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Regional variation in the effects of nicotine on catecholamine overflow in rat brain

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Abstract

The effects of acute, repeated intermittent and continuous administration of nicotine on the overflow of noradrenaline in the ventral hippocampus and dopamine in the nucleus accumbens and striatum have been studied. Daily injections of nicotine (0.4 mg/kg^{-1} for 5 days) enhanced noradrenaline and dopamine overflow in the ventral hippocampus and nucleus accumbens respectively ($P < 0.01$ and $P < 0.05$) but not dopamine in the striatum in response to a nicotine challenge. The responses in the ventral hippocampus and nucleus accumbens were attenuated ($P < 0.01$) by the constant infusion of nicotine at a dose of 1 mg kg^{-1} per day; the dopamine response in the striatum required a higher dose (4 mg kg^{-1} per day) before desensitisation was observed. The data suggest that the dopamine projections to the striatum are less sensitive to both stimulation and desensitisation by nicotine than the catecholamine projections to the ventral hippocampus and nucleus accumbens. © 1997 Elsevier Science B.V.

Keywords: Nicotine; Hippocampus, ventral; Nucleus accumbens; Striatum; Noradrenaline; Dopamine

1. Introduction

In common with amphetamine and cocaine, nicotine stimulates mesolimbic dopamine secretion (Di Chiara and Imperato, 1988; Benwell and Balfour, 1992). This effect is thought to mediate both the locomotor stimulant properties of nicotine (Clarke, 1990) and the rewarding properties of the drug which reinforce itself-administration (Singer et al., 1982; Corrigall and Coen, 1991; Corrigall et al., 1992, 1994). Repeated pretreatment with daily injections nicotine is associated with an enhanced locomotor stimulant response to the drug and sensitisation of nicotine-induced dopamine overflow in the nucleus accumbens (Benwell and Balfour, 1992). A similar pretreatment protocol also results in sensitisation of the stimulatory effects of nicotine on noradrenaline overflow in the ventral hippocampus (Mitchell, 1993). However, prolonged or repeated exposure to nicotine causes up-regulation of the central nicotinic receptors which can be labelled with [^{3}H]nicotine in smokers (Benwell et al., 1988) or in animals (Marks et al., 1983; Marks and Collins, 1985; Schwartz and Kellar, 1985). This response is thought to be related to prolonged or repeated periods of receptor desensitisation (Wonnacott, 1990) and implies that the receptors, which have high affinity for nicotine, probably desensitise readily. This conclusion is supported by the results of recent studies in our laboratory (Benwell et al., 1995) which suggest that the receptors, which mediate the enhanced stimulant effects of nicotine on dopamine overflow in the nucleus accumbens and locomotor activity, are desensitised when the plasma concentration is maintained at approximately 25 ng/ml, a level which is commonly found in the plasma of habitual smokers (Russell, 1990; Benowitz et al., 1990).

Nicotine also stimulates nigrostriatal dopamine although the evidence available suggests that this system is less sensitive to nicotine than the mesolimbic (Imperato et al., 1986; Grenhoff and Svensson, 1988; Mereu et al., 1987; Clarke et al., 1988). The influence of chronic nicotine on nigrostriatal dopamine neurones has not been studied. The present study has used in vivo microdialysis to compare systematically the effects of acute, repeated intermittent and continuous exposure to nicotine on dopamine secretion in the nucleus accumbens with the dopamine responses in the dorsolateral striatum and the noradrenaline responses observed in the ventral hippocampus. It has been suggested that the different magnitude of the responses previously

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Table 1
Experimental protocol

Infusion (days 1–14)	Intermittent injections (once/day on days 9–13)	Drug challenge on day 14
Saline	Saline	Saline + nicotine
Saline	Nicotine (0.4 mg/kg s.c.)	Saline + nicotine
Nicotine (1 mg/kg per day)	Saline	Saline + nicotine
Nicotine (1 mg/kg per day)	Nicotine (0.4 mg/kg s.c.)	Saline + nicotine
Nicotine (4 mg/kg per day)	Saline	Saline + nicotine
Nicotine (4 mg/kg per day)	Nicotine (0.4 mg/kg s.c.)	Saline + nicotine

On day 14, each animal was injected with saline (1 ml/kg s.c.) followed 60 min later by a challenge dose of nicotine (0.4 mg/kg s.c.).

observed in the striatal dopamine system may be due to a higher density of dopamine transporters in this region (Abercrombie et al., 1989). Therefore, in the present study, the selective catecholamine uptake inhibitor, nomifensine, has been included in the Ringer solution perfusing the probe to reduce the contribution any differences in the transporters may make to any apparent differences in sensitivity. Under these conditions the regional differences in the effects of nicotine on overflow are likely to predominantly reflect its influence on release rather than on reuptake.

