Short Title

Kenneth G. J. Lord
Psychology

EFFECTS OF CHRONIC MORPHINE ON AVOIDANCE BEHAVIOUR

M.Sc. PSYCHOLOGY

Kenneth G. J. Lord

EFFECTS OF REPEATED ADMINISTRATION OF MORPHINE ON SHUTTLE-BOX AVOIDANCE BEHAVIOUR

The effects of morphine on shuttlebox avoidance behaviour, and spontaneous activity in the jiggle cage and running wheel, were observed over 20 drug sessions. The first morphine injection produced facilitation of shuttle avoidance, no clear effect on jiggle-cage activity, and slight suppression of wheel running. These and other results suggest that morphine's acute effect depends upon the topography of behaviour under investigation. With repeated administration, morphine's facilitation of shuttle avoidance and jiggle-cage activity increased over the first 8 to 10 drug sessions, suggesting either the potentiation or unmasking of the stimulant effect. Over the last 10 drug sessions, facilitation of jiggle-cage activity remained stable while facilitation of shuttle avoidance declined. This suggests the development of tolerance to morphine's facilitation of instrumental responses, but not of spontaneous activity. These findings are broadly consistent with the view that the basis of tolerance development is drug-produced impairment which is proportional to the functional demand made upon the organism by the testing situation.

M.Sc. PSYCHOLOGIE

LES EFFETS DE L'ADMINISTRATION REPETEE DE MORPHINE SUR LE COMPORTEMENT D'EVITEMENT ALTERNE

Les effets de la morphine sur l'évitement alterné continue de même que sur l'activité spontanée en cage inclinée et à l'intérieur de la roue de course furent observés au cours de 20 sessions. La première injection de morphine a resulté en une augmentation du taux d'évitement continu, pas d'effet marqué sur l'activité en cage inclinée et une légère diminution de course a l'intérieur de la roue. Ces resultats et d'autres suggèrent que l'effet marqué de la morphine dépend des caractéristiques du comportement étudié. Avec la repetition des injections, l'augmentation due à la morphine du taux d'évitement et de l'activité dans la cage inclinée devinrent plus marqués au cours des premières 8 à 10 sessions, ce qui semble être soit une sensibilisation à l'effet stimulant soit une relâche de l'effet dépresseur concurrent. Au cours des 10 dernières sessions, l'augmentation de l'activité en cage inclinée est demeurée élevée tandis que l'augmentation du taux d'évitement s'est amoindrie. Ceci suggère le développement d'une tolérance à l'effet d'augmentation de la morphine sur le taux de réponse instrumentale, mais pas à l'effet sur l'activité spontanée. Ces observations sont en accord avec la conception que le développement de la tolérance est fondé sur un dommage causé par la drogue, lequel est proportionnel à la demande fonctionnelle de la situation expérimentale.

EFFECTS OF REPEATED ADMINISTRATIONS OF MORPHINE ON SHUTTLE-BOX AVOIDANCE BEHAVIOUR

by

Kenneth G. J. Lord

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Department of Psychology
McGill University
Montreal

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INTRODUCTION

Like many other drugs, morphine facilitates certain responses under some conditions and suppresses them under other conditions. These stimulant and depressant components of morphine's action have been demonstrated on a wide range of behaviour (Goldstein, Aronow, & Kalman, 1968). Over the course of repeated administrations of the drug, however, the morphine-induced changes in behaviour may become progressively attenuated, requiring increased doses to produce a given behavioural effect. This phenomenon of tolerance to morphine's effects on behaviour is well documented for the depressant component of its action in the rat (e.g., Kaymakcalan & Woods, 1956; Kumar, Mitchell, & Stolerman, 1971; Lorens & Mitchell, 1973; Martin, Wikler, Eades, & Pescor, 1963). But as yet no firm conclusion has been reached regarding the existence of tolerance to morphine's facilitatory effect on behaviour in this species. While some investigators have stated that tolerance to the stimulant component of morphine's action does not develop (Seevers & Deneau, 1963), recent positive findings indicate the need for further investigation. This thesis bears on the question of the development of tolerance to the stimulant component of morphine's behavioural action in the rat. A brief review of selected studies will aid in further defining the purpose of the present study.

Stimulant Effects of Acutely Administered Morphine

The rat is usually considered to be among those species whose predominant response to morphine is one of sedation (Maynert, 1967).

But it is likely that the lack of observed stimulant effects on behaviour reflects some peculiarities of dose or the response measured. For instance, morphine (sulfate, unless otherwise indicated) is reported to depress discriminated-avoidance behaviour whether the response is pole climbing (Cook & Weidley, 1957), running in a wheel (Verhave, Owen, & Robbins, 1959), or shuttling back and forth across a barrier (Verhave, Owen, & Slater, 1958). Small doses are observed to have no effect while large doses increase response latencies. However, since avoidance-response latencies are minimal by the time drug testing is begun, the absence of a stimulant effect of small doses may be due to a ceiling of response speed.

Heise and Boff (1962) have grouped morphine with hypnotics and phenothiazine tranquilizers on the basis of its action on free-operant avoidance behaviour. In their procedure, the pressing of an "avoidance" lever postponed the onset of footshock; this response-shock (R-S) interval was 40 sec. When the animal failed to avoid, the 5-sec. pulse of foot shock could be turned off by pressing a second or "escape" lever. However, the shock was repeated at intervals until an avoidance response was emitted; this shock-shock (S-S) interval was 20 sec. They observed that, following 0.75 mg/kg of morphine, subjects would occasionally stop lever pressing, receive several shocks, and then resume responding at a higher rate than on pre-drug baselines. After a few minutes the subjects would pause again and the cycle would be repeated. Hence, although they made approximately the same number of avoidance responses as during pre-drug sessions, the

subjects received significantly more shocks because of the irregular temporal distribution of responses. A morphine dose of 1.2 mg/kg led to a decrease in the total number of avoidance responses emitted during a session. A mean dose of 3.8 mg/kg resulted in response failure, defined as four failures per hour to press the escape lever when shocked. These findings suggest that morphine does not uniformly depress avoidance behaviour, but that its effect depends on the dose and the particular behavioural measure used.

Evidence that morphine facilitates free-operant avoidance behaviour is provided by Holtzman and Jewett (1971) who report that 1 mg/kg of morphine produced a 7.5 percent increase in avoidance rate rather than a decrease, as reported by Heise and Boff (1962). This discrepancy may be partly due to procedural differences since these investigators used chambers with only one lever and R-S and S-S intervals of 30 and 15 sec., respectively. The next largest dose tested, 2 mg/kg, produced an initial decrease of avoidance rate followed by an increase. Doses of 4 and 8 mg/kg produced a uniform rate decrease.

Facilitation following 1 mg/kg of morphine has also been reported for responses maintained by food on several fixed and variable ratio schedules (Thompson, Trombley, Luke, & Lott, 1970). Rates on interval schedules were largely decreased by this dose while 3 and 6 mg/kg decreased response rates on all schedules tested.

Glick and Rapaport (1974) report that doses of 1.25 and 2.5 mg/kg of morphine significantly increase lever-pressing rates

for lateral hypothalamic electrical self-stimulation in the rat. No significant change was produced by 5 mg/kg, while 10 mg/kg significantly decreased response rates during the 30-min. sessions. However, investigators using longer periods of testing (Adams, Lorens, & Mitchell, 1972; Lorens & Mitchell, 1973) report that, two to three hours after the injection of a large dose of morphine, there is a gradual reversal of the depressant effect to a marked increase of self-stimulation rate.

The acute effects of morphine on instrumental responses are paralleled by similar effects on certain measures of general or "spontaneous" activity. Davis and Brister (1973) have investigated the effect of morphine on activity in a circular alley in which the photocells were interconnected so as to detect only "large horizontal movements." During the first hour following injection, doses of 1.25, 2.5, and 5 mg/kg produced large increases in activity which lasted slightly longer as the dose increased and which gradually subsided by the third to fourth hour. A biphasic effect, similar to that described above, was observed following 20 mg/kg. This was the largest dose used by them. The initial decrease of activity was replaced by a marked stimulation during the third hour after injection.

Kumar, Mitchell, and Stolerman (1971) found increases in both eating and spontaneous activity over a 4 hr. period after the injection of 20 mg/kg of morphine hydrochloride. They measured activity in a square chamber containing two photocells which were positioned at right angles to each other.

Behavioural "excitation" in rats, following small doses of morphine, has also been reported by Fog (1970). He recorded the

incidence of rearing, locomotion, and three forms of grooming during the 2- to 4-hr. periods of observation. While a dose of 0.5 mg/kg had little effect, 1 mg/kg markedly enhanced grooming and slightly increased rearing and locomotion. Doses of 5 and 20 mg/kg decreased the incidence of all categories of behaviour.

