Behavioral Neuroscience
1989, Vol. 103, No. 2, 356-364

0735-7044/89/$00.75

Chronic Voluntary Nicotine Drinking Enhances Nicotine Palatability in Rats

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The response of rats to nicotine solutions was examined with the brief-exposure, taste reactivity test and a two-bottle, 24-hr preference test. In Experiment 1, naive nondeprived rats were administered intraoral infusions (0.8 ml infused during 1 min) of distilled water and 1 μg/ml, 5 μg/ml, 10 μg/ml, 25 μg/ml, 50 μg/ml, and 100 μg/ml nicotine. The oral motor, taste reactivity (TR) responses of the rats were recorded during the infusion. Nicotine solutions up to a concentration of 50 μg/ml elicited a number of ingestive TR responses similar to that by water. Ingestive responses significantly decreased and aversive TR responses significantly increased in response to 100 μg/ml nicotine. On the basis of these results, two-bottle preferences for water versus 1 μg/ml, 5 μg/ml, and 0 μg/ml (water control group) nicotine were measured in three groups of naive rats. Rats initially showed an equal preference for 0 μg/ml and 1 μg/ml nicotine. After 16 days of exposure, however, rats developed a significant preference for 1 μg/ml nicotine. The preference ratio for 5 μg/ml nicotine significantly increased during the experiment, but the preference ratio remained significantly less than that for 1 μg/ml and 0 μg/ml nicotine solutions. Last, TR responses elicited by intraoral infusions of 1 μg/ml and 5 μg/ml nicotine were then measured in these rats having had the two-bottle experience. Rats showing a two-bottle preference for the 1 μg/ml nicotine solution displayed significantly more ingestive TR responses to 1 μg/ml and 5 μg/ml nicotine than did the control rats. These data indicate that prolonged voluntary access to nicotine results in an increased preference for nicotine and modifies the immediate oral/gustatory reactivity of the rats to nicotine.

An animal's preference for a taste stimulus is determined not only by the chemical properties of the stimulus but also by associative or experiential factors. The powerful effect of experience on taste preference is demonstrated by conditioned taste aversion studies. In this procedure, a preferred taste (e.g., sucrose) is subsequently avoided and elicits aversive oral motor, taste reactivity responses after it has been paired with visceral malaise (Garcia, Hankins, & Rusiniak, 1974; Garcia, Kimeldorf, & Koelling, 1955; Grill & Norgren, 1978a; Logue, 1979). Conversely, taste preferences can be established or enhanced by experiential factors; for example, taste preferences are developed in humans and rats by associating a taste with positive reinforcement (a preferred taste or a nutritive stomach load; Booth, Mather, & Fuller, 1982; Booth, Stoloff, & Nicholls, 1974; Fanselow & Birk, 1982; Holman, 1975; Mehiel & Bolles, 1984; Revusky, 1974; Zellner, Berridge, Grill, & Ternes, 1985).

Nicotine is a highly effective positive reinforcer in humans (Russell, 1980). In addition, the preference for taste stimuli is increased after associations with nicotine's reinforcing properties; for example, chronic smokers rate cigarette smoking as "tasting good," whereas nonsmokers do not (Shor, Williams, Canon, Latta, & Shor, 1981). Similarly, with a careful selection of contingencies and injection parameters, nicotine is reinforcing to laboratory animals (Goldberg, Spealman, & Goldberg, 1981). A corresponding shift in preference for taste stimuli associated with nicotine, however, has not been reported in laboratory animals.

One objective of the following experiments was to determine whether rats given prolonged, voluntary access to nicotine solutions would develop a preference for nicotine. A change in preference for the nicotine solution may reflect the association of the taste with the reinforcing properties of nicotine. What is the effect of this association on the palatability or the oral reinforcing properties of the nicotine solution? It is often assumed that a shift in preference or intake reflects a change in the palatability of the stimulus (Fanselow & Birk, 1982; Garcia et al., 1974). This is not always the case; for example, pairing sucrose with lithium chloride toxicosis or footshock results in an equal decrease in the preference for a sucrose solution. Taste reactivity (TR; Grill & Norgren, 1978a) analysis revealed that sucrose elicits aversive responses after being paired with LiCl toxicosis. In contrast, sucrose paired with footshock continues to elicit ingestive TR responses (Pelchat, Grill, Rozin, & Jacobs, 1983). As such, intake measures alone are ambiguous indicators as to whether the palatability of the stimulus is changed by experience. Thus, the second objective of the experiments was to determine whether any change in preference that we might observe was accompanied by an increase in nicotine's immediate oral reinforcing properties.

