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# Discriminative Stimulus Properties of *levo-alpha*-Acetylmethadol and Its Metabolites

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HOLTZMAN, S G *Discriminative stimulus properties of* levo-alpha-*acetylmethadol and its metabolites* PHARMAC BIOCHEM BEHAV 10(4) 565–568, 1979 —The discriminative stimulus properties of *l*-α-acetylmethadol (LAAM), its metabolites *l*-α-acetyl-N-normethadol (NorLAAM) and *l*-α-acetyl-N,N-dinormethadol (DiNorLAAM), and methadone were evaluated in rats trained to discriminate between saline and morphine in a two-choice discrete-trial avoidance paradigm All four compounds produced dose-related increases in the number of trials completed on the morphine-appropriate choice lever after either SC or oral administration indicating that the discriminative stimulus properties of the four compounds and morphine are qualitatively similar LAAM and DiNorLAAM had a slow onset and long duration of action, and an oral parenteral potency ratio of 1 3 NorLAAM had a more rapid onset and shorter duration of action and was more potent following SC administration than either LAAM or DiNorLAAM, its oral· parenteral potency ratio was 1 10 These results are consistent with evidence from other studies that the pharmacologic activity of LAAM is dependent upon the conversion of LAAM to an active metabolite, probably NorLAAM The similarities between DiNorLAAM and LAAM suggest that the discriminative stimulus effects of the former compound are also attributable to a metabolite

levo-alpha-Acetylmethadol Methadone Discriminative stimulus

LEVO-ALPHA-acetylmethadol (LAAM) is a synthetic congener of methadone whose efficacy following oral administration and long duration of action have stimulated considerable interest in its potential as an alternative to methadone in drug maintenance programs for the treatment of opioid dependence [8, 9, 10, 15, 22] LAAM's long duration of action in man—on the order of 24 to 72 hr depending upon dose and route of administration [4,8]—is probably related to the sequential metabolic conversion of LAAM to the N-demethylated metabolites *l-alpha*-acetyl-N-normethadol (NorLAAM) and *l-alpha*-acetyl-N,N-dinormethadol (DiNorLAAM) [2] Both metabolites display typical opioid activity in vitro [14] and in vivo [19], and have a potency that is equivalent to or greater than that of the parent compound

Following the administration of single doses to volunteers experienced in the illicit use of narcotics, LAAM, like methadone, produced a characteristic opioid syndrome of subjective symptomology (i e , euphoria, [4]) However, comparable clinical studies of the metabolites of LAAM have not been reported The present study was undertaken to evaluate the stimulus properties of LAAM, NorLAAM, and DiNorLAAM in the rat. Recent investigations have demonstrated a good correlation between the discriminative stimulus properties of opioid analgesics in the rat and the subjective effects of these drugs in man [3, 12, 16, 18]. Dose-and time-effect relationships were determined for each compound administered subcutaneously and orally in rats trained to discriminate between saline and 3.0 mg/kg of morphine The stimulus effects of methadone were also determined in order to have a standard for comparison.

## METHOD

### *Animals*

The animals were 22 male Sprague-Dawley derived CFE and CD rats (Charles River Breeding Laboratories, Wilmington, Mass.) that weighed 250–350 g at the beginning of discrimination training Between experimental sessions the rats were housed in a colony room, two per cage, where they had continuous access to food and water. The colony room was illuminated between 6:00 a.m. and 6 00 p.m

### *Apparatus*

A standard one-lever rat chamber (model 1110-L, Grason-Stadler Co , Bolton, Mass.) was modified by adding two "choice" response levels to the wall opposite the original level which was designated the observing lever The choice levers were separated by a narrow Plexiglas partition that extended from the ceiling of the chamber to 1 0 cm above the grid floor. The floor was wired to a shock generator capable of delivering a scrambled electric shock at a constant current. The test chamber was housed within a ventilated enclosure that was light-proof and sound-attenuating Schedule contingencies were controlled by automatic relay programming modules

### *Discrimination Training*

The procedure for training rats to discriminate between saline and 3 0 mg/kg of morphine has been described in detail previously [16,18] Briefly, rats were trained to com-

plete a two-response chain in order to terminate a trial and avoid or escape from electric shocks delivered to the grid floor to the chamber. The beginning of a trial was signalled by the illumination of the house light and the presentation of white noise The animal was required first to press the observing response lever, then to press one of the two choice response levers on the other side of the chamber. The first observing response of the trial terminated the white noise, when the appropriate choice lever was pressed, the house light was extinguished and the trial ended. A trial was defined as correct if the rat emitted the response sequence of observing lever—appropriate choice lever, and as incorrect if the sequence of responses was observing lever—inappropriate choice lever—appropriate choice lever Beginning 5 sec after the start of a trial, a 1 0 mA electric shock was delivered intermittently (1 sec on, 2 sec off) until the two-response chain was completed The interval between trials was 50 sec during which the test chamber was illuminated dimly with red light. A session ended after 21 trials or 30 min, whichever came first The first trial of each session served as a warm-up and was not included in the data analysis