Sloan, Brooks, Eisenman, and Martin (1962) have investigated the effect of large doses of morphine on the incidence of several categories of behaviour including lying, walking, circling, standing, scratching, preening, and exploring. Doses of 15 and 30 mg/kg resulted in a biphasic effect like that described above for continuous avoidance and self-stimulation behaviour. The largest dose tested was 60 mg/kg which produced profound sedation. This reached a maximum two hours after injection and had only partially subsided by the end of the 4-hr. observation period.

The findings discussed to this point appear to support the conclusion that the stimulant effect of acutely administered morphine occurs only at low doses (Glick & Rapaport, 1974). Following large doses, the stimulant component may be delayed until the level of available morphine in the blood falls to that of a small initial dose, giving rise to the biphasic effect. Yet it appears that dosage is not the only variable of importance. Holtzman and Jewett (1972) report that the intensity of footshock can determine whether a given dose of morphine has a stimulant or depressant effect on free-operant avoidance behaviour. They found that the same dose of morphine markedly increased the avoidance rate of rats trained with 1.3 mA ("high") footshock,

but decreased the response rate of subjects trained with 0.8 mA ("low") shock. For example, during the first hour following injection of 4 mg/kg, the response rate of the high-intensity group was over 160 percent of control values compared to 40 percent for the low-intensity group. The difference between the two groups increased with dose size through a range of 1 to 8 mg/kg. Thus, the failure of Heise and Boff (1962) to observe a facilitative effect of morphine on free-operant avoidance behaviour may also be due, in part, to their use of a shock intensity of 0.6 mA. To the extent that a higher shock intensity may induce a higher level of drive, the group differences could represent a positive-multiplicative interaction of drive with morphine's stimulant effect, similar to that demonstrated for methylphenidate by Mendelson and Bindra (1962).

Behavioural Effects of Repeated Morphine Administration

As stated at the outset, repeated administration of morphine often results in the progressive attenuation of its behavioural effects. Tolerance to the depressant effect may also be associated with the earlier appearance and increased magnitude of the stimulant component. For example, Lorens and Mitchell (1973) report that response rate for lateral hypothalamic electrical self stimulation was reduced by 75 percent during the first hour following an injection of 5 mg/kg of morphine. This decrease was no longer apparent by the third session and had been replaced by a significant rate increase by day 5. Babbini and Davis (1972) describe a similar pattern of change for the level of spontaneous activity in a circular alley. Daily injections of 20 mg/kg of

morphine, for 48 days, resulted in the progressively earlier appearance of peak-activity level. Moreover, the increase in activity became larger each day right up to the final day of testing.

Investigations of tolerance to morphine's stimulant effects on behaviour, using small doses of the drug, have only recently been undertaken. Glick and Rapaport (1974) measured the facilitation of responding for electrical self-stimulation of the brain following the repeated administration of 2.5 mg/kg of morphine. They observed a decline in the total number of responses emitted during the daily 30-min. sessions until, by day 4, performance after receiving the drug was not significantly different from previous, saline baselines. Performance following saline, on day 5, was also similar to the pre-drug baseline, suggesting that the attenuation of the effect of the drug was not due to non-specific factors such as physical debilitation of the subjects resulting from repeated intraperitoneal injections.

Holtzman (1974) reports tolerance to morphine's facilitative effect on free-operant avoidance by lever pressing. Rats were trained to avoid a 1 mA footshock using S-S and R-S intervals of 15 and 30 sec., respectively. Drug injections were begun when subjects attained the criterion of an average of 16 or fewer shocks received per hour over the 4-hr., twice-weekly sessions. Immediately prior to the first experimental session of the week, subjects received either 0.3, 1, 3, or 10 mg/kg of morphine. At the end of the session, supplemental injections were given to those rats which had received one of the three smaller doses in

order to bring the total amount of morphine administered to each animal to 10 mg/kg. Then, five hours later, each subject received another 10 mg/kg of morphine which brought the daily total up to 20 mg/kg. During each of the next two days, all subjects received two 10 mg/kg injections spaced nine hours apart. The rats were tested for the second time on day 4 after receiving the same dose that was administered to them prior to the first session of the week. Experimental weeks were alternated with weeks during which the rats received saline, until all subjects had been tested at all four doses. With this procedure, Holtzman observed a significant attenuation of the facilitative effect of 0.3, 1, and 3 mg/kg of morphine on the second test day. Tolerance to the depressant component was apparent in the form of a marked facilitation resulting from the 10 mg/kg dose. However, Holtzman's use of supplemental injections leaves uncertain whether the attenuation of the behavioural response was a cumulative drug effect, or represented tolerance to the particular dose.

Babbini and Davis (1972) observed no indication of tolerance to morphine's facilitative effect on spontaneous activity in a circular alley, produced by doses of 1.25, 2.5, and 5 mg/kg. Daily 4-hr. sessions were preceded by injections of saline for the first eight days and morphine for the following 30 days. Drug-induced activity increases continued essentially unchanged throughout the month of repeated administration. The return of activity levels to pre-drug values, following a final saline injection, again suggests that there were no changes in the

behavioural baseline due to physical debilitation or cumulative drug effects.

Taken together, the results of the above three studies appear to indicate that, while the repeated administration of morphine is a necessary condition for the development of tolerance to the stimulant component of action, it may not be a sufficient one. Experimental procedure appears to be a factor; attenuation of the stimulant effect was observed in subjects required to perform some task, but was not observed when the behavioural measure was level of spontaneous activity. Similar results have been obtained with d-amphetamine by Schuster and Zimmerman (1961). They observed tolerance to the drug's disruptive effect on differential reinforcement of low response rate (DRL) for food, but not to its facilitation of spontaneous activity measured in the same subjects on alternate days.

Experimental procedure is also a factor in the development of tolerance to morphine's analgesic component of action. Using a "hot-plate" apparatus, Adams, Yeh, Woods, and Mitchell (1969) measured the effect of 5 mg/kg of morphine on the paw-lick latencies of two previously untested groups of rats. They found no significant difference between the group which had received four previous drug injections, and those animals which had previously received only saline. However, they did find evidence of tolerance in other groups which had been placed in the apparatus following each of four previous injections, whether the testing surface had been heated on those occasions or not. Gebhart, Sherman, and Mitchell (1971) have shown that, even without

exposure to the testing apparatus, a measurable degree of tolerance to morphine'a analgesic component of action will develop if the injections are given daily, instead of weekly, as done by Adams et al. (1969). They consider this to be "pharmacologic tolerance," as opposed to the "behavioural tolerance" which develops as a result of experience with the testing apparatus while under the influence of the drug. This distinction is rejected by Kalant, Leblanc, and Gibbons (1971) who conclude that the only detectable difference between these two "types" of tolerance is one of rate of development. Hence, they suggest that the term "behaviourally augmented tolerance" replace "behavioural tolerance" since experience appears to accelerate tolerance development rather than give rise to a fundamentally different process.

The Present Investigation

It is evident that, at least under certain conditions, tolerance does develop to the facilitative effect of morphine on instrumental responses. Since there are so few studies which bear directly on this question, however, there is a need for replication and extension of these findings. Hence, the purpose of the present investigation was to attempt confirmation of the development of tolerance to the facilitative effect of morphine on free-operant avoidance behaviour. "Shuttling behaviour" was selected in order to obtain information regarding morphine's facilitative effect on a response with a topography different from that of lever pressing. The suitability of this response for this purpose was established in a dose-response curve study which I carried out

before undertaking the present investigation. The selection of a dose of 5 mg/kg for this investigation was also based on the results of the dose-response curve study.

Two measures of spontaneous activity were obtained using the jiggle cage and running wheel. The jiggle cage is a sensitive instrument for measuring morphine's stimulant component of action on spontaneous activity (Eidelberg & Schwartz, 1970; Bauxbaum, Yarbrough, & Carter, 1971), but has apparently not been used in studies of tolerance. Running in a wheel was chosen on the basis of the similarity of certain of its behavioural components to those of shuttling behaviour.

METHOD

Subjects

The subjects were 21 experimentally naive male Sprague-Dawley rats purchased from Bio-Breeding Laboratories, Ottawa. Each weighed from 250 to 300 gms at the start of the experiment. They were housed individually with food and water continuously available except during experimental sessions. The daily light phase in the animal quarters was from 07:00 to 21:00 hrs.

