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Differential effects of response-contingent and response-independent nicotine in rats

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Abstract

Passive administration of nicotine activates the hypothalamic–pituitary–adrenocortical axis and sympathetic nervous system. However, little is known about the effects of self-administered nicotine. Drug-naive rats were trained to respond for food reinforcement and then tested in one, 1-h session in which they received response-contingent i.v. nicotine or response-independent i.v. nicotine or saline. Blood draws were taken immediately prior to the session, 15 min after the first infusion and immediately after the session. Both response-contingent and response-independent nicotine (RI/N) increased corticosterone within 15 min, however, corticosterone levels returned to baseline in animals receiving response-contingent nicotine (RC/N) by the end of the session while remaining elevated in those receiving RI/N. Furthermore, only RI/N increased plasma epinephrine and norepinephrine levels; RC/N produced no effect. These differences indicate that nicotine's acute effects are powerfully modified by the presence of a contingency relationship between drug administration and the animal's behavior and that this relationship develops very rapidly.

Keywords: Self-administration; Catecholamine; Epinephrine; Norepinephrine; Corticosterone; Yoking; Nicotine

1. Introduction

There is a substantial body of research on nicotine's physiological and behavioral effects and their relation to smoking behavior. Two such effects are nicotine's capacity to activate the hypothalamic–pituitary–adrenocortical axis (Andersson et al., 1983; Matta et al., 1987) and sympathetic nervous system (Kiritsy-Roy et al., 1990; Van Loon et al., 1987).

Nicotine activates the hypothalamic–pituitary–adrenocortical axis and elevates glucocorticoids in humans (Stalke et al., 1994) and laboratory animals (Cam and Bassett, 1983, 1984). With repeated dosing, chronic tolerance develops to nicotine's corticosterone-elevating effects in animals, although there is some question regarding the rate of its development and the extent of the tolerance (Benwell and Balfour, 1979; Cam and Bassett, 1984; Caggiula et al., 1991). In contrast, smokers may never show complete tolerance (Fuxe et al., 1989). This elevation of glucocorticoids has received considerable attention for its potential role in regulating nicotine-seeking behavior. Several researchers have shown that in animals, the glucocorticoid corticosterone decreases sensitivity to nicotine (Pauly et al., 1990; Caggiula et al., 1993, 1994) and some investigators have speculated that glucocorticoids may affect the pattern of nicotine use by mediating tolerance to some of nicotine's effects (Pauly et al., 1988, 1992; Caggiula et al., 1993, 1994). Nicotine-induced hypersecretion of glucocorticoids may also have immunosuppressive consequences (Munck et al., 1984; Fuxe et al., 1989; Caggiula et al., 1992; Geng et al., 1996; McAllister-Sistilli et al., 1998), as well as contribute to arteriosclerosis, osteoporosis, hyperglycemia, and hyperlipidemia (Fuxe et al., 1989). Understanding nicotine's effects on the hypothalamic–

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pituitary–adrenocortical axis may therefore be of great importance in determining the health effects of chronic nicotine intake.

Nicotine also activates the sympathetic nervous system in humans (Westfall and Watts, 1963; Volle and Koelle, 1975; Cryer et al., 1976) and laboratory animals (Grunberg et al., 1988; Morse, 1989; Kiritsy-Roy et al., 1990). Nicotine-induced activation of the sympathetic nervous system contributes to the acute cardiovascular effects of nicotine (Cryer et al., 1976; Robertson et al., 1988), such as increased blood pressure and heart rate, and may play a role in the increased risk for cardiovascular disease associated with smoking (Volle and Koelle, 1975; Benowitz, 1986; Lakier, 1992).

Although both the hypothalamic–pituitary–adrenocortical axis and sympathetic nervous system have been proposed as mediating either some of the health effects of nicotine (Fuxe et al., 1989; Volle and Koelle, 1975) or contributing to the development or maintenance of smoking behavior (Pomerleau and Pomerleau, 1991; Pauly et al., 1992), the support for these hypotheses from animal research is based on experimenter-administered drug. Evidence from research on other drugs, however, indicates that the effects obtained when a drug is administered by the experimenter, independent of the animal's behavior, may differ from those produced when the drug is self-administered, even when controlling for dose and timing of infusions. For example, Dworkin et al. (1995) found that rats receiving response-independent infusions of cocaine had increased mortality compared to rats self-administering exactly the same amount of drug. Other researchers have also reported differences between response-contingent and response-independent administration of drugs (Smith et al., 1982; Moolton and Kornetsky, 1990; Kiyatkin et al., 1993; DiCiano et al., 1996), including a recent paper on the effects of cocaine on plasma corticosterone in rats (Galica et al., 2000).