This research was supported by National Institutes of Health Grant NS-24879 awarded to F. W. Flynn and BRSG Grant 2 SO7-RR07157-08 and 09 awarded by the National Institutes of Health.

We express our appreciation to Catherine Valades and Terry Davidson for their helpful comments on the manuscript.

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In order to select the nicotine concentrations to be used in the following experiments, the taste reactivity test was used to provide normative data on the gustatory responsiveness of naive rats to nicotine (Experiment 1). Based on these results, two nicotine concentrations were selected. Naive rats were given access to one of the two nicotine solutions and water for 2 months. The results of Experiment 2 show that under voluntary intake conditions, the rat's preference for nicotine solutions significantly increases over days. Taste reactivity responses elicited by nicotine were then measured in these rats (Experiment 3) in order to determine whether the palatability of the nicotine solutions increased as a function of prolonged, voluntary nicotine intake.

Experiment 1: Taste Reactivity Responses Elicited by Nicotine in Naive Rats

Method

Subjects and surgery. Male Sprague-Dawley rats ($n = 8$) were housed in individual cages in a colony room with a 12:12 hr light/dark cycle. All testing was conducted during the light phase of this cycle. Rats were fitted with two intraoral cannulas (Grill & Norgren, 1978b). Cannulas were constructed from lengths of polyethylene (PE-50) tubing that was heat-flared at one end. Rats were anesthetized with sodium pentobarbital. The cannulas were positioned anterior-lateral to the maxillary molars and drawn out subcutaneously where they were cemented to screws anchored to the skull. Rats were treated with bicillin (10,000 U) every other day for 6 days following surgery. Throughout the experiment rats had ad-lib access to Purina chow and tap water. No formal deprivation period was used.

Taste reactivity testing. Testing took place in a clear plastic cylindrical chamber. The chamber was elevated on four legs, and a mirror attached to the legs permitted an unobstructed view of the rat's mouth. A video camera was positioned several inches from the mirror and was used to record the oral motor behaviors during the minute that the taste stimulus was infused into the rat's mouth. Subsequently, the videotapes were viewed in slow motion, and the occurrences of the TR responses were recorded. TR responses are scored as either ingestive or aversive. Ingestive taste reactivity responses include (a) TP—tongue protrusions, the tongue extends beyond the plane of the incisors; (b) LTP—lateral tongue protrusions, the tongue protrudes and breaks the plane of the incisors as it is extended laterally to one or the other side of the mouth; and (c) MM—mouth movements, the mandible moves rhythmically but the tongue is not extended. Aversive TR responses generally act to expel the taste from the rats' mouths and include (a) HS—head shakes, the head is shaken side-to-side at a rate greater than 60 Hz; (b) FF—forelimb flails, during the execution of this response fluid is deposited onto the forelimbs, and then the forelimbs are shaken rapidly; (c) CR—chin rubs, the chin is put into contact with the floor or walls as the body is projected forward by flexion of the dorsal neck and by pectoral and forelimb musculature; (d) G—gapes are large amplitude openings of the mandible with a concomitant retraction of the corners of the mouth; and (e) PD—passive drip, fluid is accumulated in the mouth and permitted to drip to the floor (Berridge, Grill & Norgren, 1981; Grill & Norgren, 1978b). With the exception of passive drip (which was scored in seconds), all these TR responses are discrete events and were scored each time they occurred during the 1-min intraoral infusion. The total number of ingestive (MM + TP + LTP) and aversive TR responses (G + HS + CR + FF) was then tabulated (Flynn & Grill, 1988), and these values were entered into the appropriate analysis of variance.