Apparatus

The shuttlebox was constructed of Plexiglas. It measured 60 cm long, 18 cm wide, and 18 cm high; the long sides and the top were transparent while the end panel on each side was translucent. A 6-watt bulb was mounted in a circular housing behind each of the two end panels. Illumination of the bulb in each

housing projected a bright disc of light, 6 cm in diameter, on the translucent panel. This light served as a discriminative stimulus during the initial phase of training. The floor of the shuttlebox consisted of 3 mm-diameter steel rods spaced 1.5 cm apart and was attached to the walls only at pivot points in the center of the chamber. Each half of the floor was independently pivoted and was held up at the end by two small coil springs. When the animal crossed to one side, its weight depressed that half of the floor slightly and activated a microswitch mounted beneath it. Thus, while an animal was in the shuttlebox, either one microswitch or the other was closed. Each half of the floor was also a separately-wired shock grid so that the 1 mA "scrambled" footshock, provided by a Grason-Stadler (model E1064GS) shock generator, was delivered only to the side on which the animal was standing. The experimental procedure was programmed for automatic operation with solid-state logic modules (BRS-Foringer Series 100 "Digi-Bits"). Responses and shocks were recorded automatically on Grason-Stadler (model E3700) digital counters and on a cumulative recorder (Scientific Prototype Mfg. Corp.).

The jiggle cage was 30 cm long, 18 cm wide, and 18 cm high. The top, bottom, and the two long sides were constructed of 1 cm square wire mesh while the two short sides were made of sheet metal. A frame, consisting of 2 1/2 cm strips of wood, was attached to the bottom of the cage to add rigidity and to permit attachment of the movement-sensing switches. One microswitch ("Robertshaw" Corp.) was attached, with the actuating lever pointing down, to the middle of each of the four sides of the

frame. The weight of the cage was supported by a swivel-joint ("Barry-Mount," Barry Manufacturing Corp.) attached to the middle of the floor. The cage was thus able to tilt freely in all directions about the central axis provided by this swivel joint. When empty, the cage was maintained in a level position by the actuating levers of the microswitches which served as "feet" for the chamber. The weight of an animal would cause the cage to tilt slightly and close one or, at most, two switches. Each switch closure was recorded on a Grason-Stadler (model E3700) digital counter.

The running wheel was of the conventional Wahmann type with a diameter of 35 cm and a width of 12.5 cm. A 5-by-5 cm cardboard panel was mounted on the circumference of the wheel to permit detection of the revolutions by interrupting the beam of two photocells placed near the circumference and directly across from each other. Both photocells had to be activated before a revolution was counted; thus, only large wheel movements were recorded. Each wheel revolution was recorded automatically on a Grason-Stadler (model E3700) digital counter.

The data from all three testing devices were cumulated on a Grason-Stadler (model E12505A) print-out counter, which printed running totals at 1-min. intervals. Each apparatus was located in a sound-attenuating chamber containing a one-way mirror to permit observation of the subject and also a blower and compressed-air outlet to provide ventilation and to mask extraneous noise.

Training Procedure

There were seven subjects in each of the three groups. Subjects in the free-operant avoidance group were introduced to the shuttlebox in groups of four, for 30 min., on the day prior to the start of training. During each daily 1-hr. training session, S-S and R-S intervals of 5 and 30 sec., respectively, were in effect. Discriminated-avoidance training was given during the first few sessions since preliminary results suggested that this facilitated the acquisition of free-operant avoidance responses. Five seconds prior to the onset of footshock, the end panel on the side on which the animal was standing was trans-illuminated until a crossing response was made. Presentation of the light was discontinued when the subject received less than 30 shocks during each of two consecutive sessions. Saline injections were begun when the subject had attained a criterion of less than 15 shocks received during each of four consecutive sessions and visual inspection of the cumulative record indicated that response rate was stable.

The daily 1-hr. activity measurement sessions for the two other groups took place between 08:00 and 18:00 hrs. Saline injections were begun on the animals' fourth session in the apparatus.

Injection Procedure

Subjects in all three groups received injections on 31 consecutive sessions. Saline injections were given on sessions 1 to 10, morphine injections on sessions 11 to 30, and a saline injection on session 31. The injection procedure was carried out exactly 15 min. after the start of the session and required the

removal of the subject from the apparatus for no more than 20 sec. All injections were intraperitoneal using 26 gauge, 3/8 in., intradermal-bevel needles. Saline injections were 0.2 cc of 0.9 percent saline solution (Laboratoire Demers) which also served as the vehicle for the morphine sulfate (May and Baker Ltd.). The latter was made up in a concentration of 10 mg/cc so that drug-injection volumes ranged between 0.17 and 0.23 cc.

RESULTS

Shuttling rate per min., activity count per min., and wheel-running count per min. were the measures of response for the three groups. Average rate or count during the 15-min. interval prior to injection was considered to be the baseline measure for a session. The relative rate or count during each of the three subsequent 15-min. intervals of each session was calculated as a percentage of the figure for the baseline interval, and is referred to below as either a response ratio, activity ratio, or running ratio, respectively.

Shuttlebox Group

Approximately 60 percent of the rats in this group were discarded because they failed to attain the criterion of receiving 15 or fewer shocks per session within 45 sessions of avoidance training. Comparable avoidance-training failure rates have been reported by Black (1958) and Kamin (1959). The remaining seven subjects attained the criterion within an average of 30 sessions (range: 14-38).

The mean number of responses emitted during each pre-injection interval and the mean number of shocks received during each session are presented in Figure 1. The number of responses emitted during the baseline period remained fairly stable and showed no significant change over the 20 sessions on which morphine was administered ($F = 0.5482$, $df = 19/114$, $p>0.05$). The mean number of shocks received by subjects tended to decrease slightly over the course of the saline sessions but did not change significantly during the 20 sessions on which morphine was administered ($F = 1.2587$, $df = 19/114$, $p>0.05$). Very few shocks were received after the initial (pre-injection) 15 min. of each session.

Mean number of responses emitted per morphine session, for blocks of four sessions, is presented in the insert of Figure 2. While there was no significant change in these means over the course of the 20 drug sessions ($F = 1.1322$, $df = 19/114$, $p>0.05$), the morphine-produced increase in mean number of responses was greater in magnitude during the second block of four sessions and then decreased.

The response ratios for certain selected sessions are presented in the main body of Figure 2. The left panel shows that response ratios changed very little during saline-session 10 but were increased by morphine, and to a progressively greater degree, during the first eight drug sessions. The right panel shows the gradual decrease in response ratios during the later drug sessions and the return to a stable level of performance on the final (saline) session of the experiment ("s-post"). Several pairs of sessions were compared using a t-test for the significance

Figure 1. Baseline performance. Upper: Mean number of responses emitted during the baseline (pre-injection) 15-min. interval of each test session. Lower: Mean total number of shocks received during each experimental session.

