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The occurrence of tolerance to a central depressant effect of nicotine

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Summary

1. Rats were injected with 0·8 mg nicotine/kg, 0·8 mg amphetamine/kg or with saline immediately before being tested for 30 min in activity boxes.

2. During the first 3 trials the nicotine group were less active than the controls but from trial 5 onwards nicotine had a stimulant effect. The stimulant effect of the amphetamine did not alter with repeated injection.

Introduction

Nicotine resembles amphetamine in its effects on bar-pressing behaviour of the rat independently of the nature of the reinforcement (Bovet & Gatti, 1965 ; Wanner & Bättig, 1968 ; Morrison, 1968a, 1969). However, Dews (1953) and Morrison & Armitage (1967 and unpublished observations) failed to demonstrate an amphetamine-like stimulant effect of nicotine on motor activity in mice and rats. Kuschinsky & Hotovy (1943) had previously reported that nicotine could increase activity. Further experiments have now been performed to reinvestigate the effects of nicotine on activity in the rat. During these it was found that tolerance developed to the depressant properties of nicotine. These findings are now reported.

Methods

Apparatus

Activity was recorded in grey plastic boxes measuring 24 by 36 by 20 cm. Electrically earthed bars in the floor alternated with others across which a low electric current was maintained. A rat placed in the box created a short-circuit between adjacent bars and any change in the rat's position activated a relay system. Activity counts were recorded at 5 min intervals by print-out counters which were in a separate room from the activity boxes. The test room was darkened during experiments.

Subjects

Twenty four male Sprague-Dawley rats were housed in groups of 4 under natural lighting conditions. Food and water were available at all times in the home cages.

Procedure

The rats were assigned to three treatment groups by means of a modified latin square design. They were injected subcutaneously with 0·9% NaCl w/v (saline),

0·8 mg nicotine/kg or 0·8 mg amphetamine/kg on each test day immediately before being placed singly in the activity boxes for 30 minutes. A rat received the same treatment each day and was tested at the same time of day in the same box. The 10 trials were on consecutive days except for intervals of 2 days (weekends) between trials 4 and 5 and trials 9 and 10.

Additional Experiments

In four further experiments the effects were examined of daily subcutaneous injections of 0·4 mg nicotine/kg on the activity of Sprague-Dawley (1 experiment) and Lister black-hooded (3 experiments) male rats.

Drugs

(+)-Amphetamine sulphate and nicotine hydrogen tartrate were dissolved in saline to give an injection volume of 0·1 ml/100 g body weight. Doses are expressed as base.

Results

Figure 1 shows the mean total activity scores for the three groups of rats during the 10 trials. The daily scores of the saline control rats fell slightly as the experiment progressed with a small increase in trial 10 (a Monday). The group that received amphetamine were much more active than the controls in all trials and the size of the response did not change with repeated dosing. The activity of the group that received nicotine, on the other hand, increased steadily during the course of the experiment. Their activity was less than that of the controls during trials 1–3 and greater during trials 5–10.