While experimenter-administered models of drug abuse have yielded many important findings, the extent to which such data can be extrapolated to human use is limited by the assumption that the effects of response-independent drug are the same as when the drug administration is contingent upon behavior. As demonstrated by the above-mentioned studies, this assumption is not always valid. To date, however, virtually all of the data available on the effects of nicotine in experimental animals is based upon experimenter-administered nicotine.

In the present study, we addressed two important issues. First, does response-contingent (i.e., self-administered) nicotine produce changes in plasma corticosterone, epinephrine, or norephinephrine when nicotine is first experienced in drug-naive rats? Determination of the acute effects of response-contingent nicotine (RC/N) on these systems was seen as a necessary first step in ultimately characterizing these effects and any change, which may occur, such as tolerance or sensitization, as self-administration progresses and drug exposure becomes chronic. Second, do these effects differ depending on whether the nicotine is contingent upon the behavior of the animal?

2. Methods

2.1. General procedures

Male, Sprague–Dawley rats, 41–44-days old and weighing between 200 and 225 g at the start of the experiment, were individually housed in a temperature-controlled environment on a 12-h reverse light/dark cycle (lights off from 7:00 a.m. to 7:00 p.m.) with unlimited access to water. All animals were habituated to the colony room for 7 days during which they received ad lib food. Following this habituation period, animals were trained to lever press for food reinforcement over 3 days. First, animals were habituated to the operant chambers in a single 20-min session. All animals were then food deprived for 24 h and placed in the operant chambers for a 25-min magazine training session. On the third day, animals were hand shaped to respond on the active lever and run through a single continuous reinforcement (CRF) session in which 75 45-mg food reinforcements were given. Lever training and the subsequent experimental session described below took place in a $10 \times 12 \times 11$ in.^{3} operant chamber (BRS/LVE model #RTC-020) with an inactive lever, an active lever with a cue light directly above it, a house light, and a pellet trough. Following food training, animals were fed 20 g at the end of each day (approximately 5:00 p.m.) for the remainder of the study. This feeding schedule results in a gradual weight gain of approximately 15 g/week (Donny et al., 1995).

After training, animals were anesthetized with Equithesin (3 ml/kg i.p.) and implanted with a catheter, similar to that used by Corrigall and Coen (1989), into the right jugular vein. After surgery, animals were given twice daily infusions of ticarcillan plus clavulanate (Timentin: 6.67 mg/kg i.v.) for 3 days and a single infusion on the fourth day after surgery. Animals were allowed 4–8 days to recovery from surgery, during which time their catheters were also flushed daily with 0.1-ml sterile, heparinized saline (5 U/ml).

The experimental session occurred following the 4–8 day recovery period. Rats that were lever trained for food reinforcement and in which catheters remained patent were divided into three groups based on a yoked design (Dworkin et al., 1995). The first group (RC/N) received nicotine bitartrate (0.03 mg/kg/infusion; dose reported as free base) contingent upon a single active lever press (except for responding during the 1-min time-out period). This procedure for presenting RC/N is equivalent to the paradigm we and others have previously used to produce robust and reliable nicotine self-administration in rats (Corrigall and Coen, 1989; Donny et al., 1995, 1998).

Individuals in the second group (Response-Independent Nicotine; RI/N) received the same number of nicotine infusions at identical times during each session as compared to their RC/N partner. Their infusions were contingent upon the partner's responding and not upon their own lever pressing. The third group (Response-Independent Saline; RI/S) was also yoked to individuals in the RC/N group, but received saline infusions instead of nicotine. None of the animals had experience with nicotine prior to this session. The session lasted for 1 h during which time the subjects were connected to a drug delivery swivel system that allowed virtually unrestricted movement in the chamber.

For RC/N animals, responding on an active lever was reinforced on a CRF with infusions of 0.03 mg/kg nicotine bitartrate delivered in a volume of 0.1 ml/kg in approximately 1 s (IITC model 100, pneumatic syringe pump). This dose has been reported by us and others to support maximum responding (Corrigall and Coen, 1989; Donny et al., 1995, 1998). Responding on an inactive lever had no consequence. All infusions were paired with a 1-s cue light and followed by a 1-min time-out period during which the chamber light was turned off and responding was recorded, but not reinforced. This served to minimize any toxic effects that might occur due to rapid and repeated infusions. All changes in the cue and house lights were identical for the three groups, and based upon the responses of the RC/N individual in each cohort. Active and inactive responses and infusions were recorded throughout the session for all three groups.

