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Cardiovascular Changes During Morphine Withdrawal in the Rat: Effects of Clonidine

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BUCCAFUSCO, J. J. *Cardiovascular changes during morphine withdrawal in the rat: Effects of clonidine.* PHARMACOL BIOCHEM BEHAV **18**(2) 209–215, 1983.—Arterial blood pressure and heart rate were measured in unrestrained rats as an index of the autonomic component of the morphine withdrawal syndrome. Physical dependence was produced by a constant infusion of morphine at increasing doses over 7 days. Signs of physical dependence observed during abrupt withdrawal included classical behavioral symptoms such as withdrawal body shakes (WBS) and increased autonomic responsiveness which was indicated by a sustained increase in mean arterial pressure (MAP) up to 23 mmHg. Injection of naloxone in morphine dependent rats also evoked a dose-related increase in MAP to about 40 mmHg. The antiwithdrawal effects of clonidine were tested in this model by pretreating dependent rats with this agent (6–60 μg/kg). Clonidine inhibited the pressor response produced by naloxone by 23–60%. These findings indicate that the increase in MAP during opiate withdrawal provides an objective and quantitative index of the intensity of the narcotic withdrawal syndrome in dependent rats.

Morphine withdrawal Physical dependence Blood pressure Naloxone Clonidine

EXAGGERATED discharges of the sympathetic nervous system, which occur during withdrawal in a subject who is physically dependent upon narcotics, produces many of the characteristic clinical signs. Mydriasis, perspiration, tachycardia and elevated arterial pressure are symptoms of morphine withdrawal that are mediated through the sympathetic nerves. Increases in arterial pressure by as much as 30 mmHg may occur at the peak intensity of withdrawal symptoms [15, 19, 25]. Also, marked increases in blood pressure have been observed in opiate-dependent [34] and narcotic overdose [4, 5, 6] patients following administration of the narcotic antagonist, naloxone.

The increase in arterial pressure which accompanies narcotic withdrawal in man originally was employed as an important criteria in the determination of the intensity of abstinence [13]. However, there have been few attempts to study these cardiovascular changes in experimental animals. The rat is widely used to study the effects of narcotic agonists and antagonists, and several techniques have been described for the production of physical dependence and the quantitation of withdrawal phenomena in this animal model [1, 8, 24, 33]. These methods of quantitation rely primarily on the appearance of several characteristic behavioral changes which are then scored for frequency and/or intensity by the observer. The purpose of this study was to determine whether measuring cardiovascular changes in addition to the behavioral signs will provide a more objective and quantitative measure of morphine withdrawal intensity.

The present study was designed to measure and compare the cardiovascular and behavioral changes produced during spontaneous or naloxone-precipitated abstinence. Rapid morphine dependence was produced by constant infusion of the narcotic through a permanently implanted arterial catheter, and all measurements, before and during withdrawal, were made in freely-moving animals. In addition, the effects of clonidine were examined on the cardiovascular and behavioral responses evoked by naloxone in the morphine dependent rat since clonidine is known to exert an antiwithdrawal action both clinically [3, 10, 32] and in morphine dependent rats [7, 29].

METHOD

Male, Wistar rats weighing 280–380 g at the time of the experiment were obtained from Harland Industries, Inc., Indianapolis, IN and housed in an environmentally-controlled room in a 12 hr light-12 hr dark cycle, Food (Wayne Lab Blocks) and tap water were supplied on an unlimited basis. Under methohexital anesthesia (65 mg/kg, IP) a midline abdominal incision was made and the viscera displaced laterally to expose the abdominal aorta and left iliac artery. Surrounding fat and fascia were cleaned from both vessels with a cotton-tipped applicator. The iliac artery was lifted up on a pair of fine forceps to occlude blood flow and a small nick was placed distally. A 50 cm length of polyethylene (PE) 50 tubing (Clay Adams, Parsippany, NJ) was filled with heparinized saline (20 units/ml) and inserted into the nick a distance of 15–20 mm so that the tip of the catheter was situated in the abdominal aorta below the origin of the renal arteries. The opposite end of the tubing was plugged with a 23 gauge stainless steel stilette and threaded through a 20 cm, 15 gauge trochar. The trochar was inserted into the soas

