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Psychology

Effects of naloxone on experimentally induced ischemic pain and on mood in human subjects

(endorphin/anxiety/opiates/enkephalin)

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ABSTRACT **The hypothesis that painful stimuli activate the endogenous opioid (endorphin) system in humans was tested by examining the effect of the opiate antagonist naloxone on experimentally induced ischemic pain and on subjective mood ratings. Intravenous injections of saline or naloxone hydrochloride (2 and 10 mg) were administered under double-blind conditions to 12 subjects. Naloxone did not affect the pain ratings. However, a significant dose-related effect of naloxone on tension–anxiety was found, suggesting that the endorphins, like exogenously administered opiates, may have antianxiety properties.**

The discovery of endogenous substances with opioid activity (endorphins) in several species, including man (1–6), raises questions about what role these substances normally play. Since narcotic antagonists block and reverse the effects of opiates, the administration of these antagonists should similarly affect behavior mediated by endorphins. However, when administered to animals or humans who have not received an opiate, the "pure" antagonists, naloxone and naltrexone, appear to have no effect, suggesting that the endorphin system is not continuously active, but only activated in certain internal or environmental conditions. In both rats (7) and humans (8) naloxone diminishes analgesia produced by electric brain stimulation; however, it does not modify hypnotic analgesia in man (9). Naloxone does not alter threshold for escape from a noxious stimulus in trained rats (10), but it reduces the latency for escape behavior when mice or rats are exposed to a noxious stimulus for the first time (11, *).

The purpose of the present study was to determine if painful stimuli activate the endorphin system in humans by examining the effect of naloxone on experimentally induced ischemic pain. In addition, recognizing the affective changes that result from administration of exogenous opiates, we hypothesized that naloxone would accentuate the stressful nature of the pain, resulting in a greater increase in anxiety, hostility, and depression than in the same situation without naloxone.

METHODS

Subjects. The subjects were six male and six female staff members who volunteered to participate in the study. The median age was 28 years. Informed consent was obtained.

Drugs. Naloxone hydrochloride was dissolved in 0.9% saline solution at 2 and 10 mg/ml. At each session an intravenous injection of 1 ml of one of these solutions or of saline alone was administered at the rate of 1 ml/min. The sequence of drug administration was counterbalanced with the restriction that, as a precautionary measure, the 10 mg dose of naloxone was never administered first. Therefore, subjects were randomly assigned to one of the following sequences: saline, 2 mg naloxone, 10 mg naloxone; saline, 10 mg naloxone, 2 mg naloxone; 2 mg naloxone, saline, 10 mg naloxone; 2 mg naloxone, 10 mg naloxone, saline. Solutions were contained in identical vials coded by two-digit random numbers. They were prepared by a person who had no contact with the experiment. The experimenter (P.G.) not only was blind to the coding system, but also was not aware of the restrictions concerning the sequence of naloxone doses. The physician (A.G.) giving the injections was also blind to the identity of the solutions.

Procedures

Pain Production. Experimental pain was produced using the submaximum effort tourniquet technique developed by Smith *et al.* (12). As a precaution, subjects were first tested with a petechiometer to be sure that capillary resistance was high enough to avoid damage by the tourniquet procedure (13). To promote venous drainage, the arm was extended toward the ceiling and an elastic bandage was wrapped around the forearm. The tourniquet, a sphygmomanometer cuff, was placed above the elbow and inflated to 250 mm Hg. (33 kPa). The bandage was removed, the arm was lowered, and the subject then exercised by squeezing a hand dynamometer, loaded to 12 kg, 20 times. Each squeeze lasted 2 sec and was followed by a 2 sec rest. Following this exercise, the arm rested on a table in front of the subject, and the subject was cued by a tape recorder at 30 sec intervals to rate on a 10-point scale the pain experienced in the occluded arm. The subject was instructed that a "0" rating indicated that the arm did not hurt at all, a "1" rating indicated just noticeable pain, up through moderate and severe pain to a "10" rating, which indicated that the pain was unbearable and the subject wished to end the experiment. The pressure of the cuff was released either when the subject gave a "10" rating or after the arm had been occluded for 10 min. Subjects were informed that, in order to participate in the study, they must be able to endure the pain for at least 3 min.

Mood Ratings. Prior to the injections, and again at the end of each session, subjects reported how they were feeling "right now" using the Profile of Mood States questionnaire (14). This questionnaire consists of a list of 65 adjectives, which the subject rates on a 5-point scale (0 = not at all, 4 = extremely). Scores for seven affective states were computed: friendliness, confusion–bewilderment, vigor–activity, depression–dejection, anger–hostility, fatigue–inertia, and tension–anxiety.

Experimental Design. Drugs were administered under double-blind conditions. Subjects were tested at three sessions at 24 hr intervals. Within each session, pain was produced twice—prior to the injection, and then again 5 min after the injection. Pain always was produced in the dominant arm first, and then in the nondominant arm. A pilot study failed to indicate any systematic difference between the two arms with respect to pain tolerance. The Profile of Mood States questionnaire was administered after the first production of pain

* P. Grevert and A. Goldstein, unpublished data.