Since the procedure of transporting the animal to the experimental room and/or attaching it to the swivel mechanism may be stressful and elevate corticosterone, epinephrine, or norepinephrine, a habituation period was used to decrease the likelihood of a stress response affecting our measurements. Habituation consisted of transporting the animals to the experimental room for two consecutive days immediately prior to the testing day, during which animals were connected to the drug delivery system for 1 h each day. For these sessions, the levers were removed to avoid possible extinction, and no infusions were available. The cue and house lights also remained off during this period. In addition, animals were transported to the operant chambers and connected to the drug delivery system for 10 min prior to collecting the pre-session blood sample (as discussed below). Access to the levers was unrestricted during this time, although lever pressing had no consequence.

2.2. Blood draws

Blood samples (0.3–0.5 ml) were collected into sterile syringes containing 80-μl saline with 12.5 mg/ml EDTA, prior to the session (at the end of the 10-min habituation period), at 15 min after the first nicotine or saline infusion, and immediately following the completion of the 1-h session. By using these three-time points, comparisons could be made using pre-session levels as a baseline for determining nicotine-induced changes. The mechanical arrangement of the drug delivery system allowed blood sampling without disconnecting the animals from the swivel attachment or handling the animals. The drug delivery tubing was disconnected outside the test chamber, blood was then drawn from the catheter, and the tubing reconnected with minimal disturbance to the animal. During this period, the computer program operating the operant chamber and infusion pumps was paused until the blood draw had been completed. Immediately after each blood draw, the same volume of sterile saline was used to replace the drawn blood to prevent a decrease in blood volume and to flush the catheter. In addition, after the pre-session and 15-min draw approximately 30 μl of drug solution (nicotine or saline) was infused to refill the connecting tubing and most of the catheter (i.e., to displace the dead space).

All blood samples were put on ice for 1–2 min and then centrifuged at 2000 rpm in a refrigerated centrifuge for 1 min. A 120-μl aliquot of plasma was added to 2.4 μl 5 M perchloric acid in a tube and frozen at −70°C until later assayed for catecholamines. A 10-μl aliquot of plasma was frozen in a separate tube and assayed for corticosterone at a later date.

2.3. Catecholamine and corticosterone assay

Plasma corticosterone was measured by a ^{125}I double antibody radioimmunoassay kit (ICN Biochemicals). The assay has a sensitivity of less than 0.5 μg/dl and an interassay coefficient of variation of 7%. Catecholamine levels were measured by high-performance liquid chromatography with electrochemical detection (Sved, 1989) following extraction of catechols from plasma using alumina (Sved and Fernstrom, 1981). The sensitivity of this assay is less than 50 pg/ml for both epinephrine and norepinephrine.

2.4. Statistical analysis

Of the 87 animals that were trained, implanted with catheters, and run through the experimental procedures, blood was obtained at all three blood draws from a subset of 36 animals. From these rats, 12 epinephrine, 5 norepinephrine and 6 corticosterone samples were lost either due to insufficient plasma volume or technical problems related to the assays. The results presented for corticosterone, epinephrine and norepinephrine each refer to only those animals from which we obtained all three blood draws and successfully completed the assay. Thus, due to the difficulty in obtaining reliable blood draws from the catheter, all three members of each yoked triad could not be used in the final analyses. Therefore, several approaches were taken to ensure that group differences in the neuroendocrine response to nicotine could not be ac-

counted for by differences in response or infusion rates. First, response rates, infusion rates, and time since the last infusion were analyzed to determine if any group difference emerged. Second, where group differences in the neuroendocrine response to nicotine were noted, the behavioral variables were added as covariates and the data were reanalyzed. Third, the subsets of samples for the complete triads were inspected for consistency with any reported group difference.

The timecourse for each measure was analyzed using 3×3 mixed analysis of variance (ANOVA), with blood draw (Draw) as the within-subject factor and treatment group (Group) as the between-subjects factor. Pre-planned comparisons were used to compare the individual groups to each other within a single-time point and to examine the change in hormone levels within each group across the three-time points. In addition, the hormonal response of each group was compared by analyzing the change from draw 1 (baseline) to draws 2 or 3 in an ANOVA with preplanned comparisons. Where significant differences in the change scores between RC/N and RI/N emerged, the data were reanalyzed using the number of active responses and infusions, and the time since the last infusion as covariates to determine if any of the physiological differences could be accounted for by group differences in the distribution of responses or infusions. A significance level of $P < 0.05$ was used for all analyses.

3. Results

The results for active responses, infusions, and time since the last infusion are summarized in Table 1. For the subset of animals used in the corticosterone analysis, active lever responding was higher at the end of the hour in RI/N as compared to RI/S ($P < 0.005$), and there was a trend for more infusions to be taken by RI/N as compared to RC/N. For the animals used in the analysis of epinephrine levels, active lever responses at draw 2 for RI/N ($P < 0.05$) and at draw 3 for both RC/N ($P < 0.05$) and RI/N ($P < 0.05$) were elevated over RI/S, but RC/N and RI/N did not differ from each other. For the norepinephrine results, active lever responding for both RC/N and RI/N was significantly greater than RI/S at draw 3 ($P < 0.05$). Importantly, there were no group differences in the number of infusions received for both the epinephrine and norepinephrine samples.

