis of Variance (ANOVA) with the factors of pretreatment condition (Ibogaine or Saline) and treatment condition (Morphine or Saline). The analysis revealed only a significant pretreatment condition effect, $F(1,22)=4.74$, $p<.05$. The ibogaine pretreated rats ($x=.79$ gm) had lower change scores than the saline pretreated rats ($x=5.42$ gm) indicating that they gained less weight over the 24 hr period.

DISCUSSION

The findings of the present experiment support earlier findings by Parker et al. (1992) and Clarke and Parker (in press) that morphine reduces the aversiveness of quinine solution. The aversive reactions of chin rubbing, head shaking, and limb flicking were specifically suppressed by treatment with morphine. Additionally, the frequency of passive dripping was enhanced in those rats treated with morphine. This enhancement of passive drips is exactly what would be predicted as morphine shifts the palatability of quinine solution toward neutrality. Morphine treatment also tended to depress activity, indicated by fewer occurances of rearing and active locomotion.

The data also suggest that ibogaine pretreatment also reduces the aversiveness of quinine solution; the frequency of paw pushes, an aversive reaction, was suppressed, and the frequency of passive drips was enhanced. This effect may be the result of a suppression of food intake in the 24 hr period after

ibogaine administration. The ibogaine pretreated rats gained less weight over the 24 hr pretreatment-treatment interval than the saline pretreated rats, suggesting that they may have been in a greater deprivation state.

Although we predicted that ibogaine would interfere with the ability of morphine to modify quinine palatability, the results do not support our prediction. The drugs appeared to act independently on the palatability of quinine solution.

EXPERIMENT 2

In Experiment 1, ibogaine did not modulate the effect of morphine on quinine palatability. Morphine reduced the aversiveness of quinine solution as evidenced by a reduction in aversive taste reactions and an enhancement in the neutral/mildly aversive taste reaction of passive dripping. Ibogaine pretreatment 24 hr prior to morphine treatment did not modify the ability of morphine to enhance quinine palatability.

Morphine has also been shown to modify sucrose palatability. In both a 5 min (Rideout & Parker, in press) and a 1 min TR test (Doyle et al., 1993), morphine pretreatment enhanced sucrose palatability. Experiment 2 assessed the ability of ibogaine to modulate morphine enhancement of sucrose palatability.

The concentration of sucrose solution employed was 10%. Although previous work has employed 20% sucrose (Parker et al., 1992; Rideout & Parker, in press), or 2-7% sucrose (Doyle et al., 1993; Rideout & Parker, in press), a 10% concentration was selected to facilitate the detection of a change in palatability in either direction. Rats display less ingestive responding to 10% sucrose than to 20% sucrose, and more ingestive responding to 10% sucrose than to 2% sucrose (Flynn & Grill, 1988).

METHOD

Twenty-eight naive male Sprague-Dawley rats, weighing 277-448 g on the day of testing, served as subjects; there were 7 rats per group. They were treated in an identical manner as in

Experiment 1 except as indicated. On the day of the test, all rats were given a 5 min intraoral infusion of 10% sucrose solution at a rate of 1 ml/min.