2. Materials and methods

2.1. Subjects and pretreatments

Male Sprague-Dawley rats, bred in the Biomedical Services Unit at Ninewells hospital, Dundee, from stock originally purchased from Interfauna, were used throughout. All animals had free access to food and water, weighed 250–350 g at the start of the experiments and were housed in pairs prior to and singly following surgery. In the experiments designed to investigate the effect of nicotine dose on nucleus accumbens and striatum dopamine or the influence of tetrodotoxin on dopamine secretion in the striatum, the rats all received 5 daily injections of saline (1.0 ml/kg s.c.) prior to implantation of the dialysis probes. In the experiments designed to investigate the effects of nicotine infusions on responses to nicotine, the rats were assigned at random to one of the six treatment groups outlined in Table 1. Osmotic minipumps (Alzet 2ML2), containing sterile saline or nicotine hydrogen tartrate solutions, were implanted subcutaneously (s.c.) under halothane (3% in oxygen) anaesthesia. Nicotine was infused at a dose of 1 mg/kg per day (one third of the animals in each group) or 4 mg/kg per day (one third of the animals in each group) for 14 days. The dose is expressed as the free base. The remaining animals in each group were infused with saline. On days 9–13 of the experiment, the animals also received daily subcutaneous injections of saline (half the animals in each group) or nicotine (0.4 mg/kg).

2.2. Implantation of microdialysis probes

The procedures for the preparation and implantation of the dialysis probes were essentially those reported previously (Benwell and Balfour, 1992). At least 3 h after the injection on day 13, the rats were anaesthetised with halothane and dialysis probes were implanted, stereotaxically, in the core of the nucleus accumbens (dialysis probe length = 1.5 mm; coordinates: ±1.7 mm rostral, +1.5 mm lateral with respect to bregma and 7.5 mm vertically from the dura), striatum (dialysis probe length = 3 mm; coordinates: ±1.0 mm rostral, −2.5 mm lateral with respect to bregma and 5.5 mm vertically from the dura) or the ventral hippocampus (dialysis probe length = 3 mm; coordinates: −5.8 mm rostral, +4.9 mm lateral with respect to bregma and 7.0 mm vertically from the dura) according to Paxinos and Watson (1986). In the studies on dopamine overflow in the nucleus accumbens and striatum,

Table 2
Regional extracellular levels of monoamines

Infusion (days 1–14)	Intermittent injections (days 9–13)	Dopamine (pmol/20 μl)		Noradrenaline (pmol/20 μl)
		Nucleus accumbens	Striatum	Ventral hippocampus
Saline	Saline	0.526 + 0.122	0.669 + 0.124	0.119 + 0.017
Saline	Nicotine (0.4 mg/kg s.c.)	0.469 + 0.049	0.868 + 0.225	0.114 + 0.010
Nicotine (1 mg/kg per day)	Saline	0.600 + 0.097	0.712 + 0.203	0.136 + 0.049
Nicotine (1 mg/kg per day)	Nicotine (0.4 mg/kg s.c.)	0.511 + 0.062	0.822 + 0.177	0.177 + 0.099
Nicotine (4 mg/kg per day)	Saline	–	0.734 + 0.160	–
Nicotine (4 mg/kg per day)	Nicotine (0.4 mg/kg s.c.)	–	0.762 + 0.232	–

The results are the means + S.E.M. of the basal extracellular levels measured at the start of the experimental day with nomifensine (5 mM) present in the Ringer.

two probes were implanted in the contralateral nucleus accumbens and striatum. The experiments on noradrenaline overflow in the ventral hippocampus were performed using single probes located in the ventral hippocampus. The position of the probes was routinely determined histologically from sections prepared at postmortem.

2.3. Microdialysis

On the day following surgery, the animals were placed into a dialysis chamber (40 cm square × 25 cm high) (Vale and Balfour, 1989). At this time, the dialysis probe was connected to a syringe pump containing Ringer solution (147.0 mM NaCl; 4.0 mM KCl; 1.25 mM $CaCl_2$) containing nomifensine (5 μM) which was perfused at a constant rate of 1.7 μl/min. In these experiments, following a period (60 min) of equilibration three baseline samples of dialysate were collected and analysed to determine the baseline concentrations of dopamine. All animals were then injected with a control injection s.c. saline followed, 60 min later, by a challenge dose of nicotine (0.4 mg/kg s.c.). Dialysate samples (20 min) were collected throughout and for at least 100 min after the injection of nicotine. The concentrations of dopamine and DOPAC in the dialysates were assayed by injecting the samples directly onto a high performance liquid chromatography system with a Coulochem electrochemical detector as described previously (Benwell and Balfour, 1992).