TABLE 1

MEAN ARTERIAL PRESSURE, HEART RATE AND THE FREQUENCY OF WITHDRAWAL BODY SHAKES FOLLOWING ABRUPT MORPHINE WITHDRAWAL

Hours Following Withdrawal	Last Prewith-Drawal Value	7	8	9	10	11	12	14	24
Mean Arterial Pressure (mmHg)	103 ± 3	118 ± 6*	115 ± 5	121 ± 5*	122 ± 6*	118 ± 5*	124 ± 5*	126 ± 5*	120 ± 3*
Heart Rate (beats/min)	374 ± 10	417 ± 16	390 ± 17	398 ± 16	385 ± 16	383 ± 14	376 ± 13	389 ± 14	418 ± 16
Body Shakes (per 1 hr interval)	0	7 ± 1	16 ± 3	16 ± 3	22 ± 4	20 ± 4	12 ± 3	21 ± 5	

Each mean is the average of 6–8 determinations.
*=Significantly different from prewithdrawal value, $p<0.05$.

muscle towards the back, under the skin to emerge at the nape of the neck. The trochar was removed through the neck incision and the catheter inserted through a stainless steel anchor button connected to a 30 cm spring support (Instech Laboratories, Philadelphia, PA). The anchor button was implanted below the skin and attached with stainless steel suture. Following surgery rats were returned to individual home plastic cages 45×25×20 cm. The spring support and arterial catheter were connected to a cannula swivel (Instech Laboratories) mounted 30 cm above the floor of the cage. This system permitted unrestricted movement of the animal to all areas of the cage. Aseptic conditions were not necessary to perform the surgical procedures, however, all implants and surgical instruments were soaked in 70% ethanol and each rat received prophylactic intramuscular injection of 60,000 units of Penicillin G. Rats were constantly infused with heparinized saline (20 Units/ml) at the rate of 8.64 ml/day to maintain patency of catheter.

One day following surgery, a heparinized saline solution of morphine sulfate was placed in a plastic 30 ml syringe which was inserted into a constant speed syringe pump (Razel Co., Stamford, CT). Morphine dependence was produced according to the schedule described by Fielding and coworkers [7] for intravenous infusions. On the day following surgery rats were infused through the arterial catheter at the rate of 0.36 ml/hr to give a total dose of 35 mg/kg/day of morphine sulfate. The concentration of morphine sulfate was adjusted each morning of the next 3 consecutive days to give, respectively, 50, 70 and 100 mg/kg/day. The last concentration was then maintained for 4 days before withdrawal.

Abrupt withdrawal was initiated by replacing the maintenance dose of morphine sulfate with heparinized saline at 2400 hours. Precipitated withdrawal was produced by rapid injection of naloxone hydrochloride (0.05–5 mg/kg) through the arterial catheter. Naloxone-precipitated withdrawal was always elicited between 1000 and 1400 hours of the eighth day of morphine infusion.

Arterial pressure and heart rate were recorded from freely-moving rats through the previously implanted aortic catheters by connecting the arterial line to a Statham, P23Db transducer coupled to a Beckman R12 polygraph recorder. Data are reported as mean arterial pressure (MAP) computed according to: MAP=diastolic + (systolic-diastolic)/3. Heart rate was monitored by a Beckman 9857B cardiotachometer which was triggered from the pressure pulses. The arterial catheters usually remained patent for at least 2 weeks if left relatively undisturbed, while more frequent blood pressure recordings often shortened the catheter's useful life. Arterial pressure and heart rate were recorded between 1000 and 1400 hours on the day before, and on each day following the start of morphine infusion for at least 20 min to obtain a stable baseline. All changes in MAP and heart rate are expressed as the difference between the prewithdrawal baseline value and the peak level of each parameter reached during the withdrawal period.

While several behavioral effects of morphine withdrawal were noted, only the characteristic "wet dog body shakes" were counted. Data is presented as the number of shakes recorded per one hour interval. All behavioral data were measured while continuously recording cardiovascular parameters.

This study was designed to measure the withdrawal-induced changes in 3 groups of morphine dependent rats: (1) A group abruptly withdrawn from morphine and the abstinence symptoms observed for 24 hr. (2) A group in which the withdrawal syndrome was precipitated by intraarterial injection of naloxone (0.05–5 mg/kg) and the abstinence symptoms observed for 1 hr. (3) A group in which clonidine 6–60 μg/kg was administered 20–40 min before withdrawal was precipitated with naloxone (5 mg/kg).

The results of two groups of observations were compared using Student's t-test for unpaired data. Comparison of more than two groups of observations was made using a one-way analysis of variance. A p-value less than 0.05 was accepted as a significant difference.

Morphine sulfate was obtained from Mallinkrodt, Paris, KY. Naloxone hydrochloride was generously supplied by Endo Laboratories, Garden City, NY. All drug doses will refer to their respective salts.

