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Aversive Environmental Stimuli as a Factor in the Psychostimulant Response to Nicotine

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VALE, A. L. AND D. J. K. BALFOUR. *Aversive environmental stimuli as a factor in the psychostimulant response to nicotine.* PHARMACOL BIOCHEM BEHAV **32**(4) 857–860, 1989.—Saline-treated rats tested on an elevated open platform were less active ($p<0.01$) than those tested on an enclosed platform of the same dimensions. Acute nicotine (0.05, 0.1 and 0.4 mg/kg SC) increased the activity ($p<0.01$) of rats tested on the open platform but had no effect on activity measured on the enclosed platform. When injected chronically, the highest dose tested increased the activity of rats tested on both platforms, whereas the two lower doses continued to exert selective effects on the activity of rats tested on the open platform. d-Amphetamine (0.1 to 0.5 mg/kg SC) and cocaine (5 and 15 mg/kg IP) evoked dose-dependent inceases in activity which were independent of the test environment used. It is concluded that nicotine appeared to be a more effective psychostimulant in the rats tested on the open platform because, at doses lower than those needed to evoke general psychostimulation, it attenuated the reduction in activity caused by exposure to the more aversive environment.

Nicotine Spontaneous locomotor activity d-Amphetamine Cocaine Aversive environmental stimuli

IT is generally accepted that nicotine is the principal rewarding component of tobacco smoke and that many people who smoke tobacco become dependent upon the drug (2,5). The predominant behavioural response to nicotine is psychostimulation, particularly when the drug is given chronically, and there is evidence that, like other psychostimulants, nicotine stimulates the dopamine (DA)-secreting pathways in the mesolimbic system of the brain (9,10). It seems likely that the effects of nicotine on this pathway contribute to both its stimulant and rewarding properties although, when compared with other psychostimulants, it is a relatively weak substrate when used in self-administration studies with infrahuman species (5,19). Smokers frequently report that tobacco smoke exerts a "tranquillising" effect and that the craving to smoke is enhanced when they are exposed to aversive environmental stimuli (7,16). It is assumed that nicotine is the agent responsible for this effect although it would be an unusual one for a stimulant drug to possess and, to date, there is no convincing evidence to show that nicotine has any of the pharmacological properties of an anxiolytic drug (3,11). Nevertheless, there is evidence that nicotine self-administration is greatly enhanced in animals exposed to aversive environmental stimuli (8) and that nicotine dependence may develop more readily if it is given to animals placed in an aversive environment (12). The purpose of the study reported in this paper was to test the hypothesis that aversive environmental stimuli also influence the psychomotor response to the drug.

METHOD

Animals

The animals used for the study were male Sprague-Dawley rats bred in the Animal Services Unit, Dundee University Medical School from stock purchased from Charles River (UK) Ltd. They were housed in groups of three in a room which was lit between 0800 hr and 2000 hr daily and allowed free access to food and water. They weighed 180–220 g at the beginning of the experiments.

Apparatus

The spontaneous activity of the rats was tested using either an elevated open platform (40 cm square) or a platform of the same dimensions enclosed with 25 cm high sides. The platforms were raised 1 m from the laboratory floor. The activity of the rats was monitored using infrared photobeams placed at 13 cm intervals along two adjacent sides of the platforms, each interruption of one of the photobeams being recorded electronically using recording equipment designed and built in the Department of Medical

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Behavioural Experiments

The responses to acute nicotine were measured using groups of rats (N = 10 per group) treated subcutaneously (SC) with nicotine (0.05, 0.1 or 0.4 mg/kg) or saline (controls) 3 minutes prior to being placed on one of the platforms for 10 minutes. Each rat was tested once on both platforms with a 5-day interval between the trials, half the rats in each treatment group being tested first on the open platform, the remainder being tested first on the enclosed platform. The psychostimulant response to higher doses of nicotine is reported to be enhanced if the animals are pretreated with the drug in order to make them tolerant to the depressant effects on locomotor activity which are observed when these higher doses are given acutely (7). Therefore, in order to investigate the effects of platform design on the psychomotor response in nicotine-tolerant rats, groups of rats (N = 6 per group) were given daily injections of nicotine (0.05, 0.1 or 0.4 mg/kg) or saline for 6 days. On day 7 and day 12 of the experiment, the rats were again injected with saline or nicotine and, 3 minutes later, tested on the open or enclosed platform using the counter-balanced design described above.