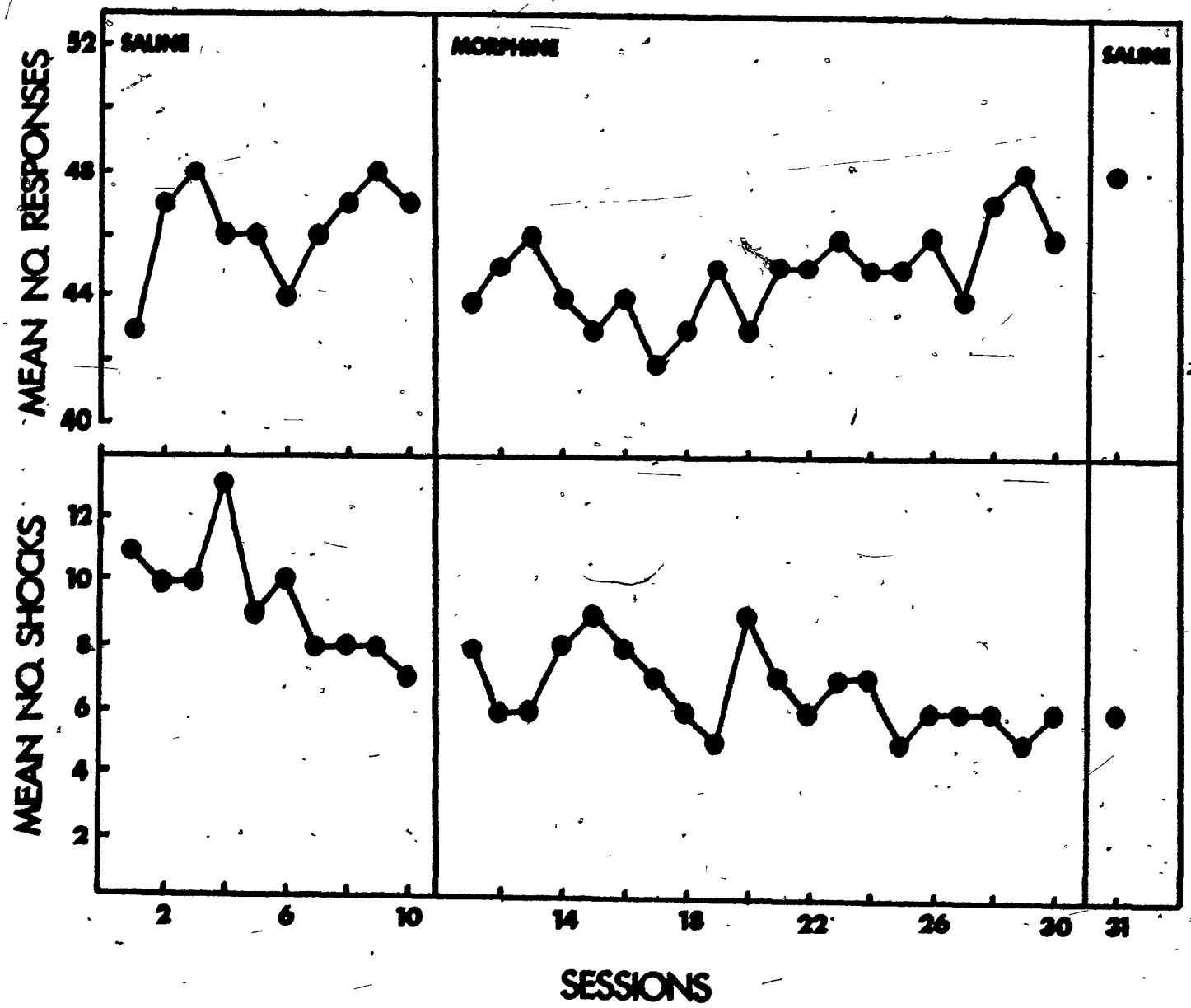
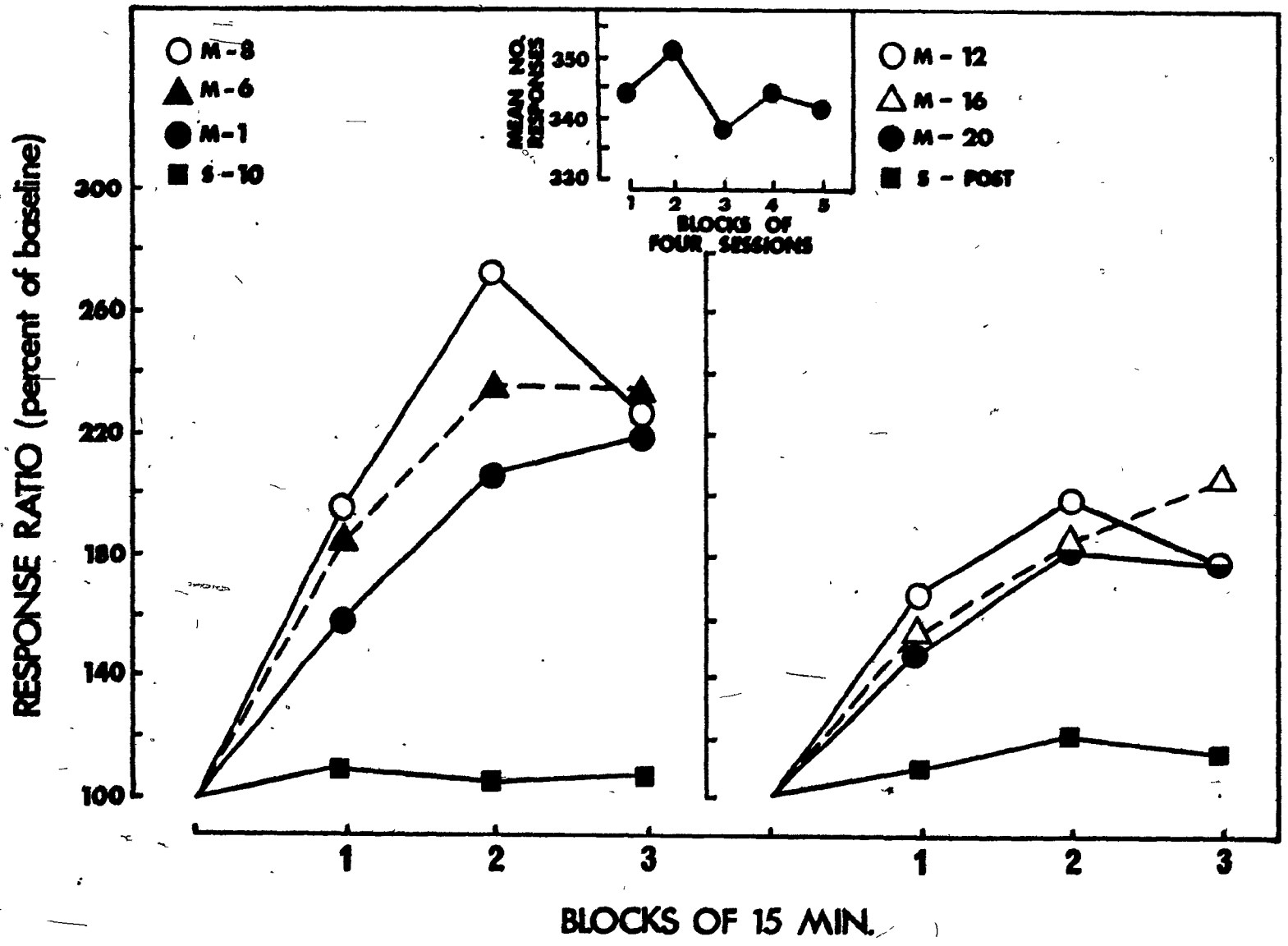

SALINE
MORPHINE
SALINE
MEAN NO. RESPONSES
52
48
44
40
MEAN NO. SHOCKS
12
10
8
6
4
2
2
6
10
14
18
22
26
30
31
SESSIONS

Figure 2. Changes in the effect of morphine on response ratios over sessions. Insert: Mean number of responses emitted per session, over blocks of four sessions, during the course of the 20 morphine sessions. Left: Mean post-injection response rate per 15-min. interval, expressed as a percentage of the corresponding baseline rate, for the tenth saline session and morphine-sessions 1, 6, and 8. Right: Mean post-injection response rate per 15-min. interval, expressed as a percentage of the corresponding baseline rate, for the final (saline) session of the experiment ("s-post").


M - 8
M - 6
M - 1
S - 10
M - 12
M - 16
M - 20
S - POST
MEAN NO. RESPONSES
350
340
330
1 2 3 4 5
BLOCKS OF FOUR SESSIONS
RESPONSE RATIO (percent of baseline)
300
260
220
180
140
100
1 2 3
1 2 3
BLOCKS OF 15 MIN.

of the difference between two means for correlated samples. The results of these comparisons are summarized in Figure 3. The upper-left panel shows that, on the first morphine session, the response ratios for the third and fourth 15-min. periods were significantly greater than for corresponding periods during the tenth saline session. As shown in the upper-right panel, this increase tended to be greater in magnitude during morphine-session 8, but was not significantly so. The panel on the lower left shows that the response ratios for the first and second 15-min. periods of morphine-session 20 were significantly less than on morphine-session 8. However, as shown in the lower-right panel, response ratios during the second and third post-injection periods of the final morphine session were still significantly greater than the corresponding ratios on the subsequent saline session. Thus, the change in mean number of responses emitted per session is paralleled by the change in the magnitude of the increase in response ratios over the course of the 20 drug sessions.

A comparison of response ratios on saline-session 10 to those on the final saline session of the experiment revealed no significant differences. This indicates that the injection procedure, as such, was not a factor in the change in morphine's effects.

Jiggle-Cage Group

Mean activity counts for baseline periods and complete sessions are presented in Figure 4. Although this group exhibited considerably more between-session variability than the shuttlebox group, there was no significant change in the mean baseline-period

Figure 3: Summary of the statistical comparison of the corresponding response ratios of selected sessions. Upper left: Saline-session 10 and morphine-session 1. Upper right: Morphine-sessions 1 and 8. Lower left: Morphine sessions 8 and 20. Lower right: Morphine session 20 and the final (saline) session of the experiment ("s-post"). The asterisks indicate significant differences between the block above which they appear and the corresponding 15-min. block in the other session plotted in the graph. One asterisk indicates $p<0.05$; two asterisks indicate $p<0.02$; three asterisks indicate $p<0.01$ ($df=6$, two-tailed t-test for correlated samples).

