Pharmacology Biochemistry & Behavior, Vol. 11, pp. 391–394. Printed in the U.S.A.

Non-Specific Enhancement of Ethanol-Induced Taste Aversion by Naloxone

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Received 25 July 1979

MICELI, D., P. MARFAING-JALLAT AND J. LE MAGNEN. *Non-specific enhancement of ethanol-induced taste aversion by naloxone.* PHARMAC. BIOCHEM. BEHAV. 11(4) 391–394, 1979.—The conditioned taste aversion paradigm (CTA) was used to examine the effects of naloxone on ethanol-induced aversion towards a saccharine solution (3 conditioning and 11 extinction trials). Six groups of rats received conditioning trials consisting of two IP injections after saccharine presentation of different combinations of either ethanol (E: 1.75 g/kg), LiCl (L: 1.2 mEq/kg, 0.1 M), naloxone (N: 10 mg/kg) or saline (S); S-S, S-N, E-S, E-N, L-S and L-N. Naloxone by itself produced no aversion to the saccharin flavor. Based on the onset and extinction of aversion, naloxone significantly enhanced ethanol but also LiCl-induced CTA. The comparative data argues in favor of different mechanisms of action (1) between the aversive central effects of ethanol and morphine and (2) between ethanol's acute behavioral effects and negatively reinforcing properties. Enhancement of ethanol and LiCl-induced CTA by naloxone is compatible with hypernociceptive action of the opiate-antagonist and with the pain-modulating role of opiates in the CNS.

Conditioned taste aversion Ethanol Naloxone

IN SPITE of earlier reports [14,28] and the finding that naloxone, a specific opiate receptor antagonist, precipitates withdrawal symptoms in morphine but not ethanol-dependent mice [12], several other lines of evidence suggest that alcohol and opiates share a common mechanism of action in the CNS (see [5] for review). These are based upon both interactions related to the induction of tolerance and physical dependence and between some acute behavioral effects of the drugs. Morphine administration has been shown to inhibit ethanol withdrawal convulsions in mice [7] and concurrent ethanol administration suppresses morphine withdrawal in rats [18]. Elsewhere, tolerance to morphine has been reported in ethanol-dependent animals [31]. Acute behavioral interactions including morphine suppression of voluntary ethanol consumption [15,29] and synergistic potentiation of acute toxicity have also been demonstrated [11,31]. Naloxone appears to modify both the acute and chronic effects of the two drugs in a similar manner [3, 4, 6, 8, 26].

The conditioned taste aversion (CTA) paradigm has also served as a useful model for the study of drug interactions. Morphine-induced taste aversion towards a saccharine solution has been shown to be attenuated by naloxone [19,30] and it consistent with the latter's antagonistic action of some acute behavioral effects of the drug. Ethanol also has been shown to be capable of generating taste aversion towards a saccharine solution [9, 10, 20, 21]. In view of the similar role of naloxone to antagonize ethanol and morphine-related behavioral alterations, the present study was undertaken to examine the effects of the narcotic antagonist on ethanol-induced taste aversion. However, another factor to be considered in naloxone's effects on CTA phenomena is linked to the pain-modulating role of opiates in the CNS as it is well known that naloxone enhances the aversiveness of a variety of nociceptive stimuli [1, 2, 13, 16]. Thus, either attenuation or no change/enhancement of ethanol-induced CTA by naloxone would suggest, respectively, that ethanol and morphine share some common mechanism of action related to their aversive-producing properties or that naloxone's attenuation effects of morphine-induced taste aversion are specific to this drug. Furthermore an enhancement of ethanol-induced taste aversion would indicate a non-specific effect of naloxone; increasing ethanol's negatively reinforcing effects on the CNS, as is the case for other aversive stimuli. For the purpose of comparison, the effects of naloxone on taste aversion produced by another nociceptive agent LiCl [27] was also investigated.