RESULTS

Rats chronically infused with morphine to produce physical dependence exhibited no uncharacteristic behavioral changes as long as the administration of narcotic was main-

TABLE 2

EFFECT OF NALOXONE ON MEAN ARTERIAL PRESSURE, HEART RATE AND THE FREQUENCY OF WITHDRAWAL BODY SHAKES IN MORPHINE-DEPENDENT RATS

Dose Naloxone (mg/kg, IA)	Prenaloxone Map*	Change in Map	Prenaloxone hr†	Increase in hr	Decrease in hr	WBS‡
0.05 (5)	104 ± 5	26 ± 6	384 ± 12	84 ± 14	75§	35 ± 10
0.5 (5)	100 ± 6	33 ± 4	388 ± 17	80 ± 15	53 ± 15	27 ± 2
5.0 (20)	105 ± 2	43 ± 2¶**	383 ± 8	100 ± 7	77 ± 13	9 ± 2¶**

*Mean arterial pressure (mmHg).
†Heart rate (beats/min).
‡Withdrawal body shakes per hr.
§A decrease in hr was observed in 2 (mean) of the 5 animals.
¶Significantly different from the 0.05 mg/kg value.
**Significantly different from the 0.5 mg/kg value.
The numbers in parentheses represent the number of experiments (animals).

tained. Morphine treated animals, however, were observed to bite or gnaw at their swivel spring support more often than saline infused rats. Over the week of morphine infusion mean arterial pressure (MAP) and heart rate (HR) showed no statistically significant change from preinfusion values (108±2 mmHg and 370±9 beats/min, respectively). A few preliminary experiments revealed no significant behavioral or cardiovascular changes until approximately 7–8 hr following abrupt withdrawal of the morphine infusion. Observations of 8 animals from 7–24 hr after withdrawal revealed many of the characteristic signs of morphine abstinence in rats, including, ptosis, writhing, piloerection and "wet-dog-like" body shakes. Withdrawal body shakes (WBS) were counted and their frequency (number counted/hr) during the observation period are listed in Table 1. WBS occurred most frequently between 10 and 12 hr following withdrawal. In addition to these behavioral changes, MAP was consistently elevated with respect to the prewithdrawal value for 9–24 hr following withdrawal (Table 1). The maximum increase, 23 mmHg, was reached by 14 hr. In contrast, HR did not change significantly over the observation period, although the mean values at each time interval were consistently higher than the prewithdrawal mean up to 44 beats/min. In addition to the changes in MAP noted above, rats withdrawing from morphine exhibited brief phasic increases in MAP and HR. Usually between 8–13 hr following withdrawal sharp increases in MAP and HR up to 30 mmHg and 130 beats/min, respectively, were observed to last 1–2 min. These increases occurred at a frequency of 4–6/hr, often coinciding with several WBS. No attempt was made to quantitate these brief cardiovascular changes.

Intraarterial injection of naloxone, 0.05–5 mg/kg, in morphine dependent rats produced a characteristic behavioral syndrome in all rats. Immediately following injection rats usually exhibited a few WBS, however, a period of behavioral excitation was evident with the animal circling about the cage and attempting to escape by jumping and often clinging to the cage top. WBS were not observed during this period which lasted about 10–15 min. Thereafter, animal behavior was subdued with occasional WBS and/or teeth chattering until the end of the experiment (1 hr after naloxone). As indicated in Table 2, the frequency of WBS was inversely proportional to the dose. It was noted that the frequency of WBS fell as the behavioral excitatory phase (and frequency of escape attempts) increased with dosage. In addition to the behavioral changes, naloxone produced an immediate, sustained and dose-related maximal increase in MAP up to 43±2 mmHg (Table 2). MAP usually reached its highest point within 1 or 2 min following naloxone. The duration of this response, however, was 13±7 to 26±5 min, depending upon the dose. Concomitant with the increase in MAP following naloxone was a biphasic effect on HR which was more prominent with the two higher doses of naloxone, i.e., an increase of 80–100 beats per min followed about 5 min later by a return to prewithdrawal levels and a subsequent decrease of 50–80 beats/min which often continued for the remainder of the experiment. Although the magnitudes of these HR changes did not exhibit a dose dependency, they appeared to parallel the behavioral effects, with the initial increase in HR associated with the period of behavioral and motor excitement, and the secondary bradycardia associated with inactivity. The elevation in MAP, however, was maintained at the same level over both phases of behavior and motor activity changes and appeared to be independent of the latter. Naloxone injection up to 5 mg/kg in control, untreated animals produced no significant effect on MAP and HR. Representative cardiovascular changes produced by 5 mg/kg of naloxone in an untreated and morphine dependent rat is presented in Fig. 1. Another feature of the naloxone-induced increase in MAP is that the response is reproducible in its time course and magnitude if the animal is returned to morphine infusion (100 mg/kg/day) for an additional 48 hr after the first naloxone injection and then given a second naloxone injection. In a group of 6 animals so treated, the pressor response to 5 mg/kg of naloxone was 36±2 mmHg, and following 48 hr of morphine infusion MAP was again at prewithdrawal levels, and a second injection of naloxone elicited a pressor response of 39±2 mmHg.