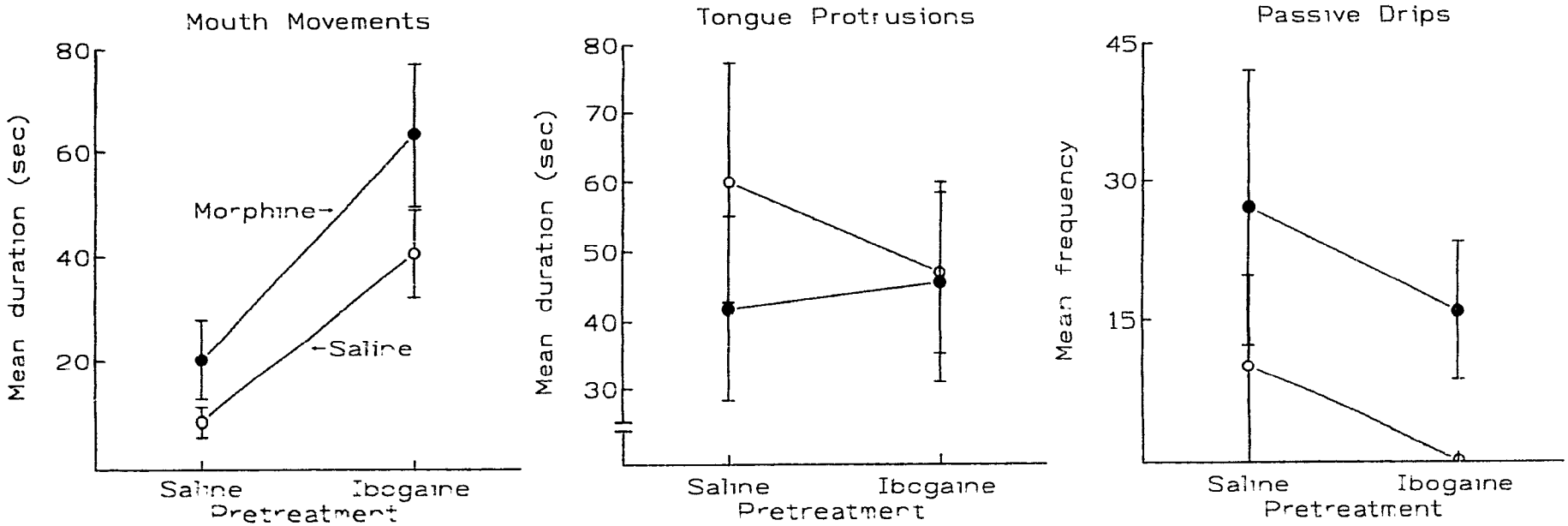
The behavioural categories measured included the following: ingestive taste reactions, activity, and passive dripping. The ingestive taste reactions included the highly ingestive reaction of tongue protrusions (TP: forward or lateral extensions of the tongue), and the mildly ingestive/neutral taste reaction of mouth movements (MM: movement of the lower mandible without extending the tongue or opening the mouth). The measures of general activity included rearing (R: lifting both front paws off of the floor without engaging in grooming), and active locomotion (AL: forward or lateral movement along the floor of the chamber with both front paws on the floor). The neutral/mildly aversive taste reaction of passive dripping (PD: allowing the solution to fall from the mouth without actively attempting to expel it) was also included in the analysis.

RESULTS

5 min Taste Reactions

Figure 4 presents the mean duration or frequency of taste reactions elicited by 10% sucrose solution during the 5 min intraoral infusion of sucrose solution. Among the three taste reactions depicted, the only significant effect revealed by the 2 x 2 ANOVAs was a main effect of pretreatment for the behaviour of mouth movements, $F(1,24)=17.29$, $p<.001$. Regardless of treatment

Figure 4. Mean (±SEM) duration (sec) or frequency of ingestive taste reactions or passive drips elicited by a 5 min intraoral infusion of 10% sucrose solution in Experiment 2.


Mouth Movements
Mean duration (sec)
80
60
40
20
Morphine
Saline
Saline
Ibogaine
Pretreatment
Tongue Protrusions
Mean duration (sec)
80
70
60
50
40
30
Saline
Ibogaine
Pretreatment
Passive Drips
Mean frequency
45
30
15
Saline
Ibogaine
Pretreatment

condition, the ibogaine pretreated rats spent more time engaged in mouth movements than the saline pretreated rats. Administration of morphine 30 min prior to the sucrose infusion did not modify the taste reactions elicited by 10% sucrose solution during the 5 min test, nor was there a significant pretreatment by treatment interaction.