Five daily training sessions were conducted each week Saline or morphine (3.0 mg/kg) were injected SC 30 min before each session according to a double alternation sequence (i e , saline, saline, morphine, morphine, saline ..) Half of the rats were trained to press the right choice lever throughout each saline session and the left choice lever throughout each morphine session, the other half were trained under the opposite conditions Training continued until the rats completed four consecutive sessions in which at least 18 out of 20 trials (i e , 90% exclusive of the first trial) were completed on the appropriate choice lever The next two sessions (one saline and one morphine session) were conducted as test sessions in which a trial could be terminated by a response on either choice lever An animal's behavior was considered to be under stimulus control if at least 18 of the 20 trials of each test session were completed on the appropriate choice lever

*Drug Testing*

After stimulus control of behavior had been established, drug test sessions were conducted on Tuesdays and Fridays provided that discrimination performance remained at the 90% level in training sessions held on Mondays, Wednesdays and Thursdays in which saline and morphine continued to be administered on a double alternation basis During test sessions both choice levers were electrically activated so that a trial could be terminated by a response on either choice lever following a response on the observing lever. Test sessions and training sessions were the same in all other respects In the determination of each dose-response curve, doses were administered in a random sequence that also included saline Points in the time-effect curves were determined in a nonsystematic order Rats were randomly assigned to drug series without regard to their history of drug testing This procedure of training and testing results in no detectable tolerance development to the stimulus effects of the drugs, and yields reproducible dose-response curves [16,18]

*Drugs*

The drugs used were morphine sulfate, dl-methadone hydrochloride, and the hydrochloride salts of l-α-acetyl-methadol, l-α-acetyl-N-normethadol, and l-α-acetyl-N,N-dinormethadol, which were provided by the National Institute on Drug Abuse All doses are expressed in terms of the free base. Drugs were dissolved in 0.9% saline and administered either subcutaneously or orally by gavage in a volume of 1 0 ml/kg of body weight

## RESULTS

Figure 1 shows dose-response curves for LAAM, NorLAAM, and DiNorLAMM determined at 0.5, 2, 4 and 6 hr after administration, and for methadone determined at 0 5 and 2 hr after administration All four drugs produced a dose-related increase in the number of trials completed on the morphine-appropriate choice lever, and, at some dose, produced stimulus control of behavior comparable to that produced by the training dose of morphine (i.e , at least 18 out of 20 trials were completed on the morphine lever points above upper dashed line) However, the pattern of development of morphine-like stimulus control of behavior varied considerably among the four compounds LAAM and DiNorLAAM had a very gradual onset of action. Both were relatively inactive at the 0 5 hr interval as evidenced by the fact that as much as 30 mg/kg of the latter resulted in the completion of an average of only 8 75 trials on the morphine lever However, by 4–6 hr after administration both drugs were approximately equipotent to the training dose of morphine in producing stimulus control of behavior The slow increase in relative potency presented a unique problem in that animals tested with the higher doses of LAAM or DiNorLAAM at the earlier time points appeared behaviorally normal at the conclusion of the experimental session but would then develop severe respiratory depression several hours after being returned to the home cage. Accordingly, 10 mg/kg of naltrexone was routinely adminstered at the end of a session to any animal that had been injected with 10 mg/kg or more of either LAAM or DiNorLAAM

The onset of action of NorLAAM was faster and its peak potency about three times greater than that of LAAM or DiNorLAAM (Fig. 1) The activity of NorLAAM remained relatively constant between 0.5 and 4 hr, and began to decline at 6 hr Methadone also displayed a rapid onset of action, but its duration of action was short in contrast to that of LAAM and its metabolites With a pretreatment interval of 0 5 hr, 3 0 mg/kg of methadone produced stimulus control of behavior comparable to that of the morphine training dose, whereas 10 mg/kg was required when the pretreatment interval was lengthened to 2 hr (Fig 1) Some of the rats became cataleptic after receiving 10 mg/kg of methadone and only two out of five responded during the session The necessity of administering even higher doses of methadone precluded the determination of dose-response curves at pretreatment intervals longer than 2 hr