To determine whether the pressor response produced by naloxone was mediated primarily via the sympathetic nerves, 3 dependent rats were pretreated with the alpha receptor blocking drug, phentolamine (3 mg/kg, IA) 15 min before receiving naloxone (0.5 mg/kg, IA) and 3 rats were pretreated with saline. Phentolamine pretreatment reduced the magnitude of the naloxone-induced pressor response in these animals by about 80% (ie., from 25±6 mmHg to 6±4 mmHg).

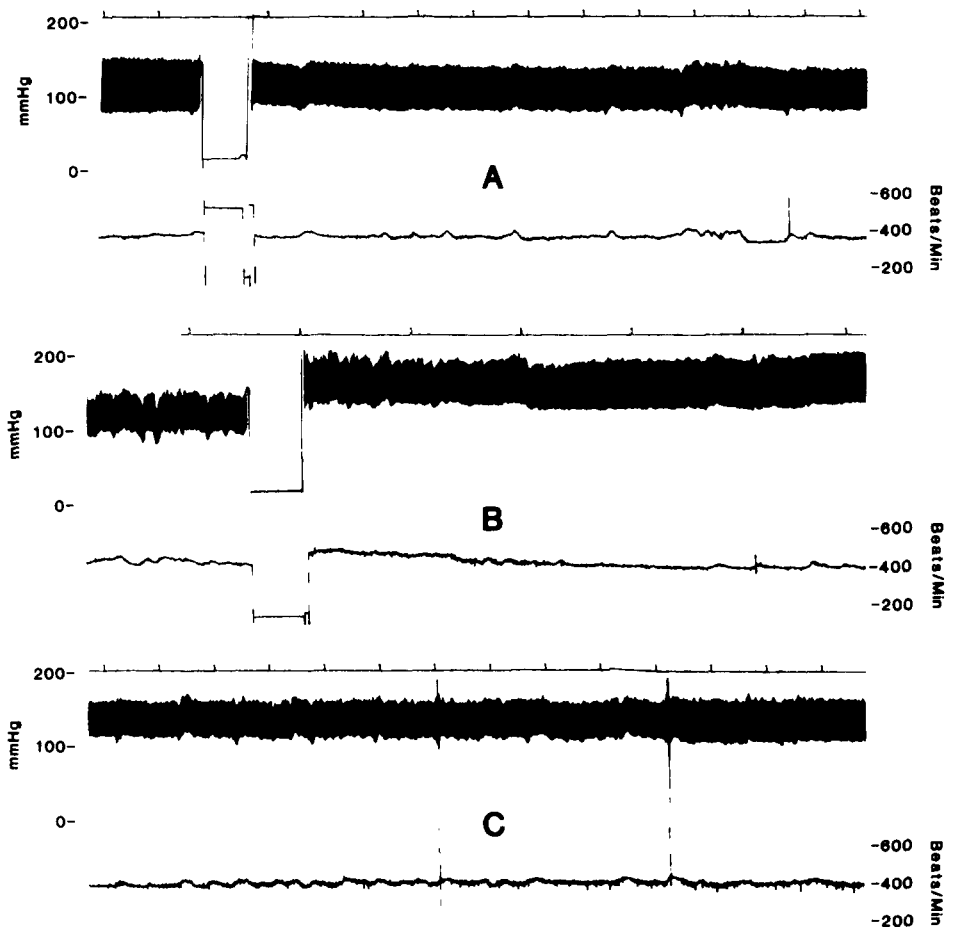

FIG. 1. Actual recordings illustrating the effect of naloxone (5 mg/kg) in control (tracings A) and morphine dependent (tracings B and C) conscious rats on arterial pressure (mmHg) and heart rate (beats/min). Interruptions in the tracings in A and B represent the points at which naloxone was injected through the arterial catheter. Tracings B and C are, respectively, 0–5 and 11–15 min following naloxone. The control and morphine dependent animals were constantly infused with, respectively, saline or morphine (35–100 mg/kg/day) over a 7-day period prior to naloxone. The time base was at 1 min intervals.