Rats were habituated to the test procedure for 3 days prior to the start of testing. During Day 1 of habituation, each rat was placed in the test chamber and remained there for 15–20 minutes. On Days 2 and 3, the intraoral cannulas of the rat were connected to stimulus-filled PE tubings. The distal end of the tubing was attached to a syringe mounted on an infusion pump. The rat was then placed in the chamber and left undisturbed for 10 min. The infusion pump and timer were then turned on, and distilled water was infused into the rat's mouth. Throughout the experiment tastes were delivered during a 1-min period at a rate of 0.8 ml/min. Following the habituation phase, the test phase was begun. Rats were given the following order of taste stimuli: distilled water, then 1 μg/ml, 5 μg/ml, 10 μg/ml, 25 μg/ml, 50 μg/ml, and 100 μg/ml nicotine. The nicotine (nicotine sulfate; Sigma Chemical Co.) was prepared fresh daily in distilled water. Taste stimuli were delivered on successive days; one taste was delivered each day.

Results

The total number of ingestive and aversive TR responses was analyzed by separate, one-way repeated (concentration) measures analyses of variance. As shown in Figure 1, the total number of ingestive TR responses varied significantly with taste stimulus, $F(6, 54) = 11.4$, $p < .001$. Further analyses indicated that distilled water and 1 μg/ml, 5 μg/ml, 10 μg/ml, 25 μg/ml, and 50 μg/ml nicotine solutions elicited similar numbers of ingestive TR responses. As the nicotine concentration was increased to 100 μg/ml, however, the total number of ingestive TR responses decreased significantly, relative to the previous nicotine concentrations ($ps < .01$).

The number of aversive TR responses varied with taste stimulus, $F(6, 54) = 16.4$, $p < .01$ (see Figure 1). Very few aversive TR responses were elicited by water and 1 μg/ml, 5 μg/ml, 10 μg/ml, and 25 μg/ml nicotine solutions. Aversive responses significantly increased as nicotine concentrations were increased to 50 μg/ml and 100 μg/ml ($ps < .01$). These two concentrations of nicotine elicited similar numbers of aversive TR responses.

In summary, nicotine concentrations of 1 μg/ml, 5 μg/ml, 10 μg/ml, and 25 μg/ml elicited primarily ingestive TR responses. The aversive quality of nicotine first appeared in response to 50 μg/ml nicotine. At this concentration the number of aversive TR responses significantly increased. The highest concentration of nicotine tested (100 μg/ml) elicited few ingestive responses and predominantly aversive TR responses.

Discussion

Nicotine was found to elicit ingestive TR responses in nondeprived rats at concentrations up to 50 μg/ml. Only in response to intraoral infusions of 50 μg/ml and 100 μg/ml nicotine did the animals respond aversively. At concentrations up to 50 μg/ml, the present taste reactivity results conflict with those of long-term, 24-hr intake tests, which indicate that the rat's preference for 1 μg/ml nicotine is equal to that for water and that the preference significantly decreases as the nicotine concentration is increased. Furthermore, during long-term intake tests, rats consume virtually none of a 48 μg/ml nicotine solution (Naquira & Argueros, 1978).