Time-effect relationships were characterized further by determining the time course of morphine-appropriate responding following the administration of 1 0 mg/kg of NorLAAM or 3 0 mg/kg of LAAM, DiNorLAAM, or methadone, the lowest dose of each drug that produced stimulus control of behavior comparable to that produced by the morphine training dose (see Fig 1) The differences in rate of onset and duration of action among the four drugs are readily apparent in Fig 2 Stimulus control of behavior comparable to that produced by morphine was maintained for 10 hr from the time of injection by LAAM and DiNorLAAM,

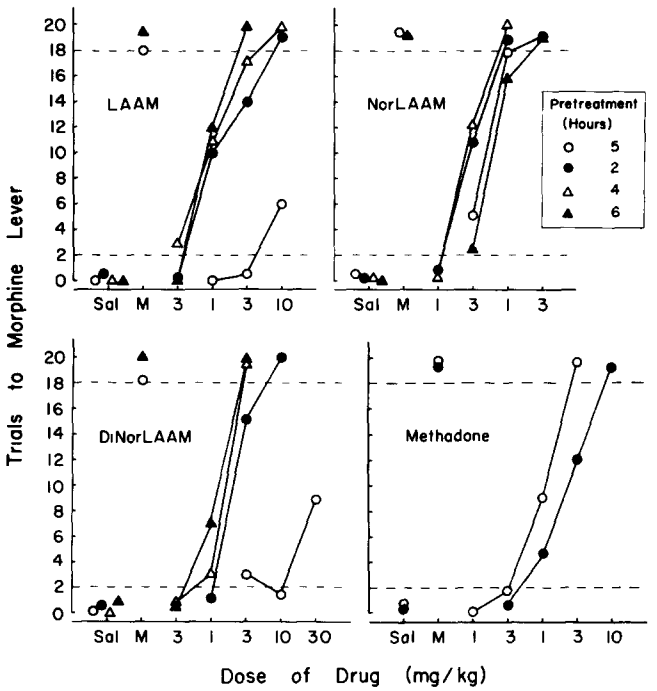


Fig 1 Dose-related discriminative effects of LAAM, NorLAAM, DiNorLAAM, and methadone administered s c as a function of the pretreatment interval (0 5, 2, 4, or 6 hr) in rats trained to discriminate between saline and 3 0 mg/kg of morphine Each point is the mean number of trials completed on the morphine-appropriate choice lever in a 20 trial session, the remaining trials were completed on the saline-appropriate lever Means are based upon one observation in each of 4–5 rats with the following exceptions only 2 out of 4 rats responded when tested 6 hr after 3 0 mg/kg of NorLAAM, and only 2 out of 5 rats responded when tested 2 hr after 10 mg/kg of methadone The mean number of trials completed on the morphine-appropriate lever 0 5 hr after the administration of 3 0 mg/kg of morphine and at the four time intervals after the administration of saline are represented by the points above M and Sal, respectively The upper and lower horizontal dashed lines indicate the minimum levels of discriminative responding to which the animals were trained with morphine and saline, respectively

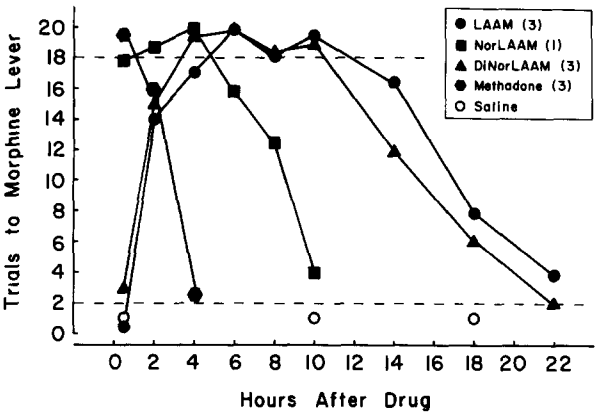


Fig 2 Time course of the discriminative effects of LAAM (3 0 mg/kg), NorLAAM (1 0 mg/kg), DiNorLAAM (3 0 mg/kg), and methadone (3 0 mg/kg) administered SC in rats trained to discriminate between saline and 3 0 mg/kg of morphine Each point is the mean number of trials completed on the morphine-appropriate choice lever in a 20-trial session, the remaining trials were completed on the saline-appropriate lever Means are based upon one observation in each of 4–5 rats Each time point was determined on a separate day