In the last series of experiments, clonidine, a non-opiate antihypertensive drug having anti-narcotic withdrawal properties, was examined for its ability to interfere with the expression of withdrawal symptoms following injection of naloxone (5 mg/kg) in morphine dependent rats. Intraarterial injection of clonidine (6–60 μg/kg) in morphine dependent rats produced a dose-dependent increase in MAP of 16–38 mmHg and a fall in HR of 43–90 beats/min. MAP and HR were allowed to return to preinjection levels (20–40 min) before the injection of naloxone. The data from these experiments is summarized in Fig. 2. Clonidine pretreatment resulted in a dose-related inhibition of the naloxone-induced pressor response. The 60 μg/kg dose of clonidine reduced the maximum pressure increase by 60%. Clonidine abolished the secondary decrease in HR produced by naloxone but had no effect on the initial increase in this parameter (Fig. 2). Likewise, the initial excitatory behavior phase also did not appear to be influenced by clonidine pretreatment. The mean value for WBS/hr in this group of control animals, 16±3 (n=7) was 8±5 (n=5), in clonidine pretreated rats; this difference was not statistically significant.

DISCUSSION

Himmelsbach's [15] studies of the cold pressor test, measurement of the reflex hypertensive response to sudden cold, in subjects addicted to narcotics led him to conclude that addiction is associated with hyperresponsiveness of the sympathetic nervous system. His studies [14,19] and those of others in man [25,34] indicated that withdrawal from narcotics in physically dependent subjects is associated with an increase in arterial blood pressure. Measurement of arterial pressure provides a good index of ongoing sympathetic nerve activity, however, cardiovascular changes are the least studied of the individual symptoms in the spectrum of opiate abstinence signs. Earlier studies in acutely narcotized

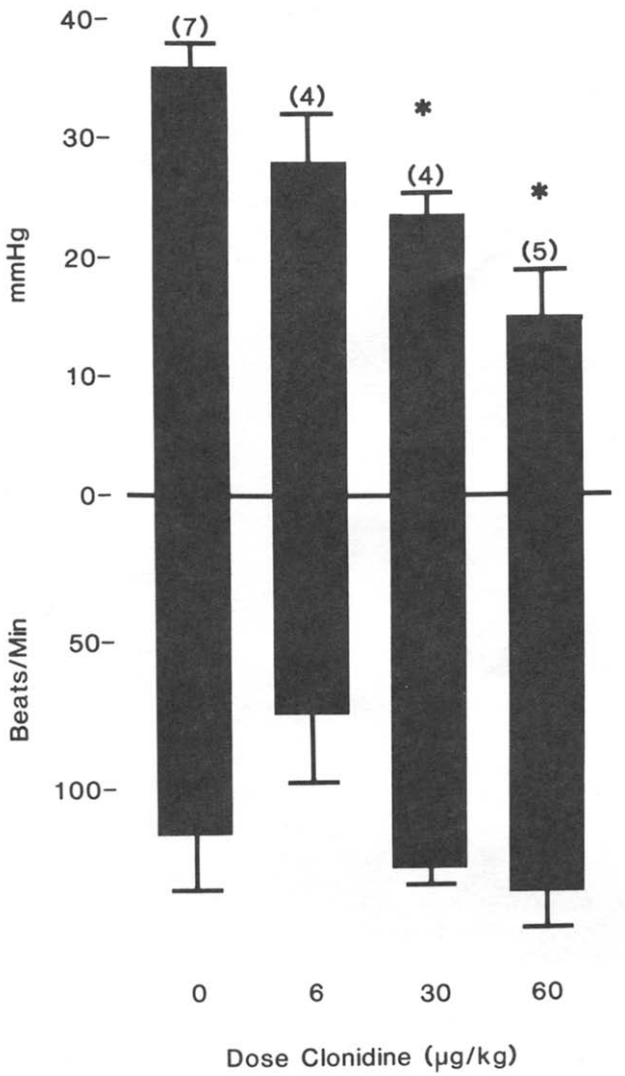

FIG. 2. Effect of clonidine pretreatment on the maximum increases in mean arterial pressure (mmHg) and heart rate (beats/min) following naloxone (5 mg/kg) in conscious, morphine dependent rats. Physical dependence was produced by constant intraarterial infusion of morphine (35–100 mg/kg/day) over a 7-day period prior to naloxone. Clonidine was administered at the doses indicated (0=saline injection) by intraarterial injection 20–40 min prior to naloxone. Values are means ± SEM with the number of experiments in parentheses. The asterisks indicate values which are significantly lower than the control (saline) value.