In another experiment, designed to investigate the effects of nicotine on habituation to the platforms, the rats (N = 7 per group) were given 24 daily injections of saline or nicotine (0.1 or 0.4 mg/kg) and then tested on the platforms for 10 minutes after each injection. In this experiment each group of rats was tested repeatedly on the same platform.

The responses to acute d-amphetamine were examined in rats treated subcutaneously with the drug (0.1, 0.3 or 0.5 mg/kg) 30 minutes prior to the trial. The effects of acute cocaine were examined in rats treated intraperitoneally with the drug (5 or 15 mg/kg) 30 minutes prior to the trial. The doses and routes of administration for each of the drugs were selected on the basis of previous studies on their psychostimulant properties.

Statistical Analysis

The data were analysed using a two-way analysis of variance (ANOVA) with drug treatment and the platform design as the two independent factors analysed. An ANOVA for repeated measures was used to analyse the data for the experiment involving repeated exposure to the platforms.

Drugs

The drug solutions were prepared by dissolving nicotine hydrogen tartrate (British Drug Houses), d-amphetamine sulphate or cocaine hydrochloride (Sigma) in saline and, when necessary, adjusting the pH to 7.4 by the addition of a small quantity of NaOH. The drug doses are all expressed as free base.

RESULTS

The administration of nicotine was associated with a significant increase, $F(7,120) = 14.0$, $p<0.001$, in the locomotor activity of the rats (Fig. 1). The reduction in activity evoked by exposure to the elevated open platform was also significant, $F(1,120) = 106$, $p<0.001$. Further analysis of the data showed that the response to nicotine was influenced by the design of the platform on which the rats were tested [drug × platform design, $F(7,120) = 4.4$, $p<0.001$]. The administration of nicotine to the rats tested on the open platform resulted in an increase in activity, $F(7,60) = 17.5$, $p<0.001$, the increase evoked by each dose of nicotine being statistically significant (Duncan's test, $p<0.01$) when compared