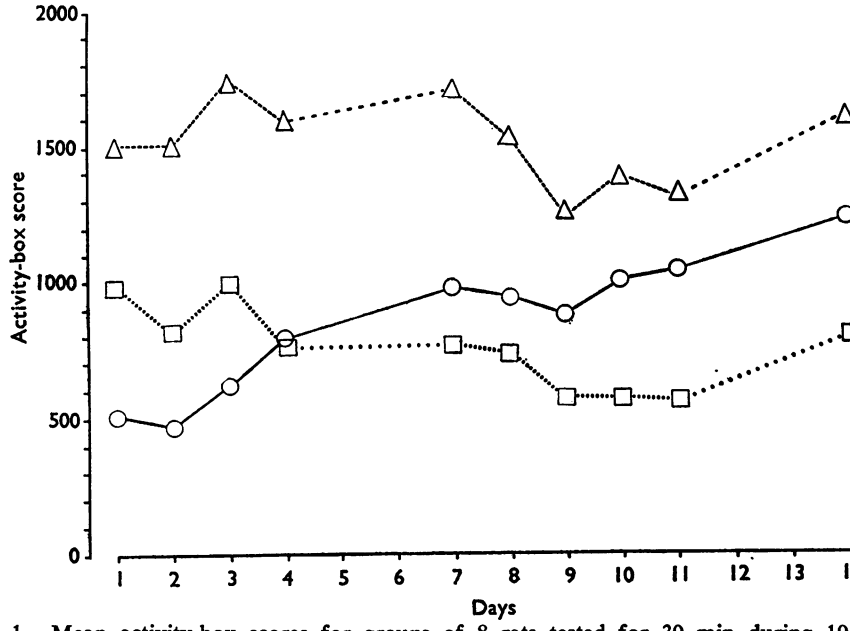

FIG. 1. Mean activity-box scores for groups of 8 rats tested for 30 min during 10 trials. Rats injected with 0·8 mg nicotine/kg (○——○——○), 0·8 mg amphetamine/kg (△--△--△) or saline (□-□-□) immediately before testing. Ordinate; activity-box score. Abscissa; days.

In the saline control and amphetamine groups the pattern of activity within each 30 min trial did not alter during the experiment (Fig. 2). The activity scores of the control rats decreased steadily during the 30 minutes. The increase in activity caused by amphetamine was most apparent during the second half of each trial. However, the pattern of activity of the nicotine group changed during the course of the experiment. In the first three trials nicotine caused an initial reduction in activity relative to the controls. By trial 4 this depressant effect had virtually disappeared and during trials 5–10 the nicotine group showed a clear phase of stimulation in the first 15 minutes. Between 15 and 20 min after the start of the trial the activity of the nicotine group was always close to that of the controls, then there was a small but significant increase in activity during the last 5 or 10 min of each trial.

Analysis of variance (Table 1) confirmed that the differences among the treatments within the trials (drugs × treatments) and the change in the effect of nicotine from depression to stimulation with repeated dosing (days × intervals × treatments) were statistically significant.