Both response-contingent and response-independent injections of nicotine elevated corticosterone during the first 15 min of nicotine administration. However, while plasma corticosterone levels declined to baseline in RC/N over the 1-h session, they remained elevated in RI/N (Fig. 1). ANOVA revealed a significant effect of both Draw [$F_{(2,54)} = 18.8$, $P < 0.001$] and the interaction between Group and Draw [$F_{(4,54)} = 3.67$, $P < 0.01$], but no overall effect of Group. There were no significant differences in baseline corticosterone levels between RC/N, RI/N and RI/S. Response-contingent ($P < 0.001$) and RI/N ($P < 0.001$) produced similar increases over pre-session levels in plasma corticosterone at the second blood draw whereas RI/S had no effect. In both groups, the change from baseline was different than in RI/S ($P < 0.001$). Corticosterone levels in the 60-min blood sample remained elevated over baseline for RI/N ($P < 0.001$), but not in RC/N. Furthermore, RC/N showed a significant decrease from draw 2 to draw 3 ($P < 0.005$), but RI/N did not. Direct comparisons

Table 1
Summary of behavioral data

	RC/N			RI/N			RI/S		
	CORT	EPI	NE	CORT	EPI	NE	CORT	EPI	NE
N	7	5	8	9	9	10	14	10	13
Draw 2									
Responses at draw 2	11.43 ± 3.60	12.40 ± 6.79	14.63 ± 5.87	18.78 ± 4.85	19.33 ± 4.80	18.20 ± 4.44	8.79 ± 2.57	5.60 ± 2.91	8.08 ± 2.68
Infusions at draw 2	3.57 ± 0.90	4.60 ± 1.89	4.50 ± 1.31	6.56 ± 1.25	5.78 ± 1.08	6.30 ± 1.17	4.71 ± 0.74	4.10 ± 0.75	4.08 ± 0.59
Min since last infusion	8.43 ± 1.75	6.98 ± 1.72	7.99 ± 1.64	5.87 ± 1.18	6.93 ± 1.33	6.86 ± 1.25	6.76 ± 1.24	5.95 ± 1.41	7.05 ± 1.31
Draw 3									
Responses at draw 3	37.29 ± 15.97	47.20 ± 23.60	52.13 ± 18.53	48.89 ± 9.06	45.22 ± 8.70	46.60 ± 8.95	15.14 ± 3.76	10.30 ± 3.83	17.08 ± 4.80
Infusions at draw 3	13.00 ± 3.60	16.40 ± 5.72	16.75 ± 4.39	20.44 ± 3.92	18.56 ± 3.87	21.20 ± 3.89	15.50 ± 3.02	14.10 ± 3.21	13.08 ± 2.53
Min since last infusion	8.44 ± 1.96	6.40 ± 2.56	6.27 ± 1.83	5.69 ± 1.61	6.11 ± 1.55	4.48 ± 1.26	8.86 ± 2.40	11.34 ± 4.11	8.49 ± 2.5

The number of animals per group, the mean (±SE) number of responses, the mean (±SE) number of infusions, and the mean (±SE) number of minutes since the last infusion prior to both the second and third blood draw for each group used in the analysis of corticosterone, epinephrine and norepinephrine. RC/N = response-contingent nicotine; RI/N = response-independent nicotine; RI/S = response-independent saline; CORT = corticosterone; EPI = epinephrine; NE = norepinephrine.