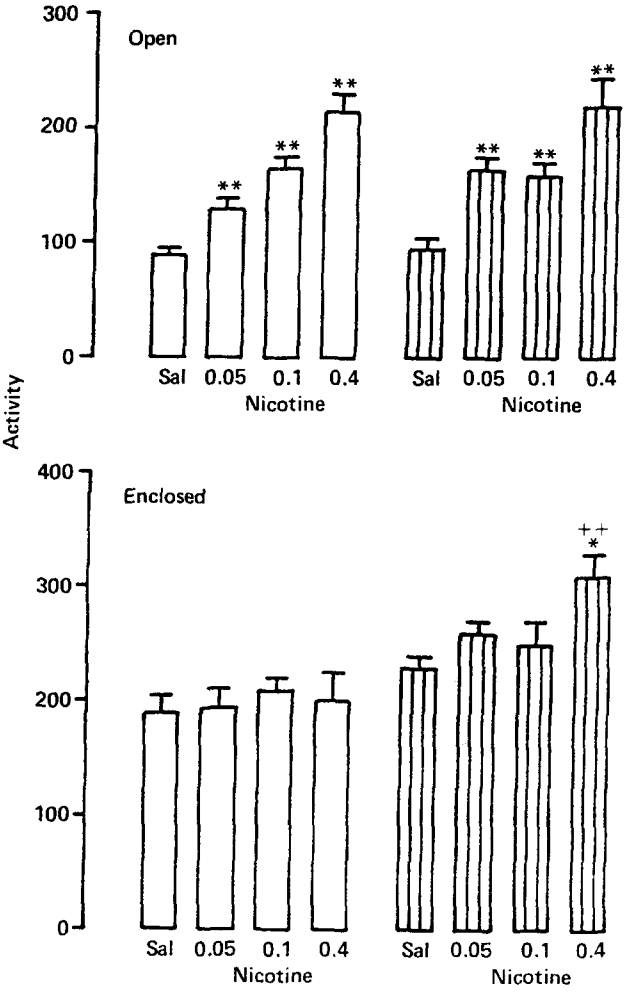

FIG. 1. The rats were treated acutely (open columns) or subchronically for 7 days (striped columns) with saline (Sal) or nicotine (0.05 to 0.4 mg/kg SC) prior to the trial (10 min). The animals were tested once in each environment 3 minutes after the injection. The results are means ± SEM of 10 observations (acute study) or 7 observations (subchronic study). The increase in activity evoked by nicotine was significant, $F(7,117) = 14$, $p<0.001$. The drug × platform interaction was also significant, $F(7,117) = 4.4$, $p<0.001$. Significantly different from saline-treated controls (Duncan's test) $^{**}p<0.01$; significantly different form rats treated acutely with the same treatment $^{++}p<0.01$.

with the saline-treated controls. Nicotine also increased the activity of the rats tested on the enclosed platform, $F(7,60) = 5.1$, $p<0.001$. However, post hoc analysis of the data (Duncan's test) indicated that, when compared with saline-treated controls, nicotine only increased activity when it was given subchronically at the highest dose tested (0.4 mg/kg). The activity of the rats tested on the enclosed platform following subchronic treatment with 0.4 mg/kg nicotine was also greater ($p<0.01$) than the activity of rats treated acutely with the same dose of the drug. However, subchronic treatment did not enhance the respoonse to nicotine (0.05 to 0.4 mg/kg) if the rats were tested on the open platform. The rats treated with saline or the lower doses of nicotine (0.05 or 0.1 mg/kg) were invariably less active when tested on the open platform (*t*-test, $p<0.01$). In contrast, the activity of the rats treated acutely with 0.4 mg/kg was not significantly reduced by exposure

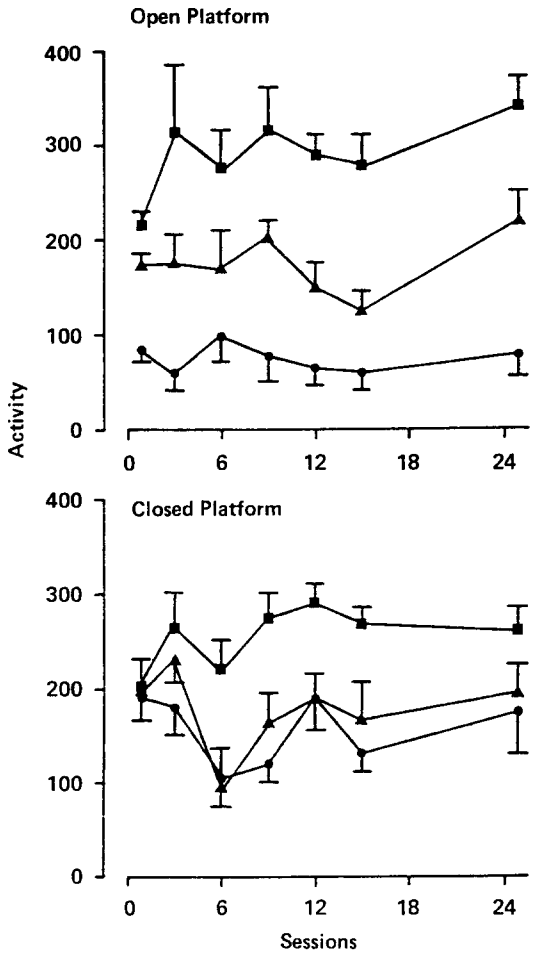

FIG. 2. The rats treated with saline (●) or nicotine (0.1 mg/kg ▲; 0.4 mg/kg ■) 3 minutes prior to each trial (10 min). The results are means ± SEM of 7 observations.