The mean frequency of rearing and active locomotion displayed by rats during the 5 min sucrose infusion is presented in Figure 5. Neither pretreatment with ibogaine, nor treatment with morphine had any significant effect on the measures of rearing or active locomotion.

Min 1 Taste Reactions

Figure 6 presents the mean duration or frequency of taste reactions elicited by the sucrose infusion during the first min of testing. The only behaviour that was affected by the manipulations was that of mouth movements; the 2 x 2 ANOVA revealed a significant main effect of treatment [$F(1,24)=9.15$, $p<.01$], and pretreatment [$F(1,24)=6.73$, $p<.025$]. During the first min of the 5 min test, rats injected with morphine 30 min prior to test spent significantly more time displaying mouth movements than rats injected with saline; likewise, rats pretreated with ibogaine 24 hr prior to the injection of either saline or morphine also spent more time displaying mouth movements than rats pretreated with saline. Ibogaine pretreatment did not, however, modify the effect of morphine on

Figure 5. Mean (±SEM) frequency of rearing and active locomotion displayed during the 5 min intraoral infusion of 10% sucrose solution in Experiment 2.

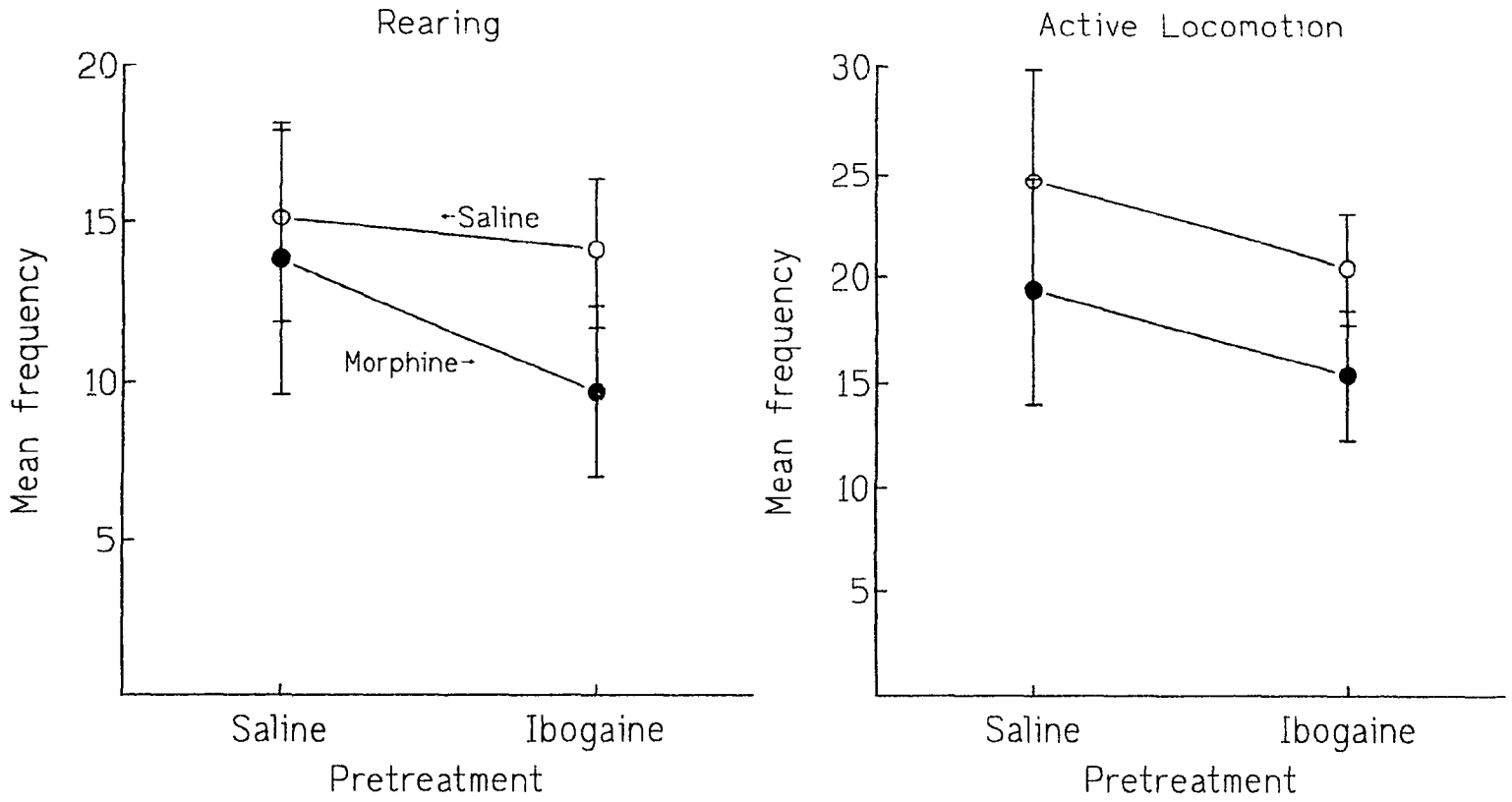
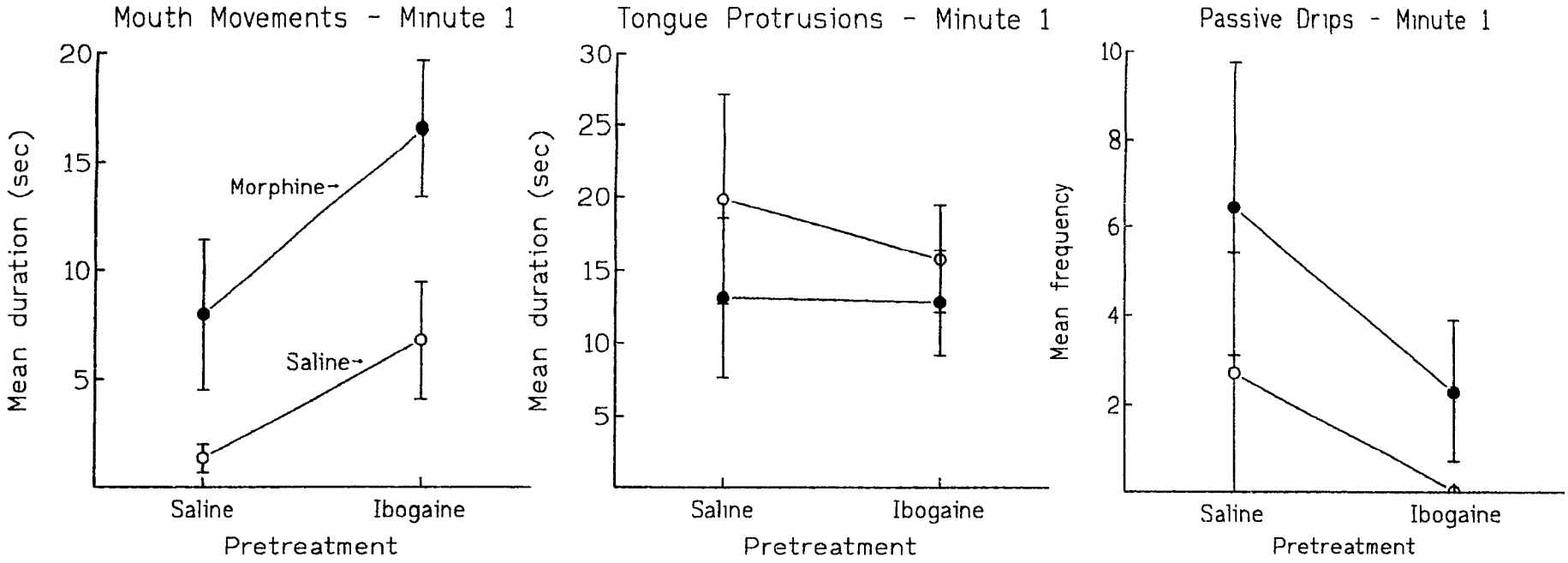