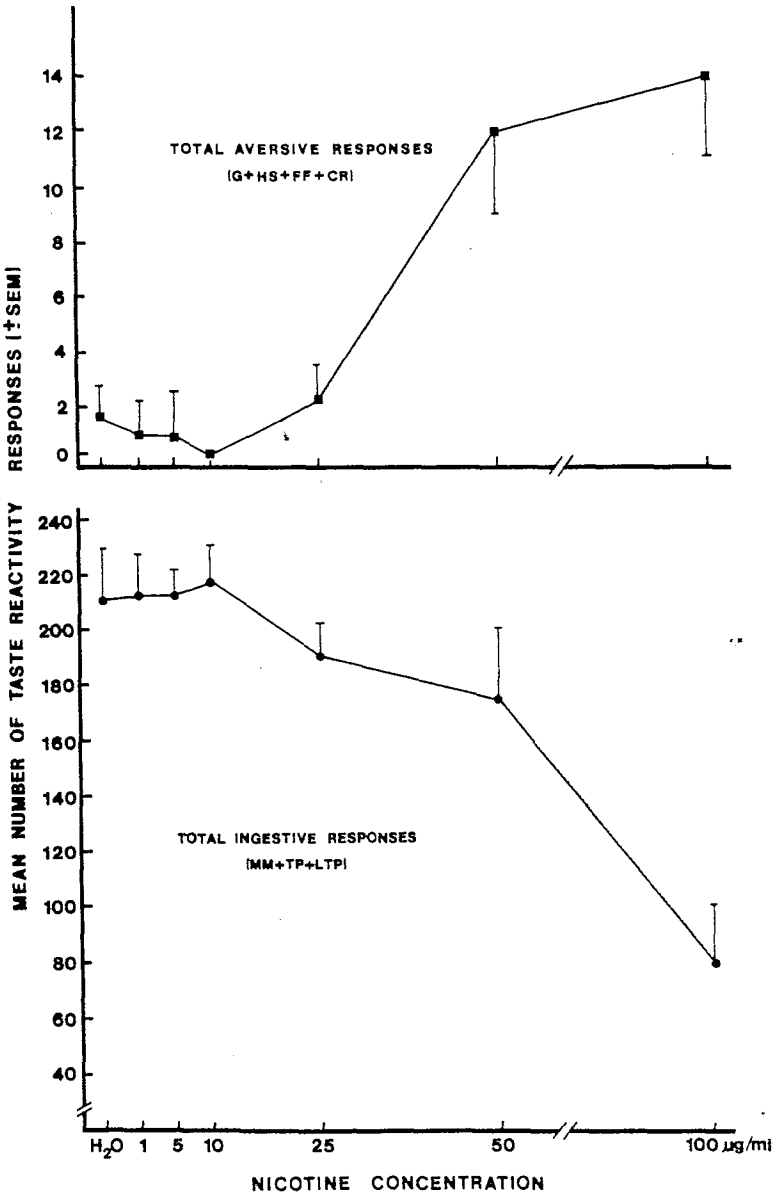

Figure 1. Mean (± *SE*) number of ingestive (lower panel) and aversive (upper panel) taste reactivity responses elicited by 0.8-ml intraoral infusions of water and nicotine solutions in naive rats having ad-lib access to chow and water. (G = gapes; HS = head shakes; FF = forelimb flails; CR = chin rubs; MM = mouth movements; TP = tongue protrusions; LTP = lateral tongue protrusions.)

Long-term intake measurement and the brief-access taste reactivity test gave opposite nicotine-concentration–response relations. Previous two-bottle intake results gave the impression that nicotine is an aversive taste stimulus, whereas in the present study the brief-exposure taste reactivity results indicate that the oral stimulating properties of nicotine are not aversive below a concentration of 50 µg/ml. Long-term, 24-hr intake tests are influenced by factors other than taste, such as inhibitory postoral signals produced by ingestion of the stimulus (Davis & Levine, 1977; Flynn & Grill, 1988; Mook, 1963). A conclusion reached by contrasting long-term and the present taste reactivity results is that the