decerebrate cats [23] and anesthetized dogs [26] have demonstrated a reversal by narcotic antagonists of the cardiovascular depressant effects of morphine. Also, Labrecque and Domino [20] reported that naloxone induced a marked pressor response in morphine-tolerant, midpontine, pretrigeminal brainstem transected cats. The present study, however, was performed in intact, conscious and freely behaving animals, thus, allowing simultaneous measures of behavioral and central sympathetic components of withdrawal. The estimation of different components of withdrawal is essential since different underlying mechanisms are involved in the expression of the many withdrawal symptoms, and drugs tested for potential antiwithdrawal properties may act selectively on one or more of these mechanisms.

Chronic infusion of morphine in conscious rats over the 7 days was associated with only a minor reduction in arterial pressure. This was surprising in view of the ability of narcotic agonists to decrease blood pressure and reduce sympathetic nerve activity following acute injection [21, 22, 30, 31]. Since the animals were unanesthetized, intact baroreceptor reflexes or other circulatory homeostatic mechanisms may have counteracted the circulatory depressant effects of a slow infusion of morphine. In contrast, during abstinence, blood pressure was elevated up to 24 hr after withdrawal. Although heart rate also was increased the mean values over the 7 days were not significantly greater than the pre-withdrawal level. Individual means during abstinence, however, were sometimes 44 beats/min greater than pre-withdrawal levels. The increase in arterial pressure during abrupt withdrawal is a more sensitive index than is the increase in heart rate. While the magnitude of the heart rate increase also was employed as an index of withdrawal in man [14, 19, 25, 34] in the rat, the lack of sensitivity of this parameter may be related to the very high resting level in this species. Also, heart rate changes closely paralleled changes in motor activity resulting from locomotor activity as well as other behaviors. The withdrawal body shakes measured simultaneously with the cardiovascular parameters occurred at a frequency similar to those values reported by other workers employing chronic injections [16,24] or yoked intravenous infusion [17] of morphine and provide additional evidence for the physical dependency of the animals.

Intraarterial injection of naloxone in morphine dependent rats produced a more severe abstinence syndrome as compared with abrupt withdrawal. Qulaitative differences also existed between the two syndromes. Naloxone precipitated withdrawal was associated with an initial excitatory and secondary inhibitory phase of behavioral and motor activity. WBS were less evident during precipitated withdrawal, however, escape behavior was much more pronounced. Wei [33] first demonstrated the inverse correlation of WBS and escape behavior, a relationship which became more apparent as the intensity of withdrawal, and the dose of naloxone, was increased. The use of WBS as the sole index of withdrawal intensity, therefore, may prove unreliable if the severity of the abstinence syndrome is greatly altered during the period of measurement e.g., with a drug such as clonidine.

Heart rate changes also paralleled the changes in behavior and although motor activity was not quantitated, it was clear that the direction and magnitude of heart rate effects following naloxone were directly related to the effects on behavior. For example, animals who appeared to be more immobile exhibited a more profound bradycardic response. In contrast, the increases in arterial pressure were independent of the behavioral changes. This correlation between heart rate (but not blood pressure) on behavior was noted in our earlier studies [2] when it was observed that the magnitude of chronotrophic responses following intrahypothalamic injection of carbachol were significantly correlated with concomitant increases in motor activity. As in the present study, blood pressure changes were independent of behavioral changes. Although behavioral or emotional outbursts can produce changes in various cardiovascular parameters, heart rate appears to be especially sensitive.

The magnitude of the increase in MAP produced by naloxone was dose-related. This suggests that the changes in

MAP are directly related to the intensity of the withdrawal syndrome and therefore, may provide an objective and quantitative estimate of the intensity of the abstinence syndrome. The hypertensive response associated with withdrawal which was the most consistent and reproducible of the abstinence symptoms appears to reflect the ongoing state of the sympathetic nervous system since it is blocked by phentolamine. Also, the observation that the pressor response to the injection of naloxone in the dependent rat could be repeated 48 hr later would allow the design of experiments where each animal could be employed as his own control. This would reduce the number of animals required for a study and reduce even further the normal variability of measurements between animals.