Rearing
Mean frequency
20
15
10
5
Saline
Morphine
Saline
Ibogaine
Pretreatment
Active Locomotion
Mean frequency
30
25
20
15
10
5
Saline
Ibogaine
Pretreatment

Figure 6. Mean (±SEM) duration (sec) or frequency of ingestive reactions and passive drips displayed during the first min of the 5 min intraoral infusion of 10% sucrose solution in Experiment 2.


Mouth Movements - Minute 1
Mean duration (sec)
20
15
10
5
Morphine→
Saline→
Saline
Ibogaine
Pretreatment
Tongue Protrusions - Minute 1
Mean duration (sec)
30
25
20
15
10
5
Saline
Ibogaine
Pretreatment
Passive Drips - Minute 1
Mean frequency
10
8
6
4
2
Saline
Ibogaine
Pretreatment

taste reactions elicited by sucrose solution; the interaction was not significant for any taste reaction.

Figure 7 presents the mean frequency of rearing and active locomotion displayed by rats during the first min of the TR test. The frequency of neither rearing nor active locomotion during min 1 of the sucrose infusion was modified by pretreatment with ibogaine, nor was it affected by treatment with morphine.

Pretreatment-Treatment Body Weight Change

As in Experiment 1, in order to determine whether the ibogaine enhancement of sucrose palatability could be the result of a differential deprivation level during TR testing, a 2 by 2 ANOVA with the factors of pretreatment condition and treatment condition was conducted. The ANOVA revealed only a significant pretreatment effect, $F(1,24)=11.44$, $p<.01$. The mean change in body weight in ibogaine pretreated rats was -4.57 gm, whereas the mean change in body weight in saline pretreated rats was 3.29 gm.