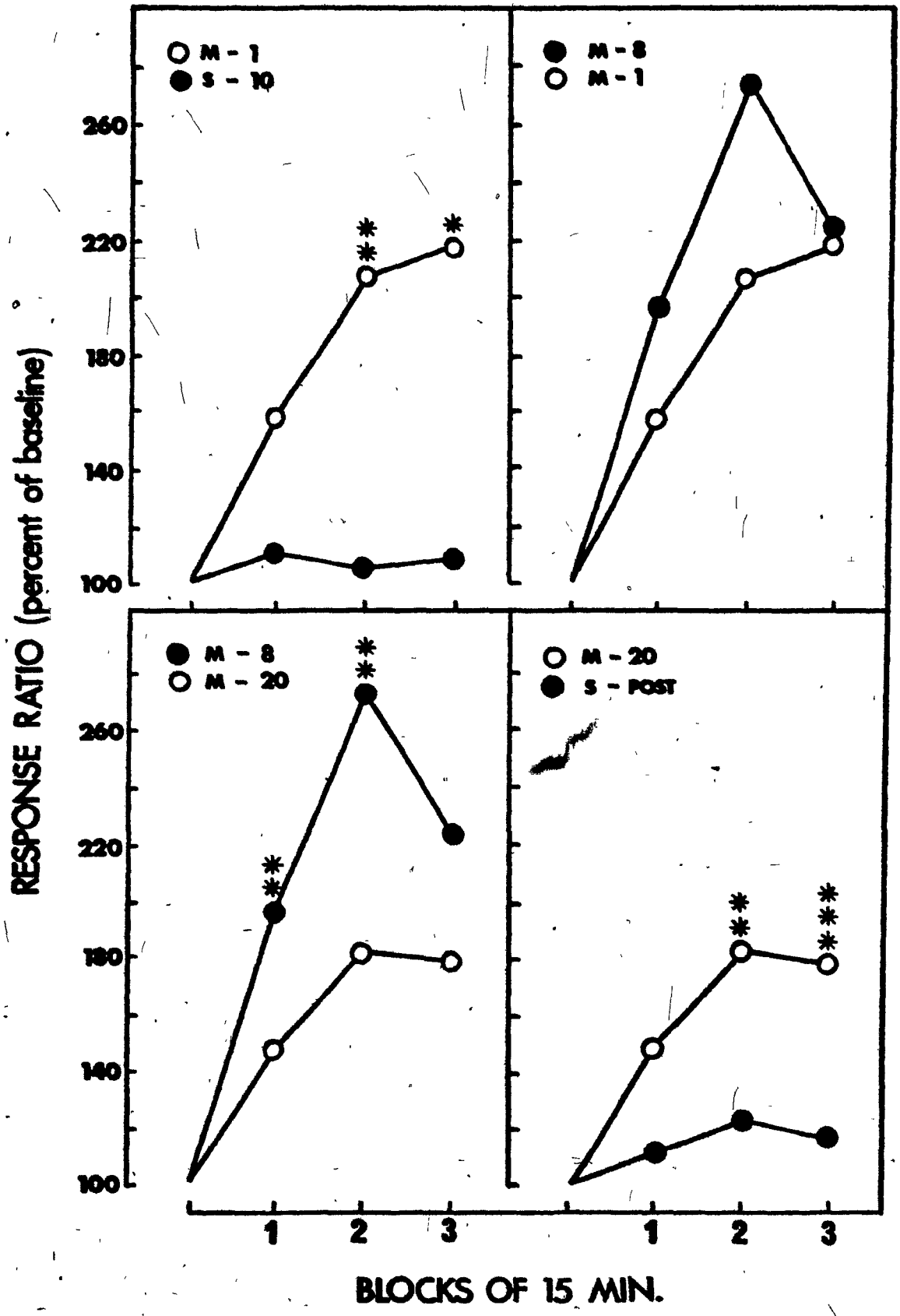
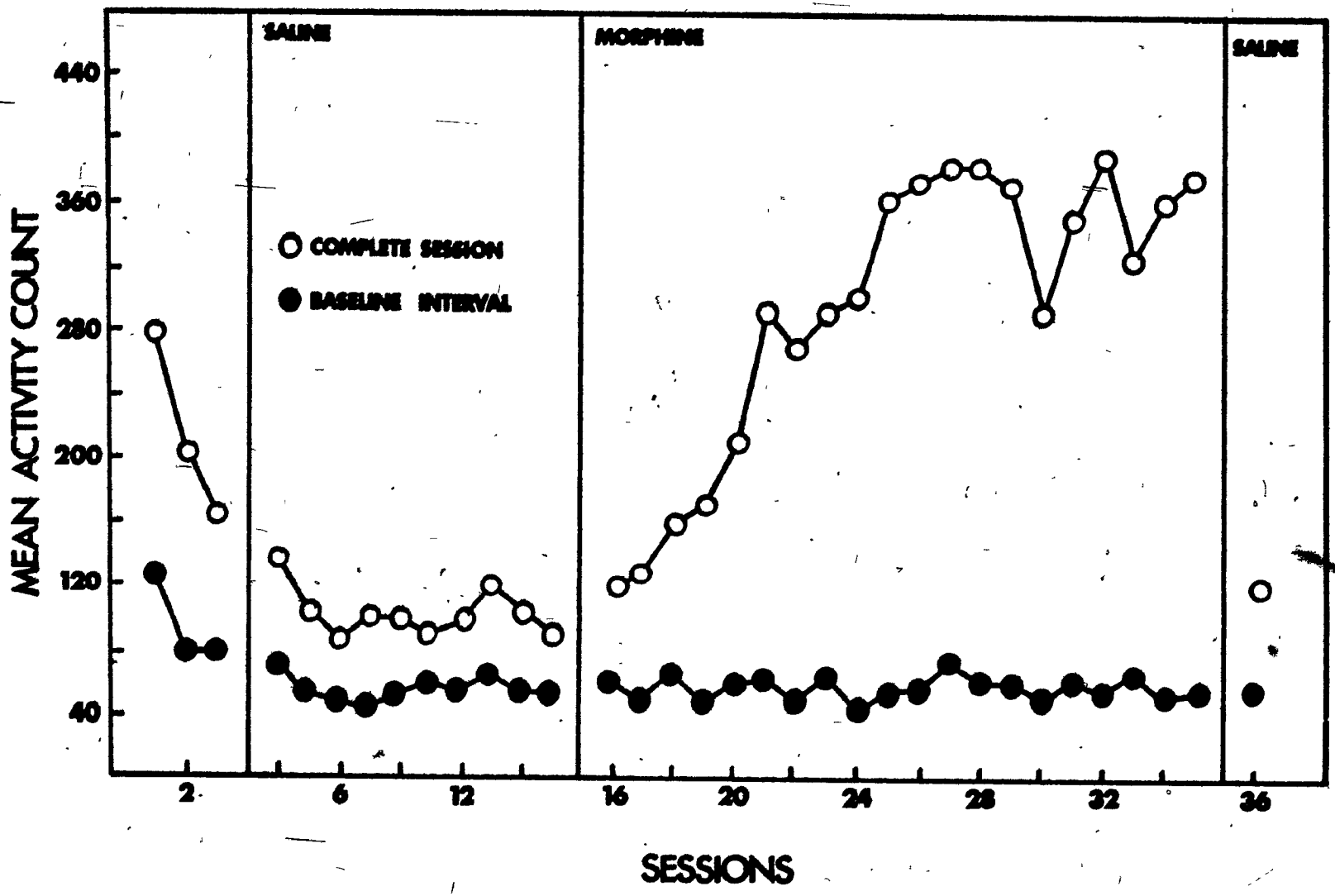

M - 1
S - 10
M - 8
M - 1
M - 8
M - 20
M - 20
S - POST
RESPONSE RATIO (percent of baseline)
100
140
180
220
260
1
2
3
BLOCKS OF 15 MIN.

Figure 4: Mean jiggle-cage activity count for each baseline interval and each complete session over the course of the experiment. Mean activity count is also presented for the first 15 min. of the three initial (familiarization) sessions, although subjects were not injected on those days.


SALINE
MORPHINE
SALINE
COMPLETE SESSION
BASELINE INTERVAL
MEAN ACTIVITY COUNT
440
360
280
200
120
40
2
6
12
16
20
24
28
32
36
SESSIONS

activity count over the course of the 20 morphine sessions ($F = 1.4459$, $df = 19/114$, $p > 0.05$). However, there was a significant increase in the mean activity count for the complete session over the course of repeated injections of morphine ($F = 4.6311$, $df = 19/114$, $p < 0.01$).

Mean activity count per morphine session, for blocks of four sessions, is presented in the insert of Figure 5. After a rapid increase during the first two blocks of sessions, the mean count apparently reached an asymptote by session 12. Activity ratios for certain selected sessions are presented in the main body of Figure 5. The left panel shows that activity decreased markedly after the baseline period on saline-session 10. Morphine initially had little apparent effect but began to increase activity ratios, and to a progressively greater degree, over the course of the first several sessions. The right panel shows that the morphine-produced increases in activity ratios changed relatively little during later drug sessions. The pattern of activity on the final (saline) session of the experiment ("s-post") is similar to that for saline-session 10.