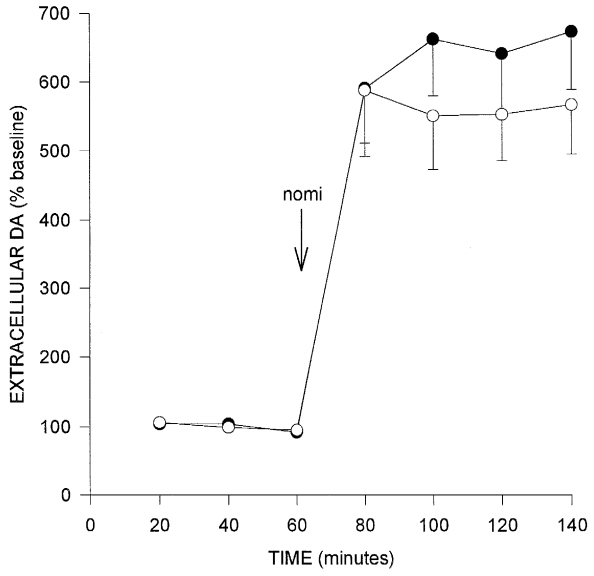

Fig. 1. Effect of nomifensine on nucleus accumbens and striatal dopamine (DA) overflow. At the time shown by the arrow, the Ringer solution, perfusing dialysis probes implanted in the nucleus accumbens (●) and the striatum (○), was changed to Ringer containing nomifensine (nomi, 5 μM). The results are the means ± S.E.M. of 12 observations at each point, expressed as percentages of the mean values obtained prior to the switch to nomifensine-containing Ringer. Basal extracellular levels were 0.102 + 0.014 and 0.166 + 0.016 pmol/20 μl in the nucleus accumbens and striatum respectively prior to switching to the nomifensine-containing Ringer.

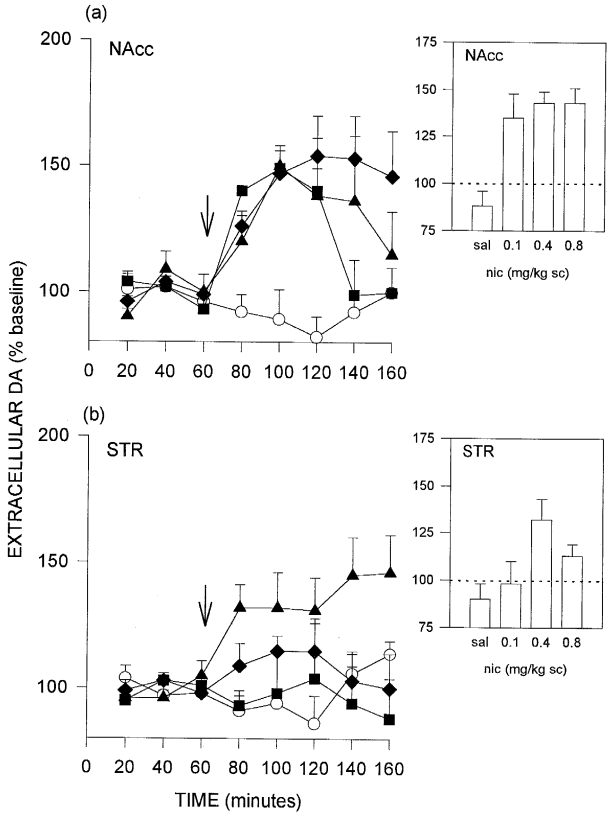

Fig. 2. The influence of acute nicotine injections on nucleus accumbens (NAcc) and striatal (STR) dopamine (DA) overflow. All animals received daily injections of saline for 5 days prior to implantation of dialysis probes in the nucleus accumbens and striatum. On day 6, the animals were challenged with subcutaneous injections of saline (○, $n = 4$) or nicotine (■, 0.1, $n = 6$; ▲, 0.4, $n = 5$; ◆, 0.8, $n = 4$ mg/kg) at the time shown by the arrow. The results are the means ± S.E.M. of 5–6 observations at each point expressed as percentages of the mean values obtained prior to the saline or nicotine injections. The results in the insets show the average dopamine overflow/20 min (means ± S.E.M.) measured over a period of 80 min after the injections of saline (sal) or nicotine (nic), expressed as percentages of the mean values obtained prior to the saline or nicotine injections. Basal extracellular levels, obtained with nomifensine-containing Ringer, were 0.697 + 0.090 and 0.980 + 0.123 pmol/20 μl in the nucleus accumbens and striatum respectively prior to the injection of saline or nicotine.