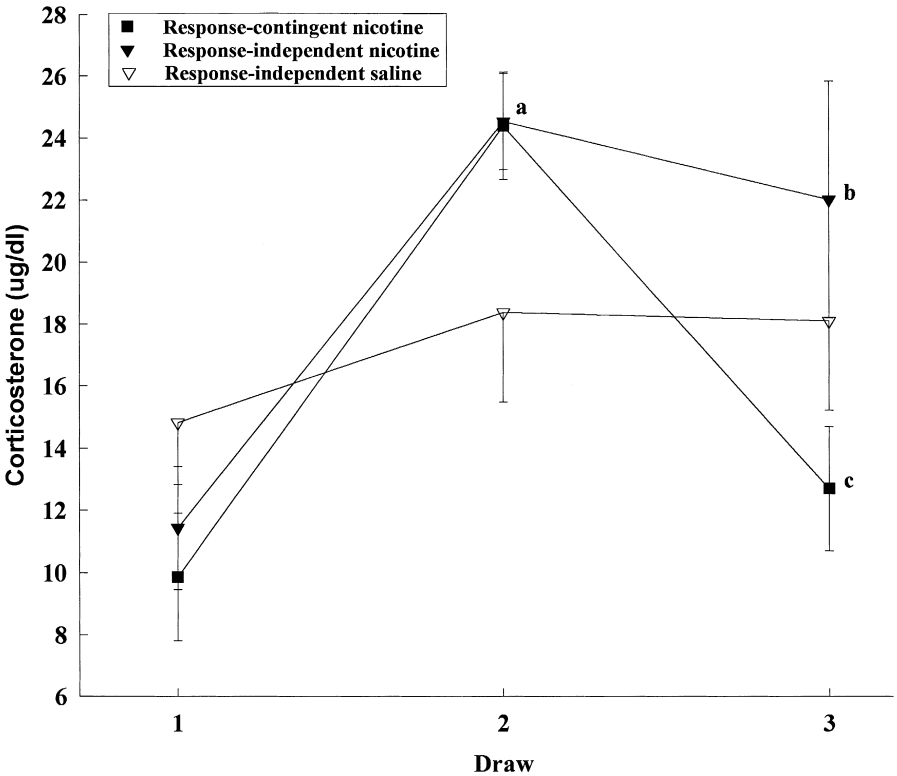

Fig. 1. Mean (±SE) plasma corticosterone levels for each of the three groups at three-time points. (a) Significantly different from draw 1 (RC/N: $F = 18.54$, $P < 0.001$; RI/N: $F = 19.42$, $P < 0.001$). (b) Significantly different from draw 1 ($F = 12.67$, $P < 0.001$). (c) Significantly different from draw 2 ($F = 11.96$, $P < 0.005$).

of the change scores (i.e., draw 1 to draw 3 and draw 2 to draw 3) in RC/N and RI/N failed to reach statistical significance ($P = 0.14$ and 0.08, respectively). These non-significant trends remained unchanged after adding responses, infusions, and time since the last infusion as covariates ($P = 0.15$ for draw 1 to draw 3; $P = 0.08$ for draw 2 to draw 3).

In order to further determine the significance of the differential infusion rates between RI/N and RC/N, we examined the correlations between the number of infusions and the change from baseline at draw 2 and draw 3 for corticosterone for each of the groups. There were no significant correlations in any of the groups ($P > 0.05$).

RI/N elevated epinephrine throughout the session. In contrast, RC/N had no effect (Fig. 2) on plasma epinephrine. There was a significant effect of Group [$F_{(2,21)} = 4.50$, $P < 0.05$], Draw [$F_{(2,42)} = 3.83$, $P < 0.05$] and a Group by Draw interaction [$F_{(4,42)} = 2.74$, $P < 0.05$]. There were no differences between the groups at baseline. RC/N and RI/S produced no change from baseline at either 15 min or at the end of the hour session. RI/N, however, significantly increased epinephrine levels after 15 min as compared to baseline ($P < 0.001$), and they remained elevated over baseline at the end of the session ($P < 0.01$). The increase from draw 1 to draw 2 in RI/N was significantly greater than the change in RC/N ($P < 0.05$) and RI/S ($P < 0.05$). The increase at draw 3 over baseline was also different between RI/N and RC/N ($P < 0.05$). When responses, infusions, and the time since the last infusion were added as covariates, there was a non-significant trend for a difference in the change from baseline between the two nicotine groups at the 15-min time point ($P < 0.10$) and a significant difference at the end of the session ($P < 0.05$).

Analysis of plasma norepinephrine levels revealed a pattern similar to epinephrine; norepinephrine levels were increased by response-independent, but not response-contingent injections of nicotine (Fig. 3). There was an overall effect of Group [$F_{(2,28)} = 5.93$, $P < 0.01$], Draw [$F_{(2,56)} = 15.38$, $P < 0.001$] and a significant interaction effect [$F_{(4,56)} = 4.02$, $P < 0.01$]. Pre-session norepinephrine was similar in all three groups. RC/N and RI/S did not change norepinephrine levels at either the 15-min or post-session time points. RI/N, however, elevated levels over baseline at both the 15 min ($P < 0.001$) and post-session time points ($P < 0.001$). The increase from baseline to draw 2 for RI/N was significantly greater than for both RC/N ($P < 0.01$) and RI/S ($P < 0.001$). Similarly, the increase from baseline to draw 3 was also significantly different between RI/N and both RC/N ($P < 0.05$) and RI/S ($P < 0.01$). Adding responses, infusions, and the time since the last infusion as covariates, did not change