Several pairs of sessions were compared using a t-test for the significance of the difference between two means for correlated samples. The results of these comparisons are summarized in Figure 6. The upper-left panel shows that the activity ratio during the second post-injection period was significantly greater on the final (saline) session of the experiment than on saline-session 10. This difference, though not large, was consistent from subject to subject and may indicate a slight tendency for the

Figure 5. Changes in the effect of morphine on activity ratios over sessions. Insert: Mean activity count per session, over blocks of four sessions, during the course of the 20 morphine sessions. Note that by the third block (i.e., by morphine-session 12) the total activity count appears to have reached an asymptote. Left: Mean activity count per 15-min. interval, expressed as a percentage of the corresponding baseline count, for the tenth saline session and morphine-sessions 1, 4, and 8. Right: Mean activity count per 15-min. interval, expressed as a percentage of the corresponding baseline count, for morphine-sessions 12, 16, and 20 and the final (saline) session of the experiment ("s-post").

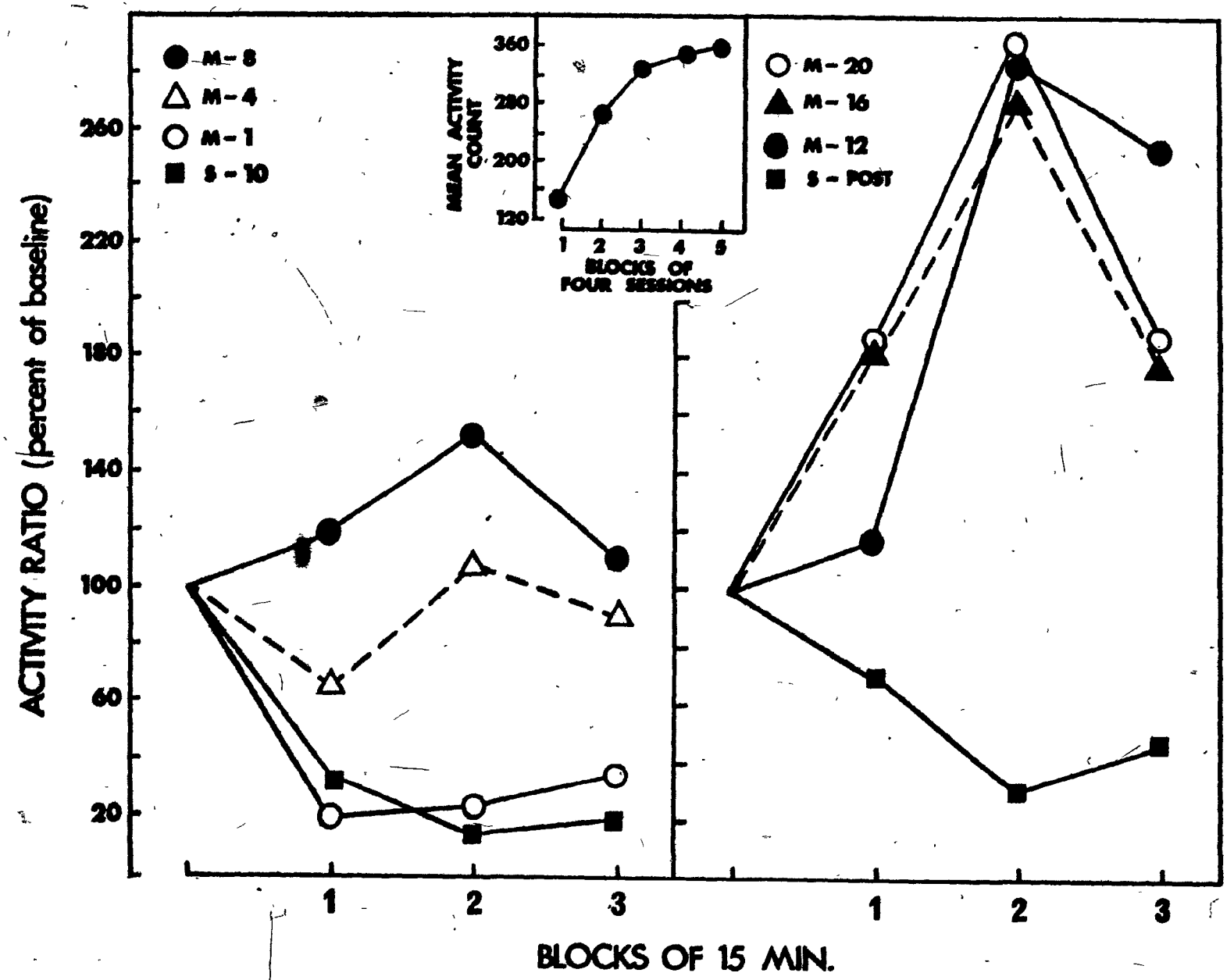
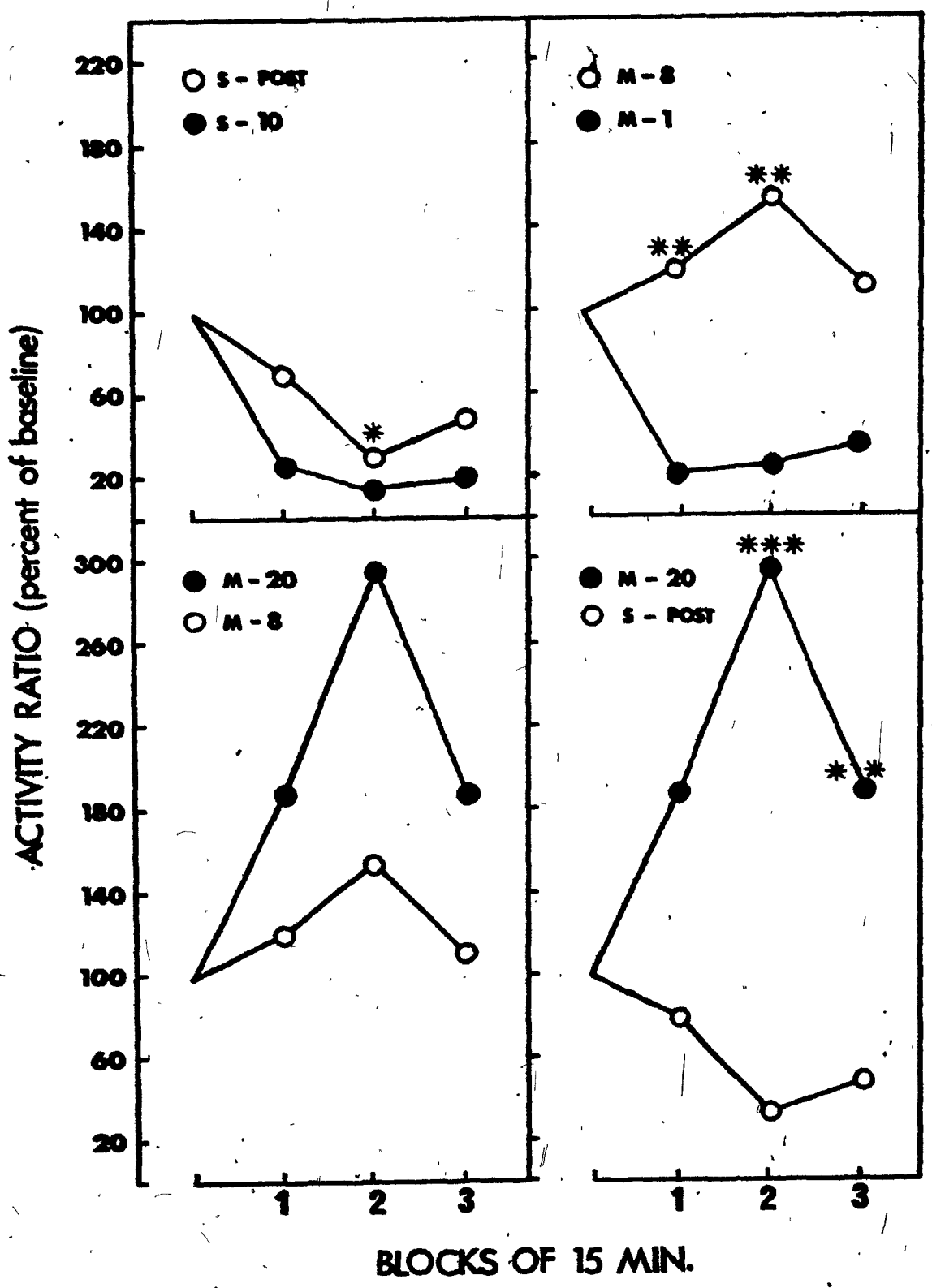

M - 8
M - 4
M - 1
S - 10
M - 20
M - 16
M - 12
S - POST
ACTIVITY RATIO (percent of baseline)
260
220
180
140
100
60
20
1
2
3
BLOCKS OF 15 MIN.
MEAN ACTIVITY COUNT
360
280
200
120
1 2 3 4 5
BLOCKS OF FOUR SESSIONS

Figure 6: Summary of the statistical comparison of the corresponding activity ratios for several sessions. Upper left: Saline-session 10 and the final (saline) session of the experiment ("s-post"). Upper right: Morphine-sessions 1 and 8. Lower left: Morphine-sessions 8 and 20. Lower right: Morphine-session 20 and the final (saline) session of the experiment ("s-post"). The asterisks indicate significant differences between the 15-min. block above which they appear and the corresponding block in the other session plotted in that graph. One asterisk indicates $p<0.05$; two asterisks indicate $p<0.02$; three asterisks indicate $p<0.01$ ($df= 6$, two-tailed t-test for correlated samples).