In the experiments designed to investigate the effect of nomifensine on extracellular levels of transmitter, drug-naive animals were connected to a syringe pump containing the basic Ringer solution to which nomifensine had not been added and left to equilibrate for 60 min before samples of dialysate were removed at 20 min intervals until a stable baseline of transmitter had been obtained. At this time, the perfusing solution was switched to Ringer solution containing nomifensine (5 μM) and at least another five samples taken.

In the experiments designed to investigate the influence of tetrodotoxin on extracellular levels of dopamine in the striatum, a separate group of saline-pre-injected animals were connected to a syringe pump containing the basic Ringer solution with nomifensine (5 μM) and were left to equilibrate for 60 min before samples of dialysate were

removed at 20 min intervals until a stable baseline of transmitter had been obtained. At this time, the perfusing solution was switched to Ringer solution containing tetrodotoxin (1 μM) and at least another five samples taken before the rats were challenged, systemically, with nicotine (0.4 mg/kg s.c.).

2.4. Statistical analysis

The data were analysed using the PC version of the Statistics Package for Social Scientists (SPSS). The microdialysis data were analysed using an analysis of variance for repeated measures with infusion, intermittent pretreatment and the challenge injection as independent factors. Where appropriate, post hoc analysis was performed using Duncan's test.

2.5. Drugs

Nicotine hydrogen tartrate and tetrodotoxin were purchased from Sigma. Nomifensine hydrogen maleate was a gift from Hoechst.

3. Results

3.1. The effect of nomifensine on extracellular monoamine levels

The addition of nomifensine (5 μM) to the Ringer perfusing the probe was associated with a significant increase in the extracellular levels of dopamine in both the nucleus accumbens ($F(6,90) = 39.1$, $P < 0.001$) and striatum ($F(6,90) = 30.7$, $P < 0.001$) as shown in Fig. 1. Statistically, the addition of nomifensine, at the dose tested, caused an equivalent increase in dopamine overflow in the nucleus accumbens and striatum. In agreement with Mitchell (1993), extracellular noradrenaline levels in the ventral hippocampus were beyond the limit of detection and could only be revealed by the addition of nomifensine to the Ringer solution used to perfuse the probe.

3.2. The effect of nicotine dose on nucleus accumbens and striatum dopamine

When the presynaptic dopamine transporter was inhibited by the inclusion of nomifensine in the Ringer solution, acute s.c. injections of nicotine caused a significant ($F(3,21) = 6.8$, $P < 0.01$) increase in the levels of dopamine measured extracellularly in the nucleus accumbens at all the doses tested (Fig. 2a). When the overflow was measured in the 60 min following the nicotine injection, the data suggest that nicotine evoked a near maximal response at the lowest dose (0.1 mg/kg) tested (Fig. 2a inset). However, beyond this, there was evidence for a dose-dependent response, data which suggest that the duration of the response to nicotine rather than the peak response is dose-dependent (Fig. 2a). In contrast, the dopamine response to nicotine in the striatum seemed to be bell-shaped with only the 0.4 mg/kg dose producing a significant increase in dopamine overflow in this area of