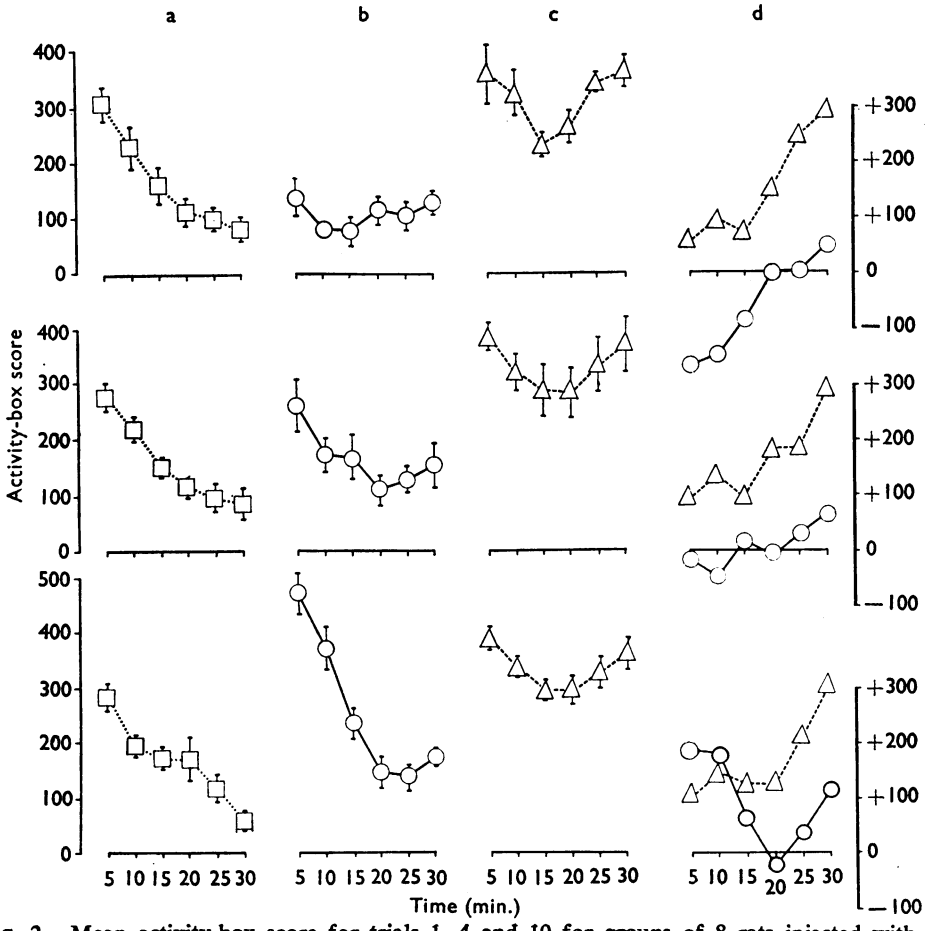

FIG. 2. Mean activity-box score for trials 1, 4 and 10 for groups of 8 rats injected with a, saline, b, 0·8 mg nicotine/kg (○——○——○) or c, 0·8 mg amphetamine/kg (△--△--△) immediately before testing. d, Drug scores plotted relative to control scores, control scores as baseline. Ordinates; activity-box score. Abscissae; time in minutes from the start of the trial.

In all 4 further experiments 0·4 mg nicotine/kg injected subcutaneously either reduced or did not affect activity; after approximately 5 trials, however, the scores of the nicotine groups were significantly higher than those of the controls. No reduction in the stimulant effect was found after 24 trials. The depressant effect of nicotine could be prevented by 4 or 5 daily injections of nicotine before testing in the activity boxes. Figure 3 compares the effects of a 5th injection of 0·4 mg nicotine/kg on the activity of rats experiencing either their first (a) or their fifth

TABLE 1. *Analysis of variance*

Source	df	SS	MS	F	P
Treatments (T)	2	7,598,671·66	3,799,335·83	23·53	<0·001
Rats (R) (within treatments)	21	3,390,159·86	161,436·18	1·00	
Intervals (I)	5	2,827,225·86	565,445·17	61·30	<0·001
I×T	10	1,539,859·44	153,985·94	16·69	<0·001
I×R (within treatments)	105	968,582·12	9,224·59	1·00	
Days (D)	9	570,589·22	63,398·80	4·15	<0·001
D×T	18	1,264,917·87	70,273·21	4·60	<0·001
D×R (within treatments)	189	2,889,943·16	15,290·70	1·00	
D×I	45	904,506·48	20,100·14	5·29	<0·001
D×I×T	90	715,799·78	7,953·33	2·09	<0·001
D×I×R (within treatments)	945	3,587,892·48	3,796·71	1·00	