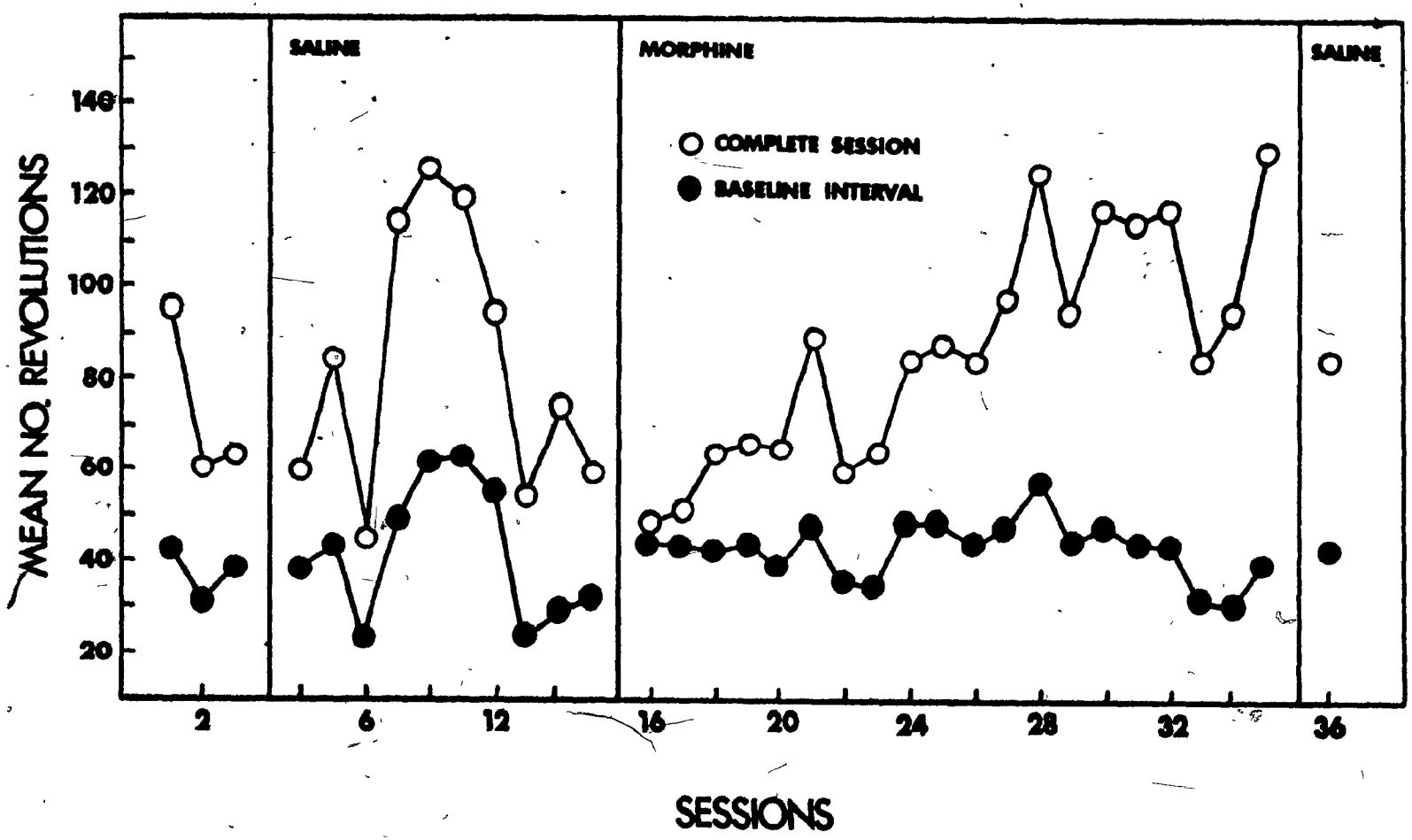

S - POST
S - 10
M - 8
M - 1
M - 20
M - 8
M - 20
S - POST
ACTIVITY RATIO (percent of baseline)
220
180
140
100
60
20
300
260
220
180
140
100
60
20
1
2
3
BLOCKS OF 15 MIN.

injection procedure to elicit an increase in spontaneous activity. The upper-right panel shows that the increase in activity ratio was significantly greater during the first and second post-injection periods of morphine-session 8 than on the first morphine session. The lower-left panel shows that the increase in activity ratios tended to be greater in magnitude by the final morphine session, but was not significantly so. As shown in the panel on the lower-right, the activity ratios for the second and third post-injection periods of morphine-session 20 were significantly greater than those during the final (saline) session of the experiment. Thus the change in mean activity count per session is paralleled by the change in activity ratios over the course of the 20 drug sessions. The progressive increase in activity ratios reached an asymptote by morphine-session 12 and, in contrast to the shuttlebox group, remained relatively stable during later drug sessions.

Running-Wheel Group

Of the three groups used in the present study, the running-wheel group was influenced the least by morphine. The mean numbers of running-wheel revolutions for baseline periods and complete sessions are presented in Figure 7. Between-session variability was high, as for the jiggle-cage group. There was no significant change in the number of wheel revolutions during the baseline periods over the course of the 20 morphine sessions (F= 1.3908, df= 19/114, $p>0.05$). Also, while there was a gradual increase in total revolutions per session following repeated administrations of morphine, this increase did not attain

Figure 7. Mean number of wheel revolutions for each baseline interval, and each complete session, over the course of the experiment. Mean number of revolutions is also presented for the first 15 min. of the three initial (familiarization) sessions, although the subjects were not injected on those days.


SALINE
MORPHINE
SALINE
COMPLETE SESSION
BASELINE INTERVAL
MEAN NO. REVOLUTIONS
140
120
100
80
60
40
20
2
6
12
16
20
24
28
32
36
SESSIONS