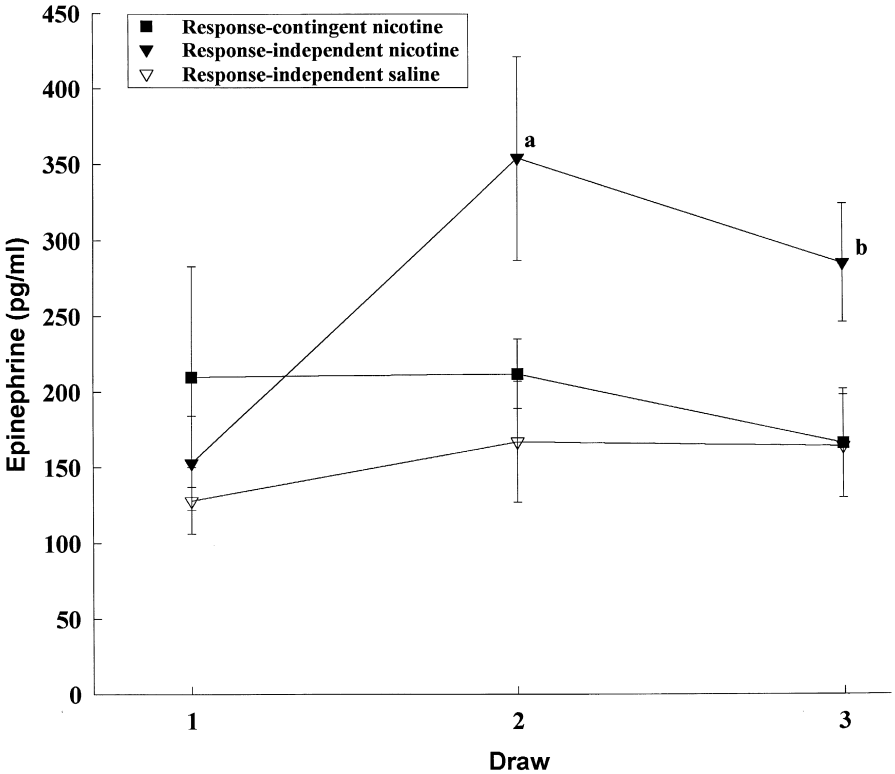

Fig. 2. Mean (±SE) plasma epinephrine levels for each of the three groups at three-time points. (a) Significantly different from draw 1 ($F = 19.68$, $P < 0.001$). (b) Significantly different from draw 1 ($F = 8.42$, $P < 0.01$).

these results; there was still a significant difference between RC/N and RI/N in the change from baseline at both the 15 min ($P < 0.01$) and post-session time points ($P < 0.05$).

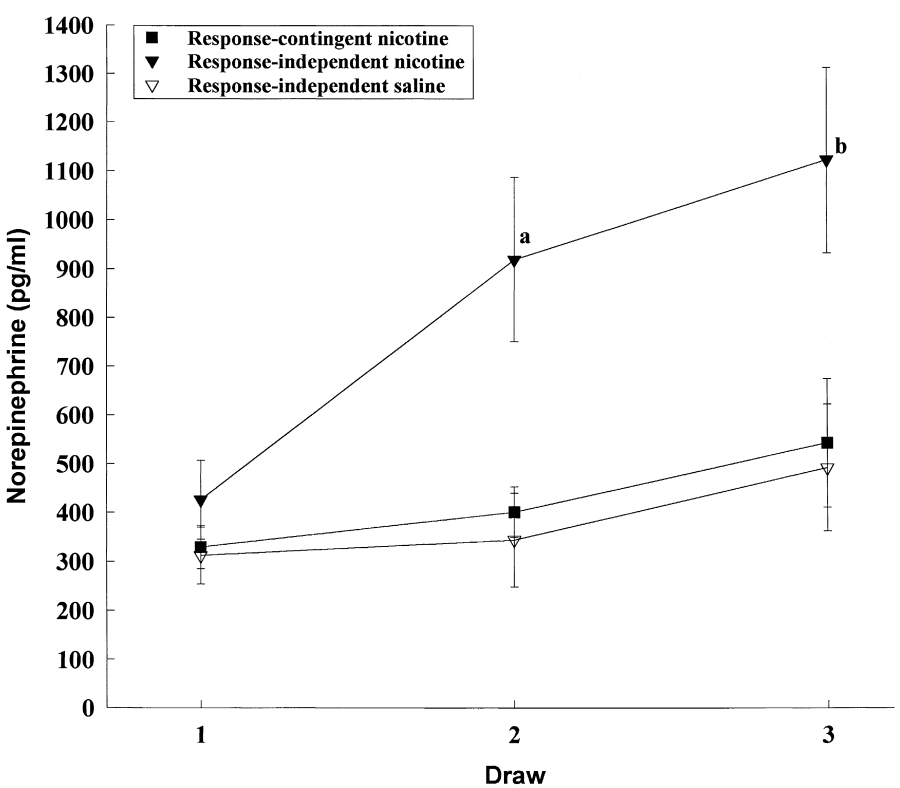

Fig. 3. Mean (±SE) plasma norepinephrine levels for each of the three groups at three-time points. (a) Significantly different from draw 1 ($F = 18.89$, $P < 0.001$). (b) Significantly different from draw 1 ($F = 37.75$, $P < 0.001$).