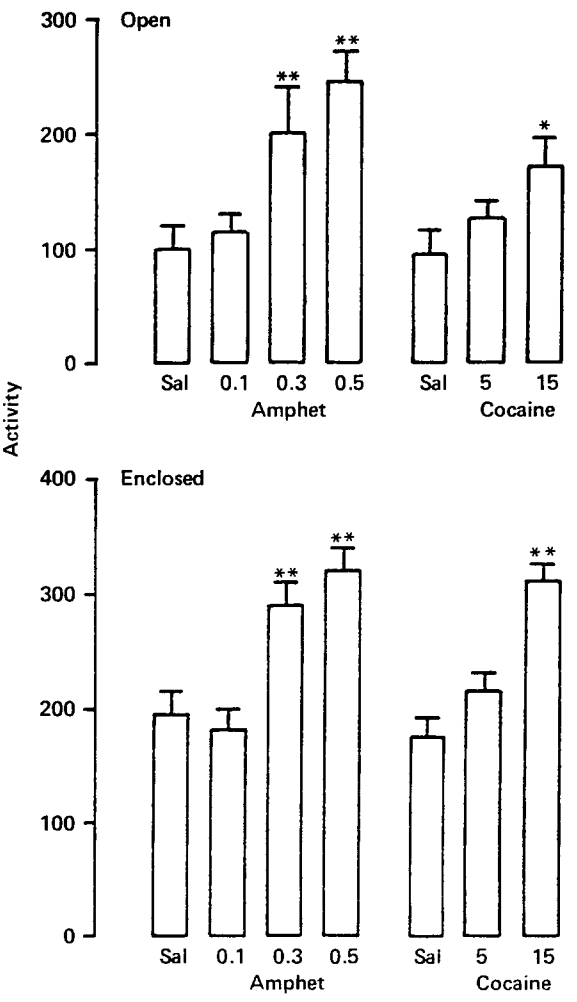

FIG. 3. The rats treated with saline (Sal), d-amphetamine (0.1 to 0.5 mg/kg SC; Amphet) or cocaine (5 or 15 mg/kg SC) 30 minutes prior to the trial (10 min). The results are the means ± SEM of 6 observations. The increases in activity evoked by d-amphetamine, $F(3,40)=18$, $p<0.001$, and cocaine, $F(2,28)=16$, $p<0.001$, were significant, $F(1,20)=45$, $p<0.001$, and unaffected by platform design. Significantly different from saline-treated control (Duncan's test) $*p<0.05$; $**p<0.01$.

to the open platform. However, in the rats treated subchronically with this dose of the drug, the reduction in activity evoked by exposure to the open platform was again significant ($p<0.05$).

Statistical analysis of the results for the chronic study in which rats were repeatedly exposed to the platforms (Fig. 2) showed that nicotine evoked a consistent increase in activity [drug, $F(1,35)=42$, $p<0.001$]. However, the response was again influenced by platform design [drug × platform design, $F(2,35)=5.3$, $p<0.01$] and also by the number of days of treatment [drug × days, $F(12,210)=2.1$, $p<0.05$]. Further analysis indicated that both doses of nicotine tested increased the activity of the rats tested on the open platform [0.1 mg/kg nicotine, $F(1,12)=22$, $p<0.001$; 0.4 mg/kg nicotine, $F(1,12)=42$, $p<0.001$] and that the higher dose also increased the activity of the rats tested on the enclosed platform, $F(1,12)=29$, $p<0.001$. Post hoc analysis (t-test) of the data showed that, for the rats tested on the open platform, the increase in activity evoked by the administration of 0.1 mg/kg nicotine was significant for all sessions (sessions 1, 3 and 25 $p<0.01$; sessions 12 and 15 $p<0.05$) except session 6. The increase in activity evoked by 0.4 mg/kg nicotine was significant ($p<0.01$) for all sessions. For the rats tested on the enclosed platform, the activity was increased significantly by the higher dose of nicotine during sessions 6 ($p<0.05$), 9 ($p<0.01$), 12 ($p<0.05$), 15 ($p<0.01$) and 25 ($p<0.01$). Analysis of the data for the saline-treated rats showed that, when compared with results for the enclosed platform, their activity on the open platform was reduced for all sessions (sessions 1, 3, 12 and 25 $p<0.01$; session 15 $p<0.05$) except sessions 6 and 9. In contrast, the activity of the nicotine-treated rats was not significantly reduced by exposure to the open platform. During sessions 6 and 9 the activity of the saline-treated rats tested on the enclosed platform reached a minimum and was significantly less than that recorded for session 1 ($p<0.05$ for session 6; $p<0.01$ for session 9).