METHOD

The study was performed using 30 male Wistar rats weighing 260 ± 2 g (mean ± SE) at the start of the experiment. They were individually housed in wire-mesh cages where standard lab chow (Pietrement) was available ad lib. A daily 12/12 hr dark/light cycle was maintained and the ambient temperature kept at 20°C. The total fluid intake was restricted to two daily drinking sessions: 30-min a.m. (10:00–10:30) and 2-hr p.m. (15:00–17:00) sessions.

On the first day the rats were water-deprived at 17:00 and starting the next day were habituated to the drinking schedule for the subsequent 5 days on tap water. This was followed by the conditioning and extinction phases of the

experiment when instead of offering water in the a.m. session, a 0.1% saccharine solution was presented on alternate days. Three conditioning trials were performed on the initial saccharine days (Trials 0–2) and consisted of administering two separate IP injections of various solutions on opposite sides of the mid-line, 5–7 min following the saccharine presentation. The animals were divided into 6 groups (N=5) depending on the solutions injected; either saline-saline (S-S), saline-naloxone (S-N), ethanol-saline (E-S), ethanol-naloxone (E-N), LiCl-saline (L-S) or LiCl-naloxone (L-N); administered in volumes of 3.0 and 0.5 ml, respectively.

Ethanol was administered in a dose of 1.75 g/kg prepared from 95% ethanol by dilution in saline. The LiCl dose was 1.2 mEq/kg (0.1 M) in saline. These moderate doses were selected on the basis of previous experiments (unpublished data) showing that they were aversive however would not induce total aversion after only one conditioning trial. A 10 mg/kg dose of naloxone was used after preliminary tests had shown this dose not to produce any aversion after 3 conditioning trials. The extinction phase extended over 11 trials (Trials 3–13) during which time no injections were performed after the a.m. saccharine sessions. No injections were given following the alternating a.m. or after p.m. water presentations. The volume of fluid consumed during the alternating a.m. saccharine and water sessions was recorded throughout the experiment.

RESULTS

The mean volumes of saccharine intake recorded during the initial 3 conditioning trials 0–2 and subsequent extinction trials 3–13 are shown in Fig. 1. Mann-Whitney U tests were performed for statistical analyses with significance at the $p<0.05$ level. No significant differences in saccharine consumption was observed between S-S and S-N animals throughout all trials. Compared to S-S controls, all groups administered either ethanol or LiCl, in combination with either naloxone or saline showed aversion to the saccharine solution after successive conditioning trials.

The E-S group drank significantly lower quantities of saccharine on the 2nd trial whereas the E-N group showed aversion after only one pairing of the UCS with the flavor (Trial 1). Both LiCl groups (L-S and L-N) showed significant aversion on the 1st trial. During the extinction phase, the E-S group consumed significantly similar amounts of saccharine as the saline controls (S-S) from the 6th trial onwards. A delay in extinction was noted in the E-N group where the intake only attained extinction levels on Trial 12. In the LiCl groups, naloxone also produced a delay in the extinction of taste aversion. Extinction occurred on the 8th trial in the L-S group and on the 11th trial in the L-N group. A comparison of E-S and L-S saccharine intakes indicated that, at the doses used, LiCl acted as the stronger negative UCS. This was demonstrated by the more rapid onset of aversion, the lower volumes of saccharine consumed and the longer delay in extinction. The levels of water consumption recorded during the same 30-min a.m. sessions on alternate days showed no significant variation across groups and trials.

DISCUSSION

The results showed that, at a dose of 10 mg/kg, naloxone by itself did not produce a significant taste aversion after 3 pairings with the saccharine flavor. Preliminary tests had shown that naloxone doses of 1.0 and 5.0 and 10.0 mg/kg all failed to generate aversion after 3 conditioning trials. Taste aversions to naloxone alone have been observed in previous investigations with experimental designs similar to that of the present study however using a greater number of conditioning trials. Le Blanc and Cappell [19] using a 12.96 mg/kg dose of naloxone in a first experiment noted aversion on the 6th conditioning trial whereas in a second experiment aversion was observed in the 3rd trial using a dose of 9.6 mg/kg. Van der Kooy and Phillips [30] reported a punishing effect of 10.0 mg/kg naloxone on the 3rd trial and animals receiving the latter dose drank significantly less saccharine than those administered lower doses (5.0 and 7.5 mg/kg) on the 5th conditioning trial.