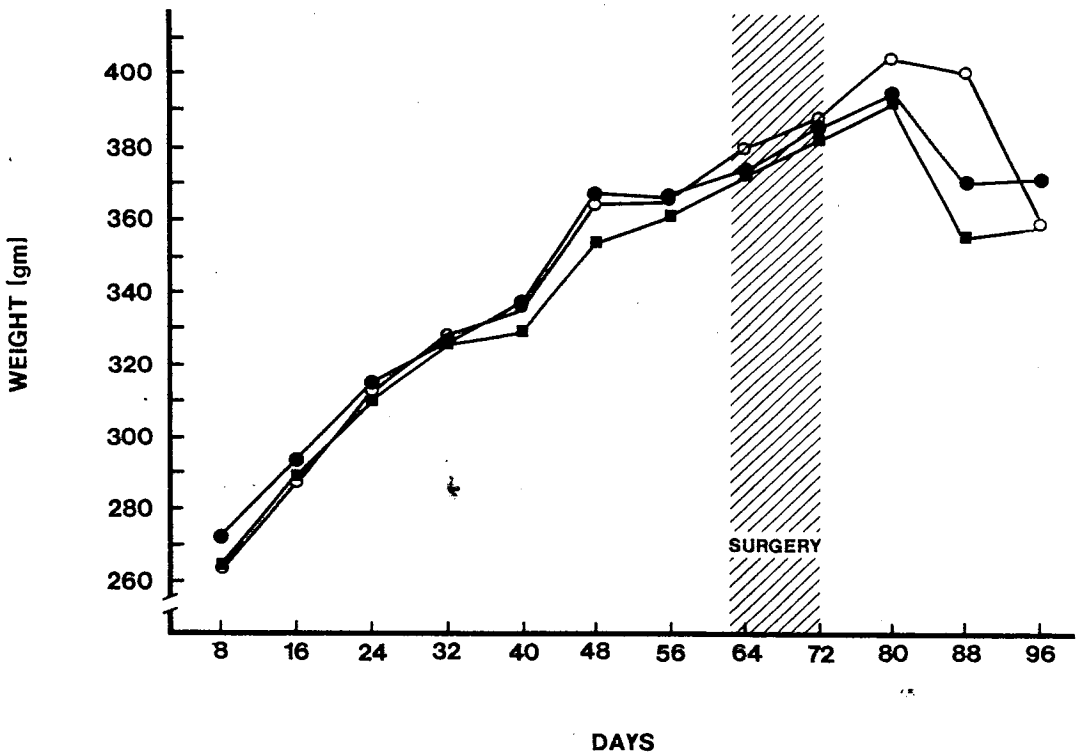

Figure 2. Body weights (in g) of rats having access to 1 μg/ml nicotine (solid squares), 5 μg/ml nicotine (solid circles), and 0 μg/ml nicotine (open circles).

rats' avoidance of nicotine solutions below concentrations of 50 μg/ml is due to postoral feedback and not aversive oral stimulation.

Experiment 2: Two-Bottle Preference for Nicotine Solutions

The objective of this experiment was to determine whether rats having access to water and a nicotine solution would develop a preference for nicotine over days. On the basis of the taste reactivity results of Experiment 1, two concentrations of nicotine, 1 μg/ml and 5 μg/ml, were selected to be used. These concentrations elicited predominantly ingestive TR responses.

Method

Subjects. Male Sprague-Dawley rats (n = 24) were matched on the basis of body weight and assigned to one of three groups. Rats were housed individually in standard suspended wire mesh cages in a temperature-controlled colony room with a 12:12 hr light/dark cycle. Two calibrated bottles were attached to the front of each cage; one contained tap water, and the other was the nicotine bottle that contained 1 μg/ml nicotine (n = 9), 5 μg/ml nicotine (n = 7), or 0 μg/ml (tap water; control group, n = 8). Nicotine sulfate solutions were prepared fresh daily. Nicotine and water intake (nearest milliliter) were recorded every 24 hr. Bottles were then refilled with fresh solutions, and the bottle positions were switched when they were reattached. Preference ratios were computed daily for 96 days.

Results

Body weight. All rats showed a similar and significant increase in body weight during the two-bottle preference test, $F(11, 231) = 284.6$, $p < .001$ (Figure 2). During this time, the body weights of the groups were not significantly different. Following intraoral cannula surgery, body weights significantly declined in all three groups ($ps < .01$).

Preference ratios (nicotine intake/total fluid intake × 100), nicotine intake (both in micrograms and milliliters), water intake, and total fluid intake volumes were averaged over 8-day blocks. In Figures 3, 4, and 5, the data points corresponding to "Day 8" reflects the average of Days 1–8, data for "Day 16" are the averages of Days 9–16, and so on. Eight-day averages were then analyzed by 3 (group) × 12 (day blocks) analyses of variance.