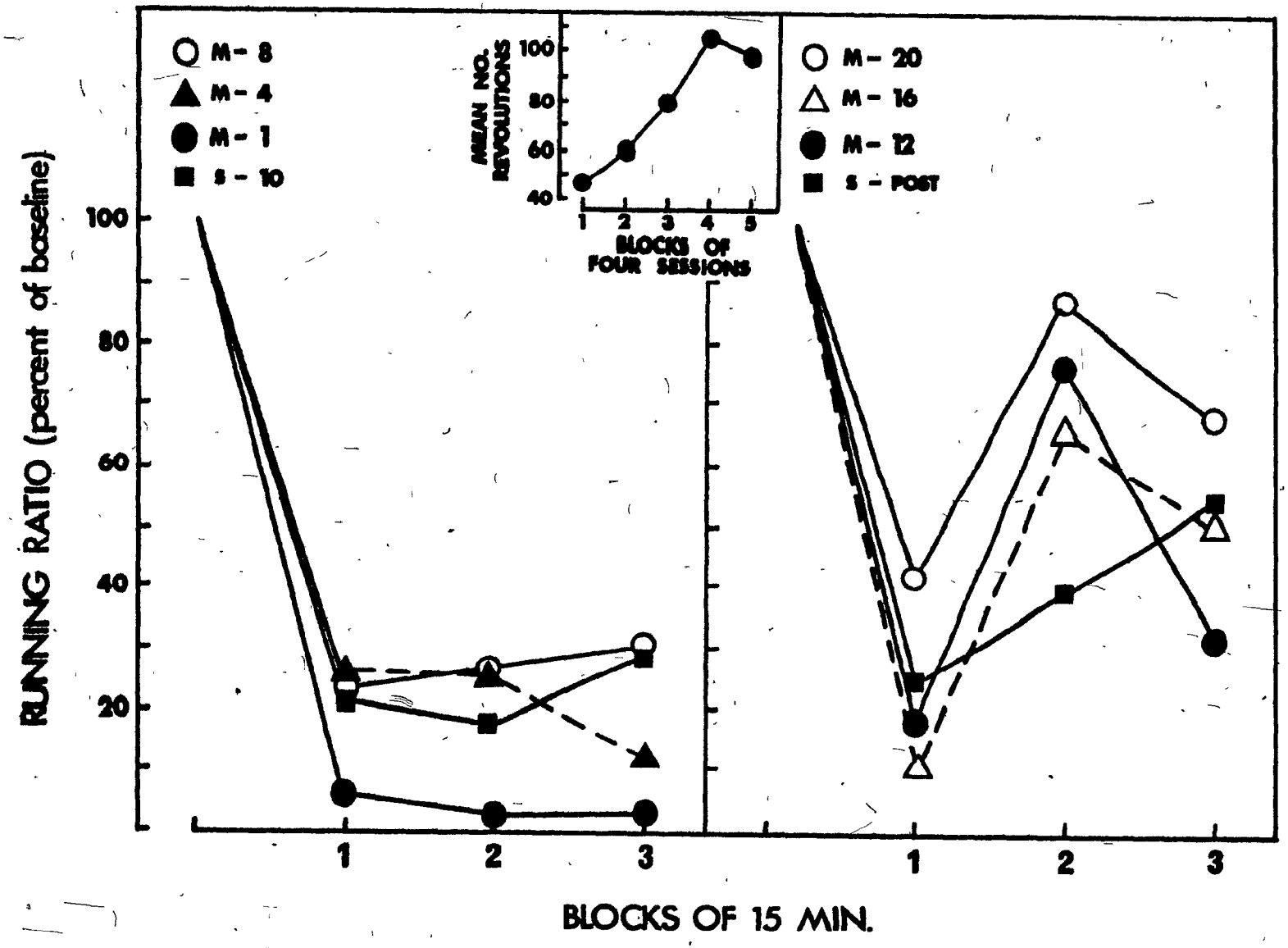
statistical significance ($F = 1.5671$, $df = 19/114$, $p > 0.05$).

Mean number of wheel revolutions per morphine session, for blocks of four sessions, is presented in the insert of Figure 8. The mean increases until it apparently reaches an asymptote by session 16. Running ratios for certain selected sessions are presented in the main body of Figure 8. The left panel shows that, compared to saline session 10, the first injection of morphine decreased running ratios while the next few injections produced little apparent effect. The right panel shows that the later morphine injections tended to increase running ratios somewhat, particularly during the second post-injection interval, when jiggle-cage activity ratios also tended to be highest. However, neither the initial suppression nor the later increase attained statistical significance ($df = 6$, two-tailed t-test for correlated samples).

DISCUSSION

The first morphine injection produced a facilitation of shuttling behaviour similar in magnitude to that observed for lever pressing by Holtzman and Jewett (1972). They report that 4 mg/kg of morphine increased avoidance-response rate to 180 percent of control values; in the present study the increase was 220 percent after 5 mg/kg. This magnitude of the initial facilitation of shuttling behaviour is also consistent with the first-session increase in "large horizontal movement" in a circular alley, following the same dose (Babbini & Davis, 1972). On the other hand, the first injection of morphine had no clear effect

Figure 8. Changes in the effect of morphine on running ratios over sessions. Insert: Mean number of revolutions per session, over blocks of four sessions, during the course of the 20 morphine sessions. Left: Mean number of revolutions per 15-min. interval, expressed as a percentage of the respective baseline value, for saline-session 10 and morphine-sessions 1, 4, and 8. Right: Mean number of revolutions per 15-min. interval, expressed as a percentage of the corresponding baseline value, for morphine-sessions 12, 16, and 20 and the final (saline) session of the experiment ("s-post").


M - 8
M - 4
M - 1
S - 10
M - 20
M - 16
M - 12
S - POST
MEAN NO. REVOLUTIONS
100
80
60
40
1 2 3 4 5
BLOCKS OF FOUR SESSIONS
RUNNING RATIO (percent of baseline)
100
80
60
40
20
1 2 3
1 2 3
BLOCKS OF 15 MIN.

on spontaneous activity in the jiggle cage. The decrease in wheel running produced by the first morphine injection is puzzling in view of the similarity of certain of its behavioural components to those of shuttling behaviour. However, Cofer and Appley (1964) have reviewed the relations among various activity measures and have concluded that wheel running is "fundamentally different" from the types of activity measured by a tilt (or jiggle) cage or runway. This would seem to be borne out in the present results, as well as by those of Lorens and Mitchell (1973), who found no increase in wheel running at doses which produced a significant facilitation of lever pressing for electrical stimulation of the brain. The above-mentioned differential effects of the initial dose of morphine on the shuttling response, spontaneous activity, and wheel running, suggest that morphine's acute effect on a given behaviour depends upon the specific topography of behaviour under investigation.

The progressive increase in the facilitation of both avoidance behaviour and spontaneous activity over the course of the initial 8 to 10 morphine sessions is similar to that observed at similar doses by Lorens and Mitchell (1973). This increase may reflect the gradual "unmasking" of the full extent of morphine's behavioural stimulation or else it may be a potentiating effect of repeated administration of the drug; i.e., a sensitization to morphine. Seevers and Deneau (1963) and Kayan, Woods, and Mitchell (1971) have suggested that the increase represents unmasking due to the development of tolerance to the depressant component of morphine's action.

The finding of chief interest in the present study is the differential response to morphine of the shuttlebox and jiggle cage groups over the course of morphine sessions 10 to 20. These results demonstrate that with daily injections of 5 mg/kg of morphine over 20 days, rats develop tolerance to the drug's facilitatory effect on shuttlebox avoidance behaviour, but not to its facilitatory effect on spontaneous activity as measured in the jiggle cage. The decrease in shuttle-response facilitation, over the last 10 morphine sessions, confirms Holtzman's (1974) report of the development of tolerance to morphine's facilitatory effect on free-operant avoidance behaviour. It appears that the effect of chronically administered morphine does not depend upon the topography of behaviour under investigation. This is indicated by the fact that tolerance developed to morphine's facilitation of lever pressing (Holtzman, 1974), which does not resemble shuttle behaviour in terms of topography. Moreover, tolerance was not found for morphine's facilitation of "large, horizontal movements" (Babbini & Davis, 1972), which does resemble the shuttle response used in the present study. The decrease in morphine's facilitation of shuttling behaviour is probably not a consequence of nonspecific factors such as physical debilitation resulting from multiple intraperitoneal injections. This is indicated by the stability of baseline performance over the 20 drug sessions and also by the absence of a corresponding decrease in the facilitation of the activity of jiggle-cage subjects. The finding of no decrease of morphine's effect on this group is consistent with Babbini and Davis' (1972) report that tolerance to morphine does not influence the facilitatory effects of the drug on spontaneous activity.

Siegel (1975) has recently proposed a conditioning theory of tolerance in which compensatory responses, opposite in direction to the unconditional effects of the drug, are elicited by environmental stimuli which reliably precede the onset of the drug's systemic effects. The close similarity between the shuttlebox group's performance on the final saline session of the experiment, and that on saline-session 10, appears to be inconsistent with this proposal. However, it is possible that a compensatory response would have been apparent during the final saline session if the morphine injections had been continued until the development of tolerance was complete; i.e., until facilitation of shuttling behaviour was no longer apparent.

It remains to be determined why tolerance has not been observed in testing situations in which subjects are not required to perform some task. The results of the present experiment, and those of Schuster and Zimmerman (1961), support Kalant et al.'s (1971) hypothesis that drug tolerance is a functional compensation which is specific to the situation in which the drug-produced impairment is experienced, rather than a fundamental alteration of the drug's central effects. While a drug may initially have the same effect on a certain behaviour in two situations, differing degrees of tolerance will develop if the situations vary in the extent to which they make a "functional demand" upon the subject. Investigations of the endocrine and cardiovascular consequences of free-operant avoidance training indicate that, in terms of stress, this behavioural paradigm makes a considerable demand upon the rat (Harris & Brady, 1974). Morphine's facilitation

of free-operant avoidance behaviour may constitute an impairment in that it alters the efficient performance, characteristic of the baseline period, to a rapid and somewhat irregular pattern resembling that seen early in training. In contrast, the low level of arousal characteristic of subjects in the jiggle-cage group was indicated by the fact that they frequently appeared to be asleep during the latter part of each saline session. Hence, if it develops at all, tolerance to morphine's facilitation of spontaneous activity might be expected to develop to a much lesser extent than for its facilitatory effect on free-operant avoidance behaviour.

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