Based on the number of conditioning trials required for the onset of aversion and test trials for extinction, naloxone significantly enhanced CTA induced by both ethanol and LiCl. The aversion lasted over trials comprised between 2–6 and 1–12 for the E-S and E-N groups; 1–8 and 1–11 for the L-S and L-N groups, respectively. Thus naloxone combined with ethanol precipitated aversion 1 trial earlier and delayed extinction by 6 trials. Although aversion was observed on the 1st trial for both LiCl groups, naloxone extended the extinction period by 3 trials (L-N) compared to the saline control group (L-S). The enhancing effect on aversion could not be attributed to additive negative reinforcement by naloxone as no aversive effects of the latter could be demonstrated using the present experimental conditions.

These findings are at variance with those which have reported an attenuation of morphine-induced CTA by naloxone [19,30] and argue in favor of a different biochemical mechanism of action related to the aversive effects produced by ethanol and morphine. Nevertheless, various processes involved in the development of dependence and in the acute behavioral effects of both ethanol and morphine appear to be naloxone-sensitive. Naloxone is capable of blocking the acute effects and development of tolerance [26] and physical dependence to opiates [8]. Administered during an ethanol inhalation treatment, naloxone has been shown to inhibit withdrawal convulsions in mice [4]. Naloxone also antagonizes morphine-induced analgesia [16, 17, 26] and changes in locomotor activity [3] as well as narcosis [5,6] and excitatory effects on locomotor activity [25] produced by ethanol. Ethanol has been shown to reduce naloxone hyperalgesia [2]. Elsewhere a dose-dependent effect of the narcotic antagonist has been reported in mice; with a 5 mg/kg dose inhibiting ethanol narcosis whereas higher doses (10 mg/kg) potentiate the narcosis [5]. Some negative data has been obtained in our laboratory using the drinking test [23,24] for measuring behavioral deficits produced by ethanol (1.75 and 2.75 g/kg doses). Naloxone was found to affect neither the initial sensitivity towards ethanol nor the subsequent increase in tolerance following chronic ethanol exposure [22].

In the case of morphine, naloxone would appear to specifically antagonize both the behavioral manifestation of its acute actions and the aversive effects produced by the drug. In contrast, and in view of the present findings, there appears to be a discrepancy between naloxone's capacity to attenuate some of the latter-mentioned behavioral effects produced by ethanol and the enhancement of its negatively reinforcing properties as demonstrated using the CTA model. This suggests that naloxone differentially influences these two components of the response to ethanol. Although