To further clarify the differences between response-contingent and RI/N, the few triads from which all three blood draws were available for each animal were examined to determine if the same pattern of results was found. There were a total of two, three and four complete triads for epinephrine, norepinephrine, and corticosterone, respectively. Consistent with the previous analyses, when only comparing individuals within a triad, receiving exactly the same number and timing of infusions, similar differences between response-contingent and RI/N were observed for both epinephrine and norepinephrine. Comparison of the four complete triads for corticosterone were less clear with a tendency for a higher baseline in the RI/N group making interpretation difficult.

4. Discussion

The key finding of the present study is that response-contingent (i.e., self-administered) nicotine had a different effect on plasma catecholamine, and possibly corticosterone, levels, compared to nicotine administered in a response-independent manner. Plasma epinephrine and norepinephrine increased in rats receiving RI/N, but not in rats in which nicotine injections were response-contingent. In addition, while RC/N produced an initial increase in corticosterone that returned to baseline by the end of the 1-h session, RI/N resulted in plasma corticosterone levels that remained elevated throughout the 1-h session. Thus, self-administered nicotine elicited responses that are markedly different from those of nicotine administered independent of the animal's behavior.

Experimenter-administered nicotine has previously been shown to elevate plasma corticosterone levels in the rat (Cam and Bassett, 1983; Caggiula et al., 1991). The present work demonstrates that RC/N also elevates corticosterone in a paradigm that supports robust nicotine self-administration (Corrigall and Coen, 1989; Donny et al., 1995, 1998). However, in rats receiving RC/N, plasma levels of corticosterone return to baseline by the end of an hour despite an average of 9.43 additional nicotine infusions. The decrease in corticosterone levels from 15 min after the first infusion to the end of the 1-h session in the RC/N animals may reflect acute tolerance. This interpretation is supported by two additional observations. First, the average time between the most recent infusion received and the time of blood draw was 8.43 min at the second draw and 8.44 min at the end of the session (Table 1), making differences due to the timing of infusions relative to blood draws an unlikely explanation for this decrease in RC/N animals. Second, corticosterone levels in RI/S animals changed little over the session and baseline corticosterone levels for all rats were in the expected range of resting levels for rats in the active (night) phase of their light–dark cycle (10–15 μg/dl), suggesting that the initial increase was a true drug effect and not the result of experimental procedures.

RI/N showed a similar acute increase in corticosterone, but in contrast to RC/N, the elevation was maintained throughout the session. However, rats in the RI/N group tended to receive more infusions of nicotine than rats in the RC/N group, limiting the interpretation of the corticosterone results. Correlational analyses revealed that the amount of nicotine given was not related to the corticosterone response, suggesting that the condition itself (contingent vs. non-contingent) may result in differential hypothalamic–pituitary–adrenocorticalactivation, independent of spurious differences in nicotine exposure. However, in the four complete triads of rats in which corticosterone was measured, the differences in the plasma corticosterone levels between RI/N and RC/N groups were ambiguous. Although these data suggest a potentially important effect of the behavioral contingency of drug delivery on the time course of acute tolerance to nicotine's hypothalamic–pituitary–adrenocortical axis activating effects, additional research with greater control over the amount of nicotine exposure and the timing of blood draws will be needed to confirm this hypothesis.

The difference between response-contingent and RI/N was clear for plasma catecholamine levels. Experimenter-administered nicotine has previously been shown to increase plasma levels of norepinephrine and epinephrine (Grunberg et al., 1988; Morse, 1989; Kiritsy-Roy et al., 1990). Consistent with this literature, we observed an increase in plasma catecholamines after RI/N. However, RC/N failed to elevate plasma epinephrine and norepinephrine. The lack of an elevation in plasma catecholamines reported here contrasts the reported increase observed in smokers (Westfall and Watts, 1963; Volle and Koelle, 1975; Cryer et al., 1976). The differential effects of self-administered nicotine in rats and humans may be accounted for by species differences or by the fact that the present results are for an acute exposure to RC/N in drug-naive animals, while the elevation in smokers is after an extended nicotine self-administration history. Other research from our laboratory supports this interpretation by demonstrating that the hypertensive effects of self-administered nicotine may in fact grow with repeated exposures (Donny et al., 1999).