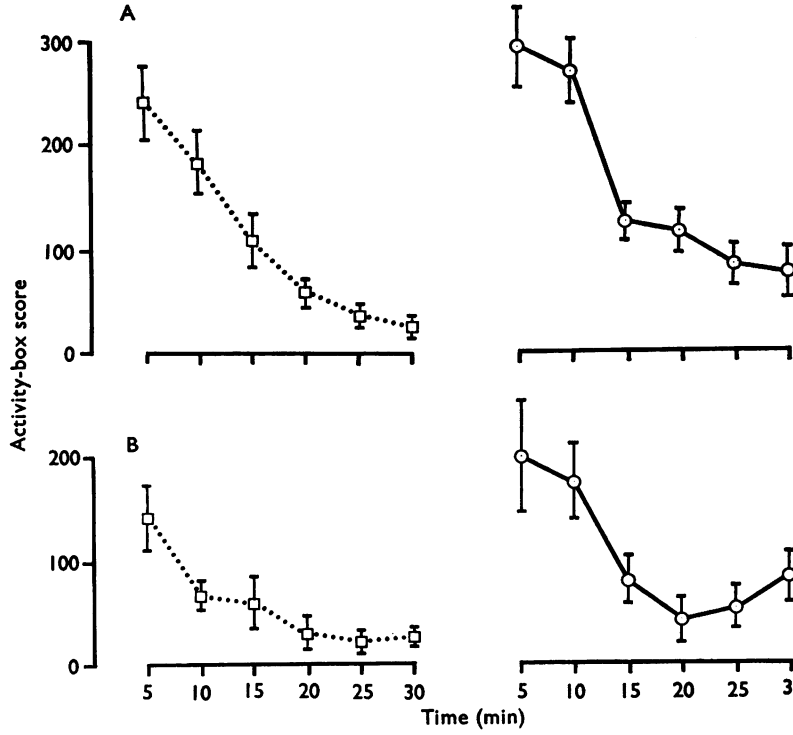

FIG. 3. Mean activity-box scores for groups of 8 rats which had received their 5th daily injection of saline (□---□---□) or 0·4 mg nicotine/kg (○——○——○) immediately before testing. Upper part: rats having no previous experience of activity boxes. Lower part: rats which had been tested in the activity boxes immediately after each daily injection. Ordinates; activity-box score. Abscissae; time in minutes from the start of the trial.

(b) trial in the boxes. In both cases nicotine increased activity, though the activity of the (b) rats had been reduced by their first injection of nicotine. On one occasion there was an interval of 23 days between two consecutive nicotine tests but the initial depressant effect did not reappear. The depressant effect of the first injection of nicotine was not affected by previous exposure to the experimental situation ; the reduction in activity was similar for rats receiving their first injection of nicotine immediately before the first or the fifth trial in the activity boxes.

Discussion

These experiments have demonstrated that rats developed a tolerance to the depressant effects of nicotine. This depended on the repeated administration of the drug and not on repeated experience of the experimental situation. Once the tolerance had developed it could, rather surprisingly, persist in the absence of further injections of nicotine. Nicotine has an initially depressant effect on bar-pressing behaviour in the rat and this has been shown to be a central effect (Morrison, 1968b ; Morrison, Goodyear & Sellers, 1969). It seems reasonable to assume that the depressant effect of nicotine on activity is also central, and the present results suggest a rapid adaptation to a central effect of nicotine. This may parallel the adaptation or development of tolerance to the unpleasant effects of smoking, such as nausea and sweating, experienced by the novice smoker.

When the depression caused by nicotine had been abolished a stimulation of activity became apparent. The pattern of stimulation was distinguishable from that of amphetamine, the greatest stimulant effect of nicotine occurring at the beginning and the greatest effect of amphetamine occurring towards the end of the trial. If the activity of the amphetamine group is compared to that of the controls then it can be shown to be a steadily increasing stimulation which could be accounted for in terms of gradual drug absorption. The stimulant effects of nicotine were most pronounced during the first 10 and the last 5 min of the 30 min trial. Subcutaneously injected nicotine is rapidly absorbed and peak blood nicotine concentrations are reached in the cat approximately 20 min after subcutaneous injection of 1·0 mg nicotine/kg (D. M. Turner, unpublished results). The period of reduced stimulation of activity found in the rat 15–25 min after injection of 0·8 mg nicotine/kg may therefore coincide with peak blood concentrations of nicotine. Possibly the rats were very active, with the activity taking a form, such as rearing, not detected by the activity boxes. Alternatively some central mechanism might be inhibited by high nicotine levels and activity slightly reduced. Since the experiments were carried out in darkness no observations of the rats' actual behaviour during this period were made.

Nicotine has now been shown to increase, decrease or to have no effect on the activity scores of rats in activity boxes, depending on the animals' previous experience of nicotine. This could explain the apparent inconsistencies in the effects of nicotine on activity previously reported.

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