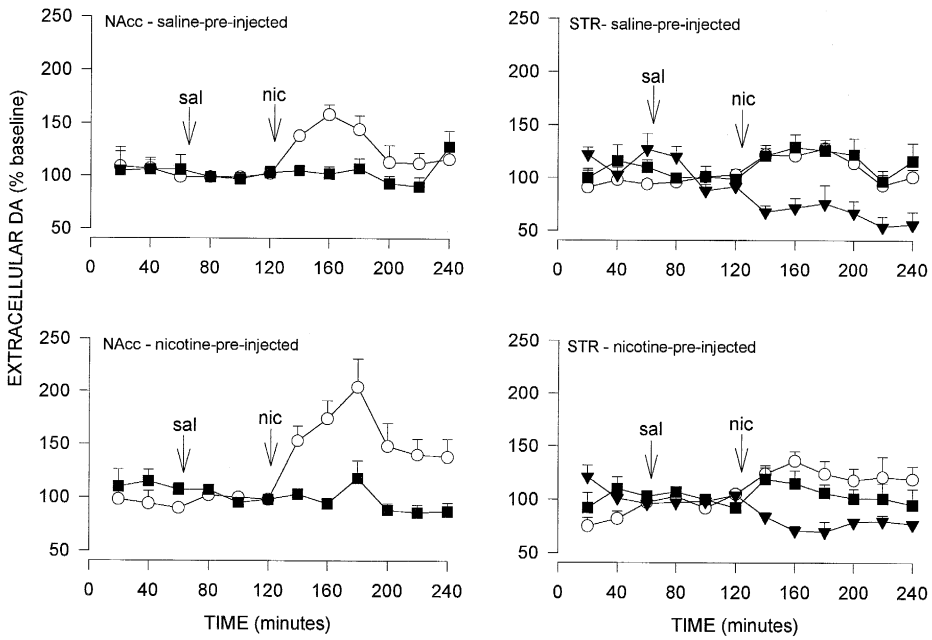

Fig. 3. The influence of the constant infusion of nicotine on nucleus accumbens (NAcc) and striatal (STR) dopamine (DA) overflow. The rats were infused with saline (sal, ○) or nicotine (nic, ■, 1 mg/kg per day; ▲, 4 mg/kg per day) for 14 days. On days 9–13, half of each of the three groups of animals also received once daily injections of nicotine (0.4 mg/kg s.c. – nicotine-pre-injected) while the remainder received saline (saline-pre-injected). On day 14, the rats were challenged with injections of saline (at the time indicated by the first arrow) followed 60 min later with a challenge dose of nicotine (0.4 mg/kg s.c. at the time indicated by the second arrow). The results are the means ± S.E.M. of 4–6 observations at each point expressed as percentages of the mean values obtained prior to the nicotine injections.

the brain (Fig. 2b and inset). Analysis of variance of the data obtained during the 60 min following the administration of the drug revealed that the increase in dopamine overflow in the nucleus accumbens was significantly higher ($F(1,21) = 6.5,\ P < 0.05$) than the response in the striatum.

3.3. Influence of nicotine pretreatment on dopamine overflow in the nucleus accumbens and striatum

The influence of nicotine pretreatment on dopamine overflow in the nucleus accumbens and striatum is shown diagrammatically in Fig. 3. With nomifensine present in the Ringer solution, there were no significant differences in the basal extracellular dopamine levels which could be attributed to the pretreatments (Table 2). Statistical analysis of the data revealed significant inter-regional differences in the overflow of dopamine in the nucleus accumbens and striatum in response to a challenge dose of nicotine in the different pretreatment groups. The responses to both the nicotine infusions ($F_{time}(8.272) = 3.05$, $P < 0.01$) and the effects of daily injections of nicotine ($F_{time}(8,272) = 2.15,\ P < 0.05$) on the responses to a subsequent injection of nicotine were significantly greater ($F_{time}(8,272) = 3.05;\ P < 0.01$ and $F_{time}(8,272) = 2.15;\ P < 0.05$ respectively) in the nucleus accumbens than the striatum. In the nucleus accumbens, the extracellular dopamine responses to nicotine were significantly ($F(8,72) = 2.08,\ P < 0.05$) enhanced in saline-infused rats which had been pre-injected with daily injections of nicotine. A significant ($F(8,64) = 4.05,\ P < 0.01$) increase was also seen in the dopamine overflow in the striatum of saline-infused rats. However, this dopamine response to nicotine was not enhanced by daily pre-injections of nicotine. The nucleus accumbens dopamine responses to nicotine were significantly ($F(8,160) = 7.41,\ P < 0.01$) attenuated during the constant infusion of the alkaloid at a rate of 1.0 mg/kg per day. The increase in striatum dopamine was unaffected by the infusion of 1 mg/kg per day but was abolished ($F(8,160) = 9.60,\ P < 0.01$) during the infusion of nicotine at the higher rate of 4.0 mg/kg per day.