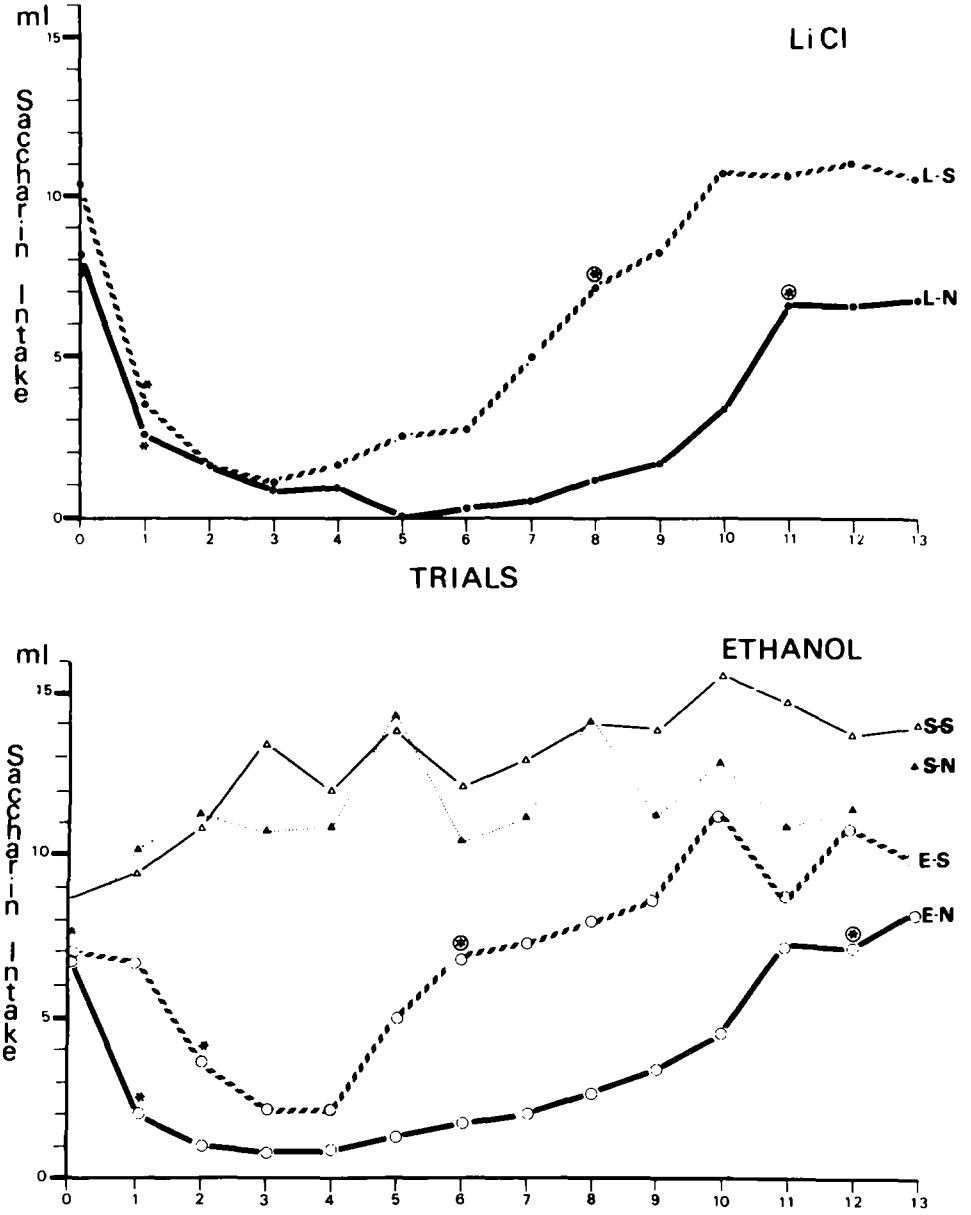

FIG. 1. The mean volumes of saccharine (0.1%) intake during the conditioning (Trials 0–2) and extinction trials (Trials 3–13) for the 6 groups tested with either ethanol (E: 1.75 g/kg), LiCl (L: 1.2 mEq/kg, 0.1 M), or saline (S) and naloxone (N: 10 mg/kg) or saline. Open and circled asterisks indicate onset and extinction of aversion, respectively, in relation to S-S control levels of consumption (Mann-Whitney U test).

both reactions are naloxone-sensitive, their underlying mechanisms, that is, those determining the acute behavioral effects of the drug and those concerned with the affective response to the agent's aversive qualities would appear to be distinct. Such a dualistic sensation/perception concept has been proposed to explain some effects of naloxone on different responses elicited by nociceptive stimuli [1, 2, 13, 16]. Naloxone effects on escape and paw-lick latencies using the hot-plate test in mice and rats have shown antagonisms in regards to the former parameter (affective component) and a lesser or no apparent effects on the latter (sensory) response [1, 13, 17].

Furthermore, the present results indicated that the enhancement by naloxone of ethanol-induced CTA is not specific to this drug as it also occurred with LiCl. These comparable effects can be explained by naloxone's demonstrated non-specific and hypernociceptive action on aversive stimuli and is compatible with the pain-modulating role of opiates in the CNS.

ACKNOWLEDGEMENTS

Naloxone HCl was a gift from Endo Laboratories (NY). This work was supported by an IREB research grant.

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