DISCUSSION

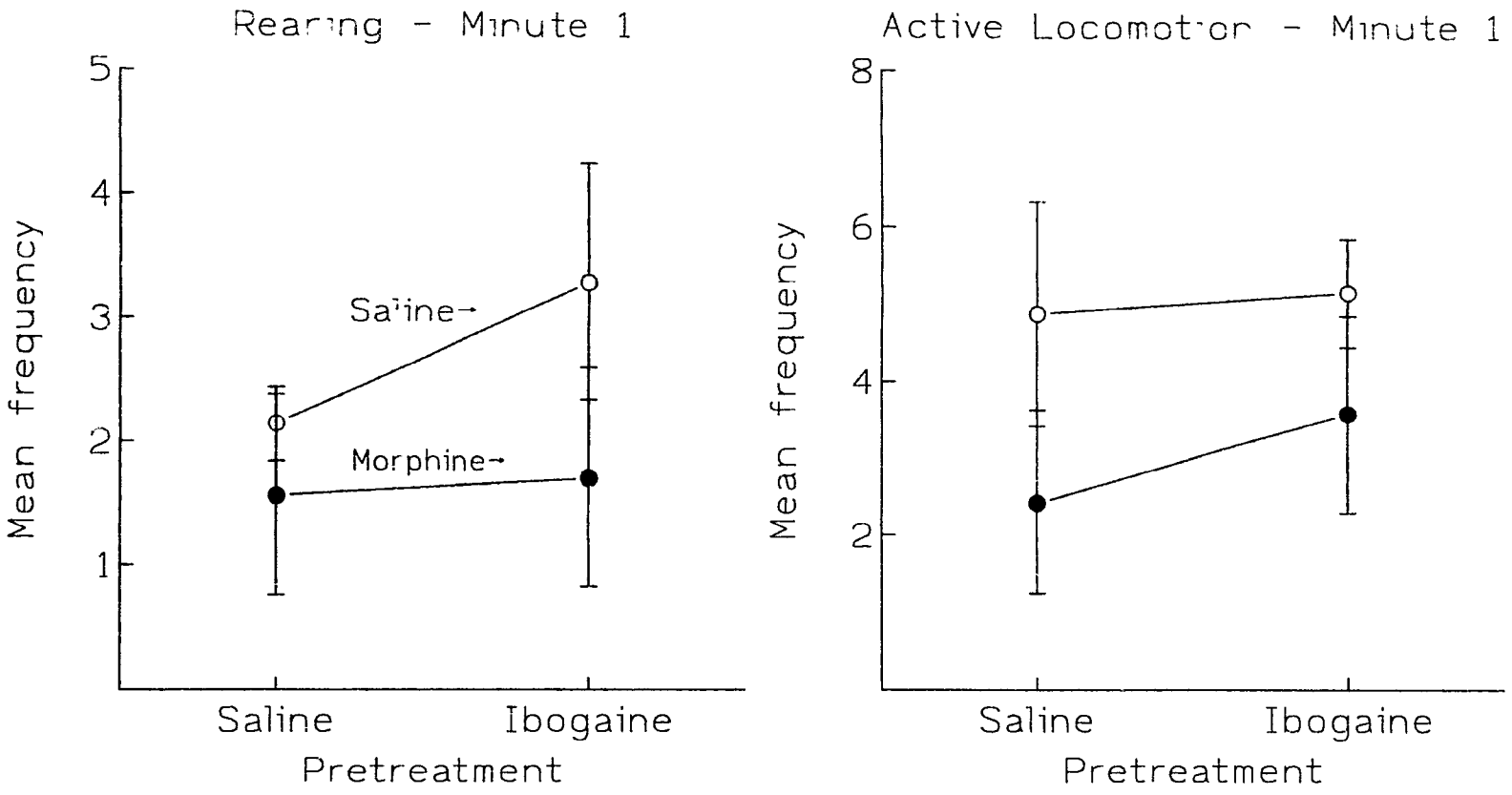
Contrary to our prediction, ibogaine did not modify the ability of morphine to shift sucrose palatability. In fact, only during min 1 of testing did morphine enhance sucrose palatability as evidenced by enhanced mouth movements elicited by 10% sucrose solution. Since this effect was not apparent throughout the 5 min of testing, it is most likely a transient modification on palatability. Interestingly, Doyle et al. (1993) also found that

morphine enhanced the palatability of a 7% sucrose/0.01% quinine mixture solution during a 1 min test. Rideout and Parker (in press) found that morphine enhances the palatability of 2% and 20% sucrose solution during the first 5 min of a 10 min TR test. This prolonged effect of morphine on sucrose palatability was not evident in the present experiment that used 10% sucrose, suggesting that this concentration may not be optimal for investigation of this effect.

These data also suggest that ibogaine may enhance the palatability of sucrose solution. Rats pretreated with ibogaine 24 hr prior to an injection of either saline or morphine spent more time displaying mouth movements during the entire 5 min TR test, and during the first min of the test. However, this effect may be the result of a suppression of food intake during the 24 hr period following ibogaine administration. Ibogaine pretreated rats appeared to be in a greater deprivation state, indicated by less weight gain than saline pretreated rats during the 24 hr pretreatment-treatment interval.

As in Experiment 1, these data indicate that ibogaine pretreatment did not effect the palatability enhancing properties of morphine.

Figure 7. Mean (±SEM) frequency of rearing and active locomotion displayed during min 1 of the 5 min intraoral infusion of 10% sucrose solution in Experiment 2.