RC/N may have produced different effects than RI/N for several reasons. One somewhat trivial explanation would be if the differences between groups could be accounted for by differences in bar pressing activity or amount of drug intake. These explanations are unlikely, however, since response rates did not differ significantly between RC/N and RI/N groups. Furthermore, when the catecholamine data were reanalyzed using active responses, infusions, and time since the last infusion as covariates, the same differences between response-contingent and RI/N were noted.

Another possible methodological explanation for the difference between the two nicotine groups could be that the hormonal response in RI/N animals resulted from extinction in previously food-trained animals, an effect that might be minimized in RC/N animals in which the operant is transferred to another reinforcer. However, the RI/S animals, which also experienced extinction, did not have an increase in catecholamine levels, making this explanation unlikely.

Predictability of drug delivery is another potential source for the difference between the effects of response-contingent and RI/N. Cues predicting drug delivery could account for differences in corticosterone or catecholamine responses (Caggiula et al., 1991). For the present experiment, this explanation seems unlikely since environmental cues predicting infusions were the same across groups. In all groups, a discrete 1-s cue light was paired with each infusion and followed by a 1-min time-out period during which the house light was turned off. There were no other planned environmental cues that would allow prediction of the infusions for any group. However, the greater predictability afforded by interoceptive cues in the response-contingent, as compared to the response-independent, condition cannot be ruled out (Mineka and Henderson, 1985).

Finally, one important theoretical distinction between response-contingent and RI/N is controllability. Controllability has been shown to be an important factor in modulating stress (Borysenko and Borysenko, 1982) and drug effects (Mineka and Henderson, 1985). Response-contingent animals, by definition, have control over drug infusions whereas response-independent animals do not. Determining whether controllability, as compared to predictability, explains the group differences is beyond the scope of the present data. Future studies are needed to examine whether predictability or controllability play an important role in mediating such differences.

Whatever the explanation for the difference between response-contingent and RI/N, the difference develops very rapidly. Part of the reason for rapid development of this contingency may be that in rats already trained to expect reinforcement following bar pressing, nicotine sufficiently substitutes for food (Swedberg et al., 1990). Without prior food training, the contingency would be expected to develop much more slowly (Shoaib et al., 1997), in which case, any difference between response-contingent and RI/N may not be seen until a greater number of pairings between lever pressing behavior and nicotine infusion.

Glucocorticoids have been suggested as possible mediators of sensitivity to nicotine and the development of nicotine tolerance (Pauly et al., 1988, 1992; Caggiula et al., 1993, 1994). However, these hypotheses have relied on studies showing increases in corticosterone in response to experimenter-administered nicotine with the assumption that self-administered nicotine produces the same effect. The data presented here are the first to demonstrate that nicotine also increases corticosterone when it is contingent on the animal's behavior. In addition, they indicate a potentially important difference in the glucocorticoid response to acute administration of response-contingent and RI/N, which implies potential differences in sensitivity to other effects of nicotine (e.g., analgesia) known to be modulated by hypothalamic–pituitary–adrenocortical axis activity (Caggiula et al., 1993).

Finally, it is important to note that active lever response rates were similar for response-contingent and RI/N — both were greater than saline. The elevation in responding for RI/N here may have been due to nicotine's direct locomotor effects, coincidental pairings of active responses with yoked infusions, or an effect of nicotine on the rate of extinction following training for food reinforcement. However, other work from our laboratory in a larger group of animals (16 triads) has shown that contingent nicotine maintains greater active lever responding than non-contingent nicotine on the first day of acquisition as well as over a 9-day period (Donny et al., 1998), suggesting that the elevation in response rate for RI/N seen here may have been spurious.

Our results suggest that RC/N produces short-lived changes in the activity of the hypothalamic–pituitary–adrenocortical axis and fails to alter sympathetic nervous system activity. The differences reported between the effects of response-contingent and RI/N challenge the assumption that self-administered and experimenter-administered nicotine invariably produce similar effects. This raises the important question: to what extent do other physiological and behavioral effects reported for experimenter-administered nicotine occur when animals self-administer this drug? Successful nicotine self-administration procedures have now been reported by several laboratories (Corrigall and Coen, 1989; Donny et al., 1995, 1998, 1999; Tessari et al., 1995; Chiamulera et al., 1996; Valentine et al., 1997; Shoaib et al., 1997) including a report examining the overlap in the neural substrates underlying nicotine and cocaine self-administration (Pich et al., 1997). Future studies are needed to explore the possibility that neuroadaptations to chronic nicotine exposure — such as tolerance or sensitization — would exhibit these differences, depending on whether the nicotine administration is contingent on or independent of the behavior of the animal. Further use of the self-administration model when studying the biology of nicotine will permit questions to be addressed in ways of greater relevance to human smoking behavior.

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