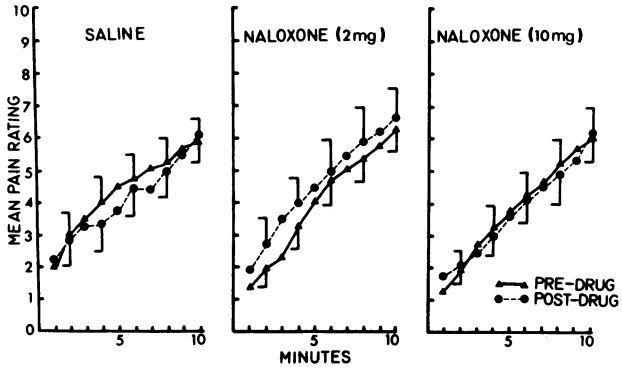

FIG. 1. Failure of naloxone to affect ratings of ischemic pain: mean pain ratings (±SEM) by 1 min intervals, before (▲) and after (●) intravenous injections of saline or naloxone (2 mg and 10 mg). $n = 12$.

(just prior to the the injection) and again after the second production of pain, following drug administration. At the end of each session, subjects were asked to judge if an active drug or saline had been administered that day.

Statistical Analysis. Pain and mood ratings were analyzed using a a one-factor (drugs: saline, 2 mg naloxone, 10 mg naloxone) repeated measures analysis of variance (15). Of specific interest was whether the changes between pre- and post-drug scores following the naloxone injections differed significantly from the changes that occurred when saline was administered. These planned comparisons were tested using a single degree of freedom F-test (15). Prior to analysis, the homogeneity of variance assumption was tested using the statistic F_{max}, the ratio of the highest treatment variance to the lowest. If the variances differed significantly, drug effects were tested using the Friedman two-way analysis of variance by ranks (16). A result was considered significant at $P < 0.05$.

RESULTS

Pain ratings

Fig. 1 presents the mean pain ratings over the 10 min rating period before and after injection of saline or naloxone. There was no significant drug effect. For each subject the change (post-drug minus pre-drug) in pain score (i.e., the sum of the pain ratings over the 10 min period) also was computed. There was an increase in pain scores after 2 mg of naloxone ($\bar{X} = 10.5$) and a decrease after saline ($\bar{X} = -4.8$) or 10 mg of naloxone ($\bar{X} = -2.2$), but these differences were not significant ($F_{2,22} = 2.34$, $P = 0.12$). No significant sessions effect of saline or 2 mg of naloxone was found when the change in pain score during the first session was compared to the change in scores during the other two sessions. Similarly, there were no sex differences.

To determine if naloxone affects the buildup of pain, the difference (post-drug minus pre-drug) in the time to reach "moderate" and "severe" pain was computed. Moderate pain was defined as the midpoint of the ratings given by a subject during each production of pain, and severe pain was the highest rating reported. Again, there were no significant drug effects.

Mood ratings

Fig. 2 presents the mean change in score on the seven scales of the Profile of Mood States. No significant drug effect was found on five of the six affective states: friendliness, confusion–bewilderment, vigor–activity, depression–dejection, and fatigue–inertia. Changes in subjects' reports of anger–hostility were in the hypothesized direction and were dose-related, but were not significant. Scores increased when pain was produced after 10 mg of naloxone and decreased the most when pain production followed saline administration. A significant dose-related drug effect was found on changes in tension–anxiety scores ($F_{2,22} = 6.00$, $P < 0.01$). The large decrease in tension–anxiety that occurred when saline was administered differed significantly both from the slight decrease after 2 mg of naloxone ($F_{1,10} = 6.38$, $P < 0.05$) and from the increase after 10 mg of naloxone ($F_{1,10} = 11.14$, $P < 0.01$) (Table 1). No sex differences were found.

Because the naloxone doses were larger than usually administered to humans, we adopted the safety precaution of never administering the 10 mg dose prior to the 2 mg dose. Consequently, the design was unbalanced, in that a reduction in anxiety in sessions 2 and 3 as compared with session 1 could be confounded with a 10 mg naloxone effect. Indeed, as shown in Table 1, the mean pre-drug anxiety score was lowest in subjects receiving the 10 mg dose. Accordingly, we analyzed the data for the balanced 2 mg versus saline comparison (each treatment appearing twice at session 1, once at session 2, and once at session 3). Post- drug scores were compared (2 mg naloxone minus saline), subject by subject. The mean difference was 1.58 ± 0.63, significantly different from zero ($P < 0.05$) by t-test. The same computation for the unbalanced 10 mg versus saline comparison yielded 1.42 ± 0.50, again significantly different from zero. Thus, despite the reduction of pre-drug anxiety in the latter sessions, naloxone at both doses increased the post-drug anxiety score as compared with saline.

Subjects were unable to differentiate saline from naloxone correctly. Only 63% of the responses were correct when subjects were asked at the end of each session if an active drug had been administered.