Rearing - Minute 1
Mean frequency
5
4
3
2
1
Saline→
Morphine→
Saline
Ibogaine
Pretreatment
Active Locomotor - Minute 1
Mean frequency
8
6
4
2
Saline
Ibogaine
Pretreatment

GENERAL DISCUSSION

The two experiments reported here support earlier findings that morphine enhances the palatability of both quinine (Parker et al., 1992; Clarke & Parker, in press) and sucrose (Doyle et al., 1993; Rideout & Parker, in press) solution. Aversive taste reactions elicited by 0.05% quinine solution in Experiment 1 were significantly suppressed in morphine treated rats. In addition, the neutral/mildly aversive taste reaction of passive dripping was enhanced in morphine treated rats. Furthermore, in Experiment 2, rats treated with morphine spent more time engaged in the ingestive taste reaction of mouth movements during min 1 of the 5 min infusion of 10% sucrose solution.

Pretreatment with ibogaine also appeared to modify quinine and sucrose palatability. Indeed, the effects of ibogaine pretreatment on palatability mimic the effects of morphine. In Experiment 1, the aversive taste reaction of paw pushes was suppressed in rats pretreated with ibogaine 24 hr prior to an injection of saline or morphine. Additionally, in rats pretreated with ibogaine, the frequency of passive dripping was enhanced. In Experiment 2, rats pretreated with ibogaine spent more time displaying the ingestive reaction of mouth movements during the 5 min infusion of sucrose solution. It is surprising that ibogaine, a drug that interferes with the ability of morphine to enhance extracellular DA levels, also enhances palatability. However, analysis of changes in body weights

during the 24 hr pretreatment-treatment interval in both experiments revealed that the ibogaine pretreated rats gained less weight than the saline pretreated rats suggesting that they were in a greater deprivation state. It is possible that the apparent effect of ibogaine on food intake may have influenced the palatability of quinine and sucrose solution. Berridge (1991) has demonstrated that ingestive reactions to sucrose are enhanced in food deprived rats.

The two measures of activity utilized, rearing and active locomotion, were both affected by morphine and ibogaine. Independently, administration of morphine and ibogaine each reduced the frequency of each of the activity measures during the 5 min quinine infusion in Experiment 1. However, administration of morphine or ibogaine had no effect on activity levels in Experiment 2 when the rats were infused with sucrose solution. This discrepancy may be related to the difference in baseline activity levels elicited by infusions of quinine or sucrose solutions. Since rats tend to be more active during quinine infusions than during sucrose infusions, they may be more sensitive to pharmacological effects on activity during exposure to quinine than during exposure to sucrose solution.

Pretreatment with ibogaine 19 hr earlier prevents the morphine-induced enhancement of DA levels in the striatum and nAcc. At this interval ibogaine also attenuates the intravenous self-administration of morphine (Glick et al., 1991) and prevents

the establishment of a one trial morphine-induced conditioned place preference (Parker, Siegel, & Luxton, submitted). Evans and Vaccarino (1990) suggested that the morphine-induced enhancement of consumption (and palatability) is mediated by DA-dependent central reward pathways. Therefore, it was expected that pretreatment with ibogaine 24 hr prior to morphine treatment would prevent the morphine-induced enhancement of sucrose and quinine palatability. The present data suggest that ibogaine does not interfere with the effects of morphine on palatability.

The reason that ibogaine is ineffective in modifying the effects of morphine in the taste reactivity test is not clear. However, in Experiment 2, morphine only weakly enhanced sucrose palatability. Morphine treated rats spent more time displaying the ingestive taste reaction of mouth movements, but only during min 1 of the 5 min TR test. With a more reliable morphine-induced enhancement of sucrose palatability, ibogaine may modulate the effects of morphine. It is also possible, however, that ibogaine interferes specifically with the rewarding properties of morphine that are assessed with the self-administration and place conditioning paradigms. This explanation is however, inconsistent with evidence provided by Evans and Vaccarino (1990) that opiate-induced enhancement of palatability is mediated by activation of reward mechanisms. Further research is necessary to determine the generality of the effects of ibogaine on opiate-induced feeding systems.

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