Two-bottle preference. As shown in Figure 3, the groups differed significantly in their preference ratios for nicotine, $F(2, 21) = 57.1$, $p < .001$. Multiple comparisons revealed that the preference ratio for 1 μg/ml nicotine was significantly

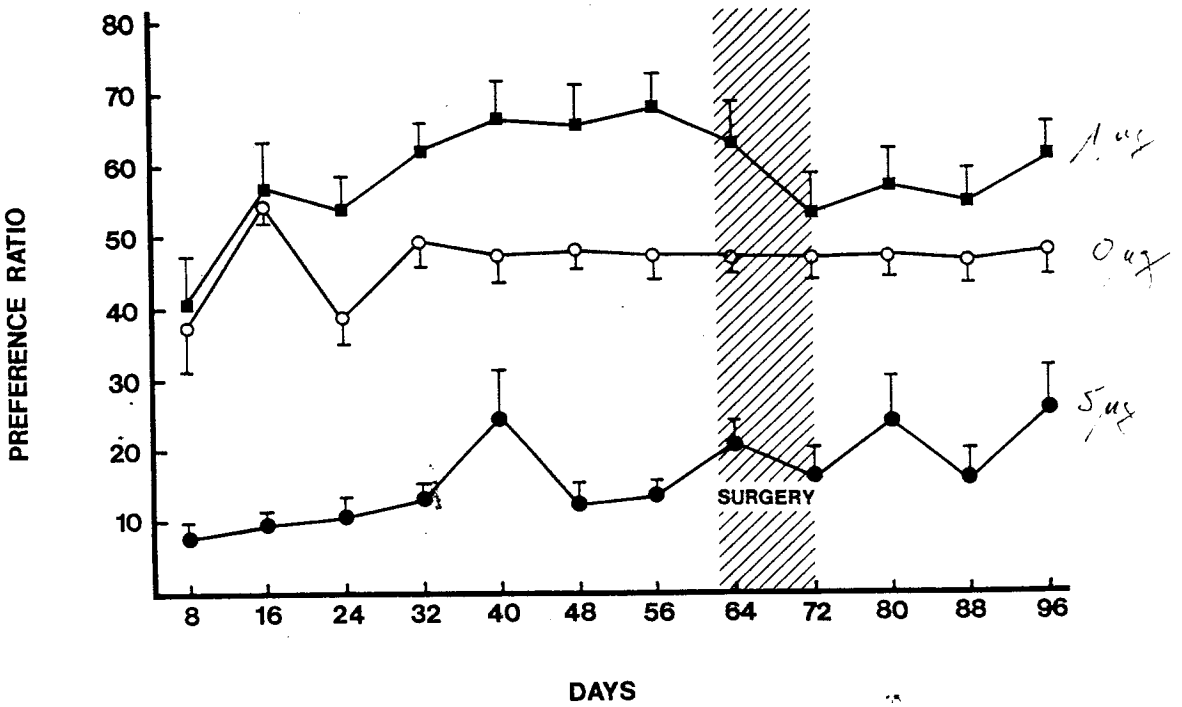

Figure 3. Mean (± *SE*) preference ratios of rats given access to 1 µg/ml nicotine (solid squares), 5 µg/ml nicotine (solid circles), and 0 µg/ml nicotine (open circles). (Each data point reflects an 8-day average.)

greater than that for 0 µg/ml (water) and 5 µg/ml nicotine (*p*s < .02). Also, the preference ratio for 5 µg/ml nicotine was significantly less than that for 0 µg ($p < .01$).

Preference ratios showed a significant change over days, $F(11, 231) = 5.1$, $p < .001$, and a marginally significant Group × Days interaction, $F(22, 231) = 1.6$, $p < .06$. Initially, the preference ratio for 1 µg/ml nicotine was not significantly different from that of the control group's preference for 0 µg/ml nicotine. The preference ratio for 1 µg/ml nicotine, however, significantly increased during the experiment ($p < .001$), whereas that for 0 µg/ml did not vary significantly over days. Also, the preference ratio for 5 µg/ml significantly increased from approximately 8% at the start of the experiment to 26% at Day 96 ($p < .001$).