The final portion of this study was designed to determine whether the cardiovascular changes which occur during withdrawal could be influenced by an agent having the property of inhibiting withdrawal symptoms. Clonidine pretreatment in clinically relevant doses [3,11] to morphine dependent rats produced a dose related inhibition of the pressor response to subsequent injection of 5 mg/kg of naloxone. This was the highest dose of naloxone used and the most intense withdrawal syndrome observed in the present study. Conceivably lower doses of clonidine might block withdrawal signs associated with less severe abstinence syndromes. The mechanism of clonidine's ability to inhibit the naloxone-induced pressor response is not known, however, the ability of clonidine to lower arterial pressure in essential hypertension is related to its ability to inhibit sympathetic nerve activity by a central action [12, 18, 27, 28]. Since many of the signs and symptoms of narcotic withdrawal are due to heightened autonomic responsiveness, including hypertension and tachycardia, clonidine's antiwithdrawal action may be related to its central sympatholytic effects.

The present findings indicate that measurement of cardiovascular parameters in freely-moving rats may provide an objective measure of the intensity of the morphine abstinence syndrome. Behavioral models of narcotic withdrawal in the rat are useful for the determinations of abuse potential of new narcotic agonists and in the determination of the antiwithdrawal potential of certain drugs like clonidine. The simultaneous measurement of arterial blood pressure makes this a more powerful animal model for these purposes and provides direct information concerning the state of the sympathetic nervous system which mediates many of the abstinence signs in man.

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REFERENCES

1. Bhargava, H. N. Rapid induction and quantitation of morphine dependence in the rat by pellet implantation. *Neuropharmacology* **52:** 55–62, 1977.
2. Buccafusco, J. J. and H. E. Brezenoff. Pharmacological study of a cholinergic mechanism within the rat posterior hypothalamic nucleus which mediates a hypertensive response. *Brain Res* **165:** 295–310, 1979.
3. Channabasavanna, S. M. and M. Subrahmanya. Clonidine for opiate withdrawal. *Lancet* **1:** 313–314, 1981.
4. Desmonts, J. M., G. Bohm and E. Couderc. Hemodynamic responses to low doses of naloxone after narcotic-nitrous oxide anesthesia. *Anesthesiology* **49:** 12–16, 1978.
5. Estilo, A. E. and J. E. Cottrell. Naloxone, hypertension, ruptured cerebral aneurysm. *Anesthesiology* **54:** 352, 1981.
6. Evans, L. E. J., P. Roscoe, C. P. Swainson and L. F. Prescott. Treatment of drug overdosage with naloxone, a specific narcotic antagonist. *Lancet* **1:** 452–455, 1973.
antagonist. *Lancet* **1:** 452–455, 1973.
7. Fielding, S., J. Wilker, M. Hynes, M. Szewczak, W. J. Novick and H. Lal. A comparison of clonidine with morphine for antinociceptive and antiwithdrawal actions. *J Pharmacol Exp Ther* **207:** 899–905, 1978.
8. Gianutsos, G., R. Drawbaugh, M. Hynes and H. Lal. The narcotic withdrawal symdrome in the rat. In: *Methods in Narcotics Research, Vol. 5, Modern Pharmacology–Toxicology*, edited by S. Ehrenpreis and A. Neidle. New York: Marcel Dekker, 1975, pp. 293–309.
9. Gold, M. S., D. E. Redmond and H. D. Kleber. Clonidine in opiate withdrawal. *Lancet* **1:** 929–930, 1978.
10. Gold, M. S., D. E. Redmond and H. D. Kleber. Clonidine blocks acute opiate-withdrawal symptoms. *Lancet* **2:** 599–602, 1978.
11. Gold, M. S., A. L. C. Pottash, I. Extein and H. D. Kleber. Clonidine and opiate withdrawal. *Lancet* **2:** 1078–1079, 1980.
12. Haeusler, G. Clonidine-induced inhibition of sympathetic nerve activity. No indication for a central presynaptic or indirect sympathomimetic mode of action. *Naunyn-Schmiedebergs Arch Pharmac* **287:**97–111, 1974.
13. Himmelsbach, C. K. Clinical studies of drug action. II. "Rossium" treatment of drug addiction. *Public Health Rep. Suppl. 125*, 1–18, 1937.
14. Himmelsbach, C. K. Studies of certain addiction characteristics of (a) dihydromorphine ("Paramorphan"), (b) dihydrodesoxymorphine-D ("desomorphine"), (c) dihydrodesoxycodeine-D ("desocodeine"), and (d) methyldihydromorphinone ("metopon"). *J Pharmacol Exp Ther* **67:** 239–249, 1939.
15. Himmelsbach, C. K. Studies on the relation of drug addiction to the autonomic nervous system: Results of cold pressor tests. *J Pharmacol Exp Ther* **73:** 91–98, 1941.
16. Hynes, M. D., G. Gianutsos and H. Lal. Effects of cholinergic agonists and antagonists on morphine-withdrawal syndrome. *Psychopharmacology* **49:** 191–195, 1976.
17. Hynes, M. D., M. D. McCartin, G. Shearman and H. Lal. Differential reduction of morphine-withdrawal body shakes by butaclamol enamtiomers. *Life Sci* **22:** 133–136, 1978.
18. Klupp, H., F. Knappen, Y. Otsuka, I. Streller and H. Tiechmann. Effects of clonidine on central sympathetic tone. *Eur J Pharmacol* **10:** 225–229, 1970.
19. Kolb, L. and C. K. Himmelsbach. Clinical studies on drug addiction. III. A crifical review of the withdrawal treatments with method of evaluating abstinence syndromes. *Am J Psychol* **94:** 759–799, 1938.
20. Labrecque, G. and E. F. Domino. Tolerance and physical dependence on morphine: Relation to neocortical acetylcholine release in the cat. *J Pharmacol Exp Ther* **191:** 189–200, 1974.
21. Laubie, M., H. Schmitt, J. Canellas, J. Roquebert and P. Demichel. Centrally-mediated bradycardia and hypotension induced by narcotic analgesics: Dextromoramide and fentanyl. *Eur J Pharmacol* **28:** 66–75, 1974.
22. Mansour, E., R. Capone, D. T. Mason, E. A. Amsterdam and R. Zelis. The mechanism of morphine-induced peripheral arteriolar dilation—Central nervous sympatholysis. *Amer J Cardiol* **26:** 648, 1970.
23. Martin, W. R. and A. J. Eisenman. Interactions between nalorphine and morphine in the decerebrate cat. *J Pharmacol* **138:** 113–119, 1962.