The administration of d-amphetamaine increased, $F(3,40)=18$, $p<0.001$, the activity of the rats on both platforms (Fig. 3). The response to d-amphetamine was not influenced significantly by the platform design, the effects of the two higher doses tested (0.3 and 0.5 mg/kg) being significant (Duncan's test, $p<0.01$) on both platforms. Intraperitoneal injections of cocaine also increased the activity of rats tested on both platforms, $F(2,28)=16$, $p<0.001$. Again the response was not influenced by platform design, the

increase being statistically significant (Duncan's test; $p<0.05$ for the open platform; $p<0.01$ for the enclosed platform) for the higher dose tested (15 mg/kg).

DISCUSSION

The stimulant effects of nicotine on spontaneous locomotor activity are well established and there is clear evidence that the response to higher doses of the drug is enhanced if the drug is given chronically (6, 13, 17, 18). The data presented in this paper are entirely consistent with these earlier studies since they have shown that subchronic nicotine, when given at a dose of 0.4 mg/kg, evoked an increase in activity which was independent of the test environment used to make the measurements. In addition, however, the study has also shown that, when the drug is given acutely or subchronically at low doses, nicotine appears to be a more potent stimulant in rats in which the spontaneous activity has been suppressed by the nature of the test environment used.

Studies with the elevated X-maze test for anxiety suggest that open spaces are significantly more aversive to rats than those which are enclosed (15). It seems reasonable to suggest, therefore, that the lower levels of activity observed for the rats tested on the open platform were caused, at least in part, by the aversive properties of the environment. Nelsen (14) has already reported that nicotine can ameliorate the behavioural disruption observed in rats exposed to an unavoidable stress and there is also evidence that nicotine self-administration is enhanced in animals exposed to stressful stimuli (8). Thus, the increase in activity evoked by the administration of low doses of nicotine to the rats tested on the open platform could be related to its ability to attenuate responses to stress.

Analysis of the results of the experiments with d-amphetamine and cocaine indicated that, over the dose range used, the stimulant properties of these drugs were unaffected by the platform design. Their properties were, therefore, similar to those of chronic nicotine when it was given at a dose of 0.4 mg/kg. Thus, the selective effects of the lower doses of nicotine on the activity of rats tested on the open platform do not appear to be a property which is common to all stimulant drugs. The psychostimulant responses to d-amphetamine and cocaine are thought to be mediated by enhanced neurotransmission at DA synapses within the brain, particularly in the mesolimbic system (19). There is also evidence that nicotine, at doses in excess of approximately 0.2 mg/kg, also stimulates DA secretion in the mesolimbic system and that the psychostimulant response to chronic nicotine is mediated by this pathway (5,9). The results of preliminary studies in this laboratory suggest that, in contrast, the effects of low doses of nicotine on the spontaneous activity of rats tested on the open platform may not be dependent upon mesolimbic DA secretion (Vale and Balfour, unpublished).

The results of the study, therefore, suggest that the locomotor stimulant properties of nicotine may be biphasic, the response to acute nicotine or the chronic administration of low doses of the drug being primarily the result of attenuation of reductions in activity evoked by factors such as aversive environmental stimuli. In contrast, the chronic administration of moderate or high doses of the drug appears to evoke increases in activity which are independent of the test environment used. The data are consistent with the hypothesis that the effects of nicotine on rat behaviour on the open platform may be associated with its reported ability to ameliorate the effects of stress (1,14). However, Bovet *et al.* (4) reported that acute nicotine also attenuates the reduction in the activity of rats tested in a running wheel during the daytime when their activity was at a minimum, whereas when given to rats tested at night, when they are more active, it either had no effect or, at higher doses (1 mg/kg), suppressed activity. Thus, the results of the present study do not preclude the possibility that the changes observed for the rats tested on the open platform are not a specific response to a stress-induced fall in activity but a measure of the ability of the drug to attenuate reductions in activity evoked by a number of different physiological or environmental stimuli.

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