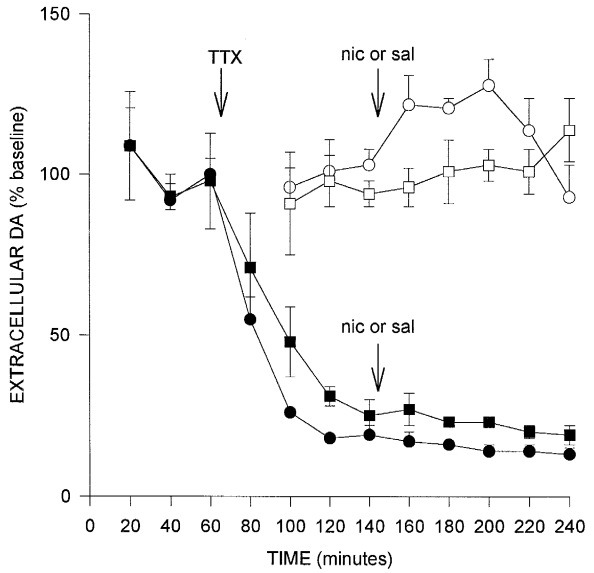

Fig. 4. The influence of tetrodotoxin (TTX) on striatal dopamine (DA) responses to nicotine. The rats received a daily pretreatment of nicotine (0.4 mg/kg s.c.) once/day for 5 days. On day 6, the striatum was dialysed by perfusing with Ringer containing nomifensine (5 μM) and at the time indicated by the first arrow, the Ringer solution was switched to one containing tetrodotoxin (1 μM, filled symbols; $n = 4$ per group). At the time indicated by the second arrow, the animals were challenged with an injection of nicotine (nic, ●, 0.4 mg/kg s.c.) or saline (sal, ■). Another group of animals were dialysed with Ringer containing nomifensine (open symbols, $n = 4$ per group) but no tetrodotoxin and challenged with an injection of nicotine (○, 0.4 mg/kg s.c.) or saline (□) at the time indicated by the second arrow. The results are the means ± S.E.M., expressed as percentages of the mean values obtained prior to the tetrodotoxin administration (filled symbols) or prior to nicotine in the Ringer plus nomifensine group (open symbols).

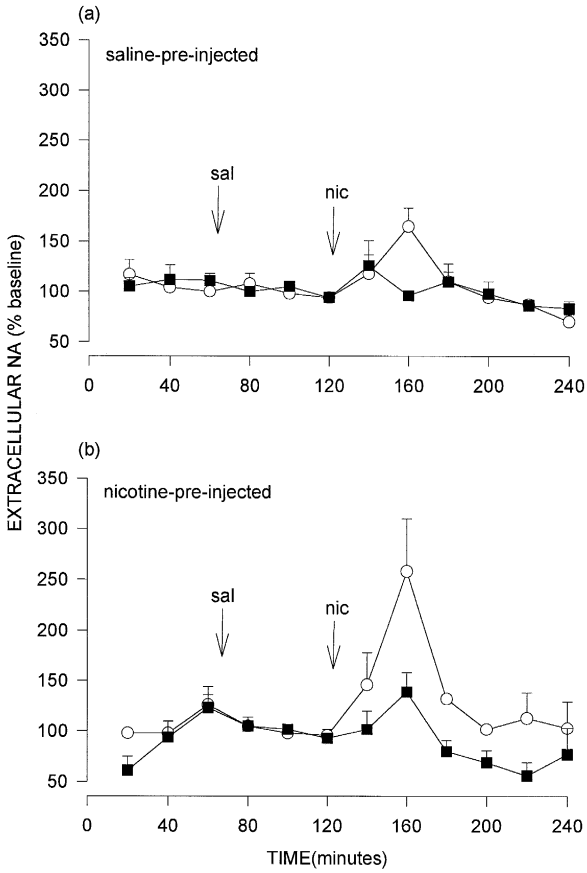

Fig. 5. The influence of the constant infusion of nicotine on ventral hippocampal noradrenaline (NA) overflow. The rats were infused with saline (sal, ○) or nicotine (nic, ■, 1 mg/kg per day) for 14 days. On days 9–13, half of each group of animals also received once daily injections of nicotine (0.4 mg/kg s.c. – nicotine-pre-injected) while the remainder received saline (saline-pre-injected). On day 14, the rats were challenged with injections of saline (at the time indicated by the first arrow) followed 60 min later with a challenge dose of nicotine (0.4 mg/kg s.c. at the time indicated by the second arrow). The results are the means ± S.E.M. of four observations at each point expressed as percentages of the mean values obtained prior to the nicotine injections.

3.4. The influence of tetrodotoxin on nicotine-induced striatum dopamine overflow

The infusion of tetrodotoxin (1 μM), via the dialysis probe, resulted in a very significant ($F(6,42) = 34.3$, $P < 0.001$) reduction in the overflow of dopamine measured in the striatum (Fig. 4). The stimulant effect of systemically administered nicotine (0.4 mg/kg), on striatum dopamine overflow, was abolished when tetrodotoxin was present in the probe.