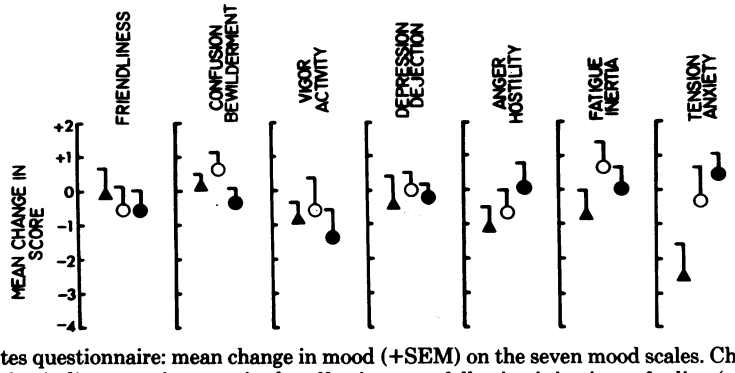

FIG. 2. Profile of Mood States questionnaire: mean change in mood (+SEM) on the seven mood scales. Change is the post-drug score minus the pre-drug score. A positive value indicates an increase in the affective state following injections of saline (▲), 2 mg of naloxone (○), or 10 mg of naloxone (●). $n = 12$.

Table 1. Effect of naloxone on tension–anxiety scores

	Saline			2 mg naloxone hydrochloride			10 mg naloxone hydrochloride		
Subject	Pre-drug	Post-drug	Difference	Pre-drug	Post-drug	Difference	Pre-drug	Post-drug	Difference
S001	3	2	−1	4	7	3	4	5	1
S002	10	4	−6	7	6	−1	6	8	2
S003	8	2	−6	14	5	−9	9	4	−5
S004	15	9	−6	9	13	4	9	11	2
S005	10	3	−7	2	2	0	2	2	0
S006	6	5	−1	10	9	−1	4	6	2
S007	5	4	−1	4	4	0	6	8	2
S008	1	2	1	1	2	1	1	1	0
S009	3	0	−3	1	3	2	1	2	1
S010	9	10	1	9	9	0	9	10	1
S011	2	2	0	4	1	−3	2	2	0
S012	3	1	−2	1	2	1	2	2	0
Mean	6.3	3.7	−2.6	5.5	5.3	−0.3	4.6	5.1	+0.5

The table shows tension–anxiety scores from the Profile of Mood States (14), obtained immediately after the ischemic pain procedure. With saline, the reduction after the second production of ischemic pain was significantly different from zero ($P < 0.05$). This reduction was blocked by naloxone at 2 mg ($P < 0.05$) and at 10 mg ($P < 0.01$).

DISCUSSION

Naloxone did not affect subjects' ratings of the experimentally induced ischemic pain. This result is in agreement with the recent finding of El-Sobky *et al.* (17), using electric shocks as painful stimuli and much lower doses of naloxone. However, we found a significant effect of naloxone on mood. The reduction in tension–anxiety that occurred at the completion of pain production when saline was administered did not occur after naloxone, and this effect was dose-related. A similar but not statistically significant effect of naloxone on anger–hostility also occurred. Resnick *et al.* (18) found that seven of 37 ex-addicts reported feeling nervous and irritable after receiving the first dose of oral naltrexone, but these symptoms disappeared in a few days and were attributed to a protracted abstinence syndrome. Gritz *et al.* (19), using the Multiple-Affect Adjective Checklist to systematically assess mood changes in eight ex-addicts after oral doses of naltrexone (20, 40, 80, and 160 mg), found no effect on anxiety. Thus, naloxone or naltrexone administered in a neutral environment does not affect anxiety. However, in the present study, naloxone blocked the reduction in tension–anxiety that normally occurred after the stressful pain production was terminated. This result suggests that the endorphin system may be activated during stress, and that the endorphins, like exogenously administered opiates, may have antianxiety properties. Further research on the effects of naloxone on anxiety is needed to test this hypothesis.

If the endorphin system was activated, why was there no significant effect on the pain ratings? For an antagonist to produce a pharmacologic effect, two preconditions must be met. *First,* the antagonist must be present at sufficient concentration to occupy the receptors. The naloxone doses (2 and 10 mg) given here by the intravenous route were certainly more than sufficient. For comparison, a dose of only 0.4 mg will, within a few minutes, arouse a comatose person suffering from an opiate overdose; and a dose of 0.2 mg reversed analgesia produced by electrical stimulation in the human brain (8). *Second,* the corresponding agonist (in these experiments endorphin) must be present and acting at the receptor sites; otherwise the antagonist could do nothing. This principle underlies our attempt to activate endorphin release. We inflicted a non-trivial painful stimulus of several minutes duration, twice at each session—initially, to obtain baseline response data and (we hoped) to initiate endorphin release, then again 15 min later, after saline or naloxone administration.

It is interesting that there was no change in pain response under the saline condition, and also no effect of naloxone upon pain. However, there was a definite decrease in tension-anxiety at the termination of the second ischemic pain procedure, and this change was blocked by naloxone. Evidently, then, the mood change was a more sensitive indicator of endorphin release than was the pain procedure. Possibly significant reduction of pain requires greater activation of the endorphin system(s) than does mood alteration. Alternatively, the several components of endorphin action (such as effects on pain and on mood) may be independently controlled.

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