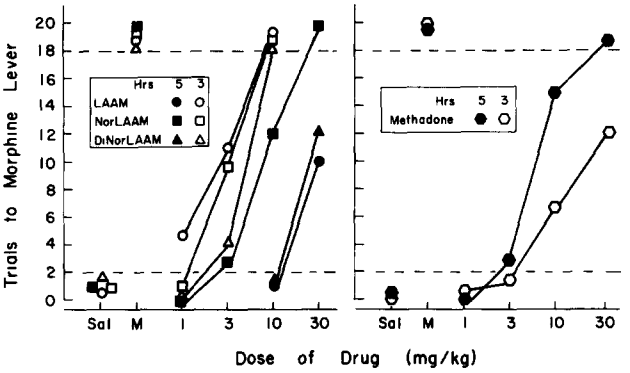


Fig 3 Dose-related discriminative effects of LAAM, NorLAAM, DiNorLAAM, and methadone at 0 5 and 3 hr after oral administration in rats trained to discriminate between saline and 3 0 mg/kg of morphine Each point is the mean number of trials completed on the morphine-appropriate choice lever in a 20-trial session, the remaining trials were completed on the saline-appropriate choice lever Means are based upon one observation in each of 5 rats The mean number of trials completed on the morphine-appropriate lever 0 5 hr after the SC administration of 3 0 mg/kg of morphine and at 0 5 and 3 hr after the oral administration of saline are represented by the points above M and Sal, respectively

for 4 hr by NorLAAM, and for only 0 5 hr by methadone. At least 22 hr were required for predominantly saline-appropriate responding to return after the administration of LAAM and DiNorLAAM, 10 hr after NorLAAM, and 4 hr after methadone

The relative time- and dose-effect relationships among the four drugs for producing morphine-like stimulus control of behavior after oral administration were similar to those observed after SC administration (Fig. 3). The potency of LAAM and DiNorLAAM increased an estimated 10 fold between 0.5 and 3 hr, the potency of NorLAAM increased about 3 fold during this interval, and the potency of methadone decreased an estimated 3 fold At the 3 hr interval LAAM, NorLAAM and DiNorLAAM were approximately equipotent, producing morphine-appropriate responding comparable to that produced by the morphine training dose at 10 mg/kg, and were an estimated 10 times more potent than methadone which, at the highest dose that could be safely tested (30 mg/kg), resulted in an average of only 12 trials being completed on the morphine-appropriate lever

## DISCUSSION

The results of this study indicate that the discriminative stimulus properties of LAAM and its two principal metabolites NorLAAM and DiNorLAAM are qualitatively similar to each other and to those of methadone It is well documented

that drugs identified by experienced users as dope on the basis of distinctive drug-induced alterations in thought, mood and perception [5] reliably elicit drug-appropriate responding in rats trained to discriminate between saline and morphine [6, 16, 18, 21] Both LAAM [4] and methadone [7] have been shown to produce a morphine-like or opioid syndrome of subjective effects in human volunteers. Based upon the findings of the present study, NorLAAM and DiNorLAAM would also be expected to produce typical morphine-like subjective symptomology in man

All of the drugs—LAAM, NorLAAM, DiNorLAAM, and methadone—were capable of producing morphine-like stimulus control of behavior when administered by either the SC or oral routes However, in going from SC to oral administration, LAAM and DiNorLAAM retained a greater degree of their activity than did NorLAAM and methadone The oral parenteral potency ratios, estimated at the time of peak stimulus effects, were approximately 1:3 for LAAM and DiNorLAAM and 1:10 for NorLAAM and methadone. The ratio for methadone is in accord with that obtained in a previous study [17].

The slow onset and long duration of action of LAAM had prompted Sung and Way [20] to suggest that the pharmacologic activity of LAAM is largely dependent upon the conversion of the parent compound to an active metabolite This view was supported by the subsequent isolation of the two N-demethylated metabolites of LAAM [1,13] and the demonstration of their opioid activity in vitro [14] and in vivo [19] In the present study, the gradual onset and prolonged action of LAAM is consistent with the concept that the discriminative stimulus effects of LAAM are similarly attributable to a metabolite NorLAAM is the most likely candidate for this role because of its rate of onset and duration of action, and because it is more potent than LAAM when administered s c However, the possible contribution of other metabolites, including methadone [11], to the activity of LAAM cannot be ruled out On the other hand, the profile of activity of DiNorLAAM is strikingly similar to that of LAAM with respect to the gradualness of onset and long duration of action, and the relatively high oral· parenteral potency ratio Hence, it is conceivable that the discriminative stimulus effects of DiNorLAAM, like those of LAAM, are attributable to a metabolite of the injected compound

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