3.5. Influence of constant nicotine infusion on ventral hippocampus noradrenaline responses

There were no significant changes in the basal noradrenaline overflow in the ventral hippocampus which could be attributed to the pretreatments (Table 2). Analysis of the data revealed that nicotine significantly ($F(8,24) = 7.0$, $P < 0.001$) increased noradrenaline overflow in the ventral hippocampus of saline-pretreated rats (Fig. 5a). This response was significantly ($F(8,104) = 2.71$, $P < 0.01$) enhanced following nicotine pretreatment (Fig. 5b). The responses to both an acute and subchronic challenge with nicotine were significantly ($F(8,104) = 5.1$, $P < 0.01$) attenuated in both groups of rats if the animals were also constantly infused with the drug at a rate of 1.0 mg/kg per day.

4. Discussion

Electrophysiological and biochemical studies suggest that mesolimbic dopamine neurones respond to lower doses of nicotine than those required to stimulate the nigrostriatal dopamine pathway (Clarke et al., 1988; Di Chiara and Imperato, 1988; Imperato et al., 1986; Mereu et al., 1987). This phenomenon can be more difficult to demonstrate using the microdialysis technique because there is a higher density of dopamine transporters in the striatum when compared with the nucleus accumbens (Marshall et al., 1990; Garris and Wightman, 1994). As a result, stimulation of the projections to the striatum elicits a smaller increase in dopamine overflow into the extracellular space sampled by a dialysis probe when compared with the increase evoked by stimulation of the projections to the nucleus accumbens (Garris and Wightman, 1994). In order to reduce the contribution of this regional difference in the efficiency of the dopamine transporter system, the experiments reported in this paper were performed using dialysis probes perfused with a solution containing the catecholamine uptake inhibitor, nomifensine, to inhibit the transporter system. Under these conditions, dopamine responses to acute nicotine could be measured in both the striatum and nucleus accumbens.

Previous studies in our laboratory using probes perfused with a nomifensine-free Ringer solution revealed a modest response to acute nicotine in the nucleus accumbens when it is given at a dose of 0.4 mg/kg (Benwell and Balfour, 1992) and no response to this dose of the drug in the striatum (Birrell and Balfour, 1995). The data are in complete agreement with the results obtained by Brazell et al. (1990) but contrast to the reports of Imperato et al. (1986) and Di Chiara and Imperato (1988) who recorded relatively robust increases in extracellular dopamine in both the nucleus accumbens and striatum following a slightly higher dose (0.6 mg/kg s.c.) of nicotine. One explanation for the differences may be related to the calcium concentrations of the Ringer solutions, higher calcium concentrations increasing the overflow of dopamine (Westerink et al., 1988). The present study and that of Brazell et al. (1990) used a concentration of calcium (1.25 mM) which is a close approximation to the endogenous extracellular concentration of calcium in the brain (Somjet et al., 1987) whereas the calcium levels used by Imperato et al. (1986) were almost 3-fold higher (3.4 mM). Therefore, it is possible that the effects of nicotine on dopamine overflow are amplified to more detectable levels at higher calcium concentrations. The results measured in the presence of the uptake inhibitor are consistent with previous results in two respects. Firstly, in agreement with a number of previous reports (Imperato et al., 1986; Grenhoff and Svensson, 1988; Mereu et al., 1987; Clarke et al., 1988), the mesoaccumbens dopamine neurones were found to be more sensitive to the stimulatory effects of nicotine than those which innervate the striatum. Secondly, the sensitised effects of nicotine on dopamine overflow in the nucleus accumbens, reported previously in the absence of nomifensine for animals which had been pretreated with the drug prior to the test day (Benwell and Balfour, 1992), were still apparent in the presence of nomifensine. No sensitisation was observed in the striatum in spite of the fact that the animals were pretreated with a dose of nicotine (0.4 mg/kg) which was clearly able to stimulate the nigrostriatal neurones. These data are consistent with the hypothesis that sensitisation may be an intrinsic property of mesoaccumbens dopamine neurones which is not shared by the dopamine-secreting neurones which innervate the striatum.

The studies with the rats which received infusions of nicotine also revealed differences in the sensitivity of the two dopamine systems to nicotine. The infusion of nicotine, at a dose of 1 mg/kg per day, abolished or markedly inhibited the stimulatory effects of a challenge injection of nicotine on dopamine overflow in the nucleus accumbens. These data both confirm the results of earlier experiments obtained with nicotine-sensitised rats in the absence of nomifensine (Benwell et al., 1995) and imply the acute dopamine response to nicotine in this area of the brain is also desensitised by nicotine when it is constantly infused at this dose. In contrast, desensitisation of the dopamine response to nicotine in the striatum was unaffected by the lower dose (1 mg/kg per day) used for the infusion

studies whereas desensitisation of the response was observed in this area of the brain when the higher dose (4 mg/kg per day) was infused. These results suggest that nigrostriatal neurones are less sensitive to both the stimulatory effects of nicotine when the drug is given in bolus form and to desensitisation when the drug is infused. The reasons for these regional differences remain unclear.