24. Martin, W. R., A. Wikler, C. G. Eades and F. T. Pescor. Tolerance to and physical dependence on morphine in rats. *Psychopharmacologia* **4:** 247–260, 1963.
25. Martin, W. R. and C. W. Gorodetzky. Demonstration of tolerance to and physical dependence on n-alkylnormorphine (nalorphine). *J Pharmacol Exp Ther* **150:** 437–442, 1965.
26. Paddleford, R. R. and C. E. Short. An evalutaion of naloxone as a narcotic antagonist in the dog. *J Am Vet Med Assoc* **163:** 144–146, 1973.
27. Sattler, R. W. and P. A. Van Zwieten. Acute hypotensive action of 2-(2,6-dichlorophenylamine)-2-imidazoline hydrochloride (St 155) after infusion into the cat's vertebral artery. *Eur J Pharmacol* **2:** 9–13, 1967.
28. Schmitt, H., H. Schmitt, J. R. Boissier and J. F. Guidicelli. Centrally-mediated decrease in sympathetic tone by 2-(2,6-dichlorophenylamino)-2-imidazoline (S.T. 155, Catapressan). *Eur J Pharmacol* **2:** 147–148, 1967.
29. Sparber, S. B. and D. R. Meyer. Clonidine antagonizes naloxone-induced suppression of conditioned behavior and body weight loss in morphine-dependent rats. *Pharmacol Biochem Behav* **9:** 319–325, 1978.
30. Stickney, J. L. and D. C. Eikenburg. Peripheral sympatholytic effects of 1-alpha-acetylmethadol. *J Cardiovasc Pharmacol* **3:** 369–380, 1981.
31. Varagic, V. and M. Kristic. Effects of analgesics on the hypertensive response to eserine. *Neuropharmacology* **5:** 237–240, 1966.
32. Washton, A. M., R. B. Resnick and R. A. Rawson. Clonidine for outpatient opiate detoxification. *Lancet* **1:** 1078–1079, 1980.
33. Wei, E. Assessment of precipitated abstinence in morphine-dependent rats. *Psychopharmacology (Berlin)* **28:** 35–44, 1973.
34. Wikler, A., H. F. Fraser and H. Isbell. N-alkylnormorphine: Effects of single doses and precipitation of acute "abstinence syndromes" during addiction to morphine, methadone or heroin in man (post addicts). *J Pharmacol Exp Ther* **109:** 8–20, 1953.