Nicotine solution intake (in milliliters). The amounts consumed from the nicotine bottles (1 µg/ml, 5 µg/ml, and 0 µg/ml) are shown in Figure 4. Multiple comparisons of the significant main effect of group, $F(2, 21) = 42.3$, $p < .001$, revealed that rats consumed significantly more 1 µg/ml nicotine than 5 µg/ml nicotine and 0 µg/ml nicotine solution (*p*s < .02). Also, intake of 0 µg/ml was significantly greater than that of 5 µg/ml nicotine ($p < .001$).

Intake of the solutions in the nicotine bottles (1 µg/ml, 5 µg/ml, and 0 µg/ml) changed significantly during the days of the experiment, $F(11, 231) = 4.9$, $p < .001$. As shown in Figure 4, intake of 1 µg/ml nicotine and 5 µg/ml nicotine increased over days (*p*s < .01), whereas intake of 0 µg/ml nicotine remained at approximately 20 ml.

Nicotine intake (in micrograms). Analysis of variance showed a nonsignificant trend for rats of the 5 µg/ml nicotine solution group to consume more nicotine than did rats having access to 1 µg/ml nicotine solution, $F(1, 14) = 3.8$, $p < .07$ (see Figure 5). The significant Group × Days interaction reflected that after Day 56, rats having access to the 5 µg/ml nicotine solution consumed significantly more nicotine than did rats having the 1 µg/ml solution, $F(11, 154) = 2.8$, $p < .01$.

Water intake. Rats having access to 5 µg/ml nicotine drank significantly more water (36.4 ± 3.0 ml) than did rats of the 1 µg/ml nicotine group (17.9 ± 2.6 ml), $F(2, 21) = 211.8$, $p < .001$. The significant decrease in water intake during the course of the experiment, $F(11, 231) = 7.8$, $p < .001$, was due to the change in water intake by rats having access to 1 µg/ml nicotine. Intake of water by rats in the 0 µg/ml group and water intake by rats in 5 µg/ml group were stable during the experiment.

Total fluid intake. The total two-bottle fluid consumption by rats in the 1 µg/ml nicotine group (43.5 ± 2.6 ml), 5 µg/ml nicotine group (43.0 ± 3.2 ml), and 0 µg/ml group (41.9 ± 2.8 ml) was not significantly different. This indicates that rats accurately compensated for the intake of the nicotine solutions by decreasing their water intake. As such, nicotine solution consumption did not suppress the overall fluid intake by the rats.

The significant main effect of days, $F(11, 231) = 8.9$, $p < .001$, reflected that the total fluid intake transiently decreased