It is known that nicotine exerts its effects in the brain acting on a family of neuronal nicotinic receptors (Wonnacott, 1990) and that nicotinic cholinoceptors are present on both the cell soma and terminal membranes of the mesolimbic and nigrostriatal dopamine systems (Clarke and Pert, 1985). However, when administered systemically at the doses used in this study, the effects of nicotine on mesolimbic dopamine secretion are impulse-dependent and almost entirely mediated by stimulation of the nicotinic receptors present on dopamine cell bodies on the ventral tegmental area rather than those located on dopamine terminals (Benwell et al., 1993; Nisell et al., 1994). The effect of nicotine on dopamine secretion from striatum nerve terminals is not sensitive to tetrodotoxin (Giorgieff-Chesselet et al., 1979; Rapier et al., 1988). However, the nicotine-induced striatal dopamine release, seen in the present study, appeared to be entirely impulse-dependent, since it was abolished by tetrodotoxin. Therefore, it is unlikely that the differences in sensitivity to nicotine between the mesolimbic and nigrostriatal dopamine systems reflect a greater influence of the nicotinic receptors located on dopamine nerve terminals in the striatum. It is possible that the nicotinic receptors, expressed by the dopamine neurones in the substantia nigra and ventral tegmental area, are different subtypes with different affinities for nicotine. However, there is no evidence for marked regional variation in the receptor subunits (Wada et al., 1989; Wonnacott, 1990). Therefore, the differential regional responses to the drug may reflect differences in the intrinsic properties of the dopamine neurones rather than the nicotinic receptors present in the mesolimbic and nigrostriatal systems. Previous studies (Benwell et al., 1995) have shown that when nicotine is constantly infused at a dose of 1 mg/kg per day it maintains plasma nicotine at a concentration of approximately 25 ng/ml, a concentration similar to that found in the plasma of many smokers (Benowitz et al., 1990). When nicotine is constantly infused at the higher dose tested, plasma nicotine is maintained at a concentration close to 90 ng/ml, a plasma nicotine concentration which is only likely to be found in people who smoke very heavily. Thus, it seems reasonable to suggest that, in many smokers, nigrostriatal dopamine neurones are unlikely to be desensitised by the concentrations of nicotine routinely achieved in their plasma.

In order to be able to compare the effects of nicotine on forebrain dopamine systems with the response of another catecholamine system known to respond to nicotine, the effects of nicotine pretreatment on noradrenaline overflow in the ventral hippocampus were also studied. In agreement with previous studies (Mitchell, 1993), nicotine injections were found to increase noradrenaline overflow in the hippocampus. The experiments also confirmed the results reported by Mitchell (1993) which showed that pretreatment with nicotine results in sensitisation of its effects on noradrenaline overflow in the hippocampus. These experiments extend these observations by showing that constant infusions of nicotine (1 mg/kg per day) suppress both the acute noradrenaline response to nicotine and the sensitised response observed in nicotine-pretreated animals. These data suggest that the noradrenaline responses to nicotine in the hippocampus are similar in many respects to the dopamine responses to the drug seen in the nucleus accumbens. In particular, the dose of nicotine required to elicit the responses are the same. Thus, the noradrenaline response in the hippocampus and the dopamine responses in the nucleus accumbens may be mediated by the same isoform of the receptor although, clearly, this conclusion requires confirmation. The putative role of hippocampal noradrenaline in nicotine psychopharmacology has not been established. However, the data presented in this paper suggest that the responses may well be blunted in many smokers who maintain their plasma nicotine levels at 25 ng/ml or higher and that when considering the role of the possible system in the tobacco smoking habit, this factor should be considered.

In summary, these experiments have confirmed the differences in the sensitivity of the mesoaccumbens and nigrostriatal dopamine systems to nicotine reported previously and have shown that the systems also differ in their responses to nicotine pretreatment. The data are consistent with the possibility that there may be significant intrinsic differences in the way the two groups of neurones respond to chronic nicotine. However, the possibility that these differences may reflect heterogeneity in the isoforms of the nicotinic receptors which are expressed by the neurones in the ventral tegmental area and substantia nigra cannot be excluded.

Acknowledgements

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