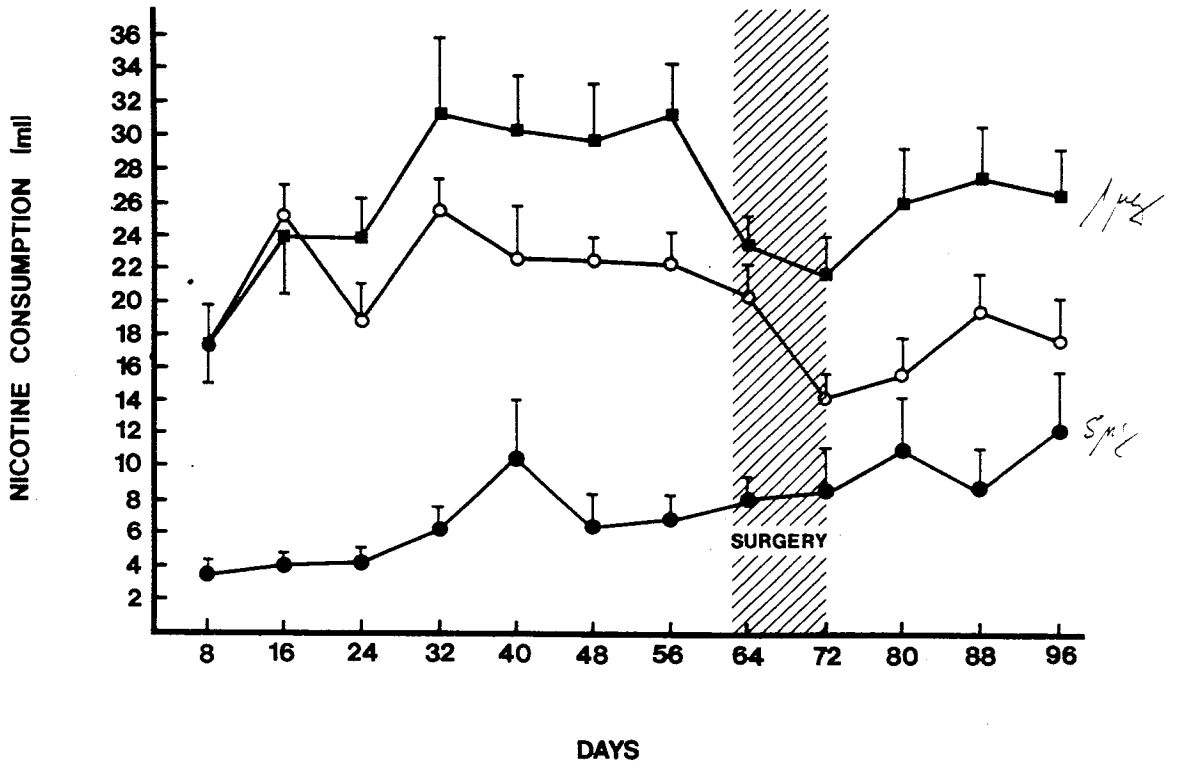

Figure 4. Daily nicotine solution intake (in ml) averaged over 8-day blocks. (Rats having access to 1 μg/ml nicotine [solid squares] consumed significantly more of the solution than rats given 5 μg/ml nicotine [solid circles] and 0 μg/ml [water; open circles].)

immediately following surgery (Days 64–72) in all three groups ($ps < .05$). The total fluid intake by all groups then increased, and intake returned to preoperative levels by Day 88.

Experiment 3: Taste Reactivity Responses Elicited by Nicotine Following Two-Bottle Preference Test

The results of Experiment 2 demonstrate that rats given voluntary access to 1 μg/ml nicotine develop a significant preference for this solution over water. Also, the preference for 5 μg/ml nicotine was shown to increase over days, but the preference ratio did not exceed that for water. The shift in preference for 1 μg/ml nicotine may or may not reflect an increase in the palatability of the nicotine solution. Rats may consume more of the nicotine because of its postoral reinforcing consequences (Goldberg et al., 1981) independently of any change in the oral reinforcing properties or palatability of the stimulus. In order to determine whether the two-bottle preference for nicotine reflected a change in the animal's response to the oral stimulating properties of the nicotine solution, TR responses elicited by 0.8-ml intraoral infusions of 1 μg/ml and 5 μg/ml were measured in those rats used in Experiment 2.

Method

Intraoral cannulas. As indicated in Figures 2–5, intraoral cannula surgery began on Day 60. Rats used in Experiment 2 ($n = 24$) were anesthetized with Nembutal, and intraoral cannulas were positioned according to the procedure described above. Following surgery, rats were returned to their home cages with ad-lib access to Purina chow and their two-bottle regimen (water and 1 μg/ml, 5 μg/ml, or 0 μg/ml nicotine). Intakes from the nicotine-labeled bottles and from the water bottle were recorded daily, and bottle positions were reversed every 24 hr. Rats were allowed to recover for 2 weeks prior to the start of TR adaptation and testing.

Taste reactivity testing. The apparatus and procedure described in Experiment 1 were used. Taste reactivity adaptation and testing took place during Days 80–96. During this time, rats continued to have two-bottle access and ad-lib Purina chow. All testing was conducted between 0900 and 1200 hours. Rats were adapted to the intraoral infusion procedure for 4 days. On Days 1 and 2, the rat's intraoral cannulas were attached to PE tubing filled with distilled water. The rat was then placed in a clear plastic chamber and left to adapt for 10–15 min. An infusion pump was then turned on, and the distilled water was infused at a rate of 0.8 ml/min for 1 min. On Days 3 and 4, rats were adapted to intraoral infusions of nicotine. Nicotine sulfate solutions were prepared fresh daily in distilled water. The exposure to intraoral nicotine infusions was intended primarily to familiarize the control rats (0 μg/ml group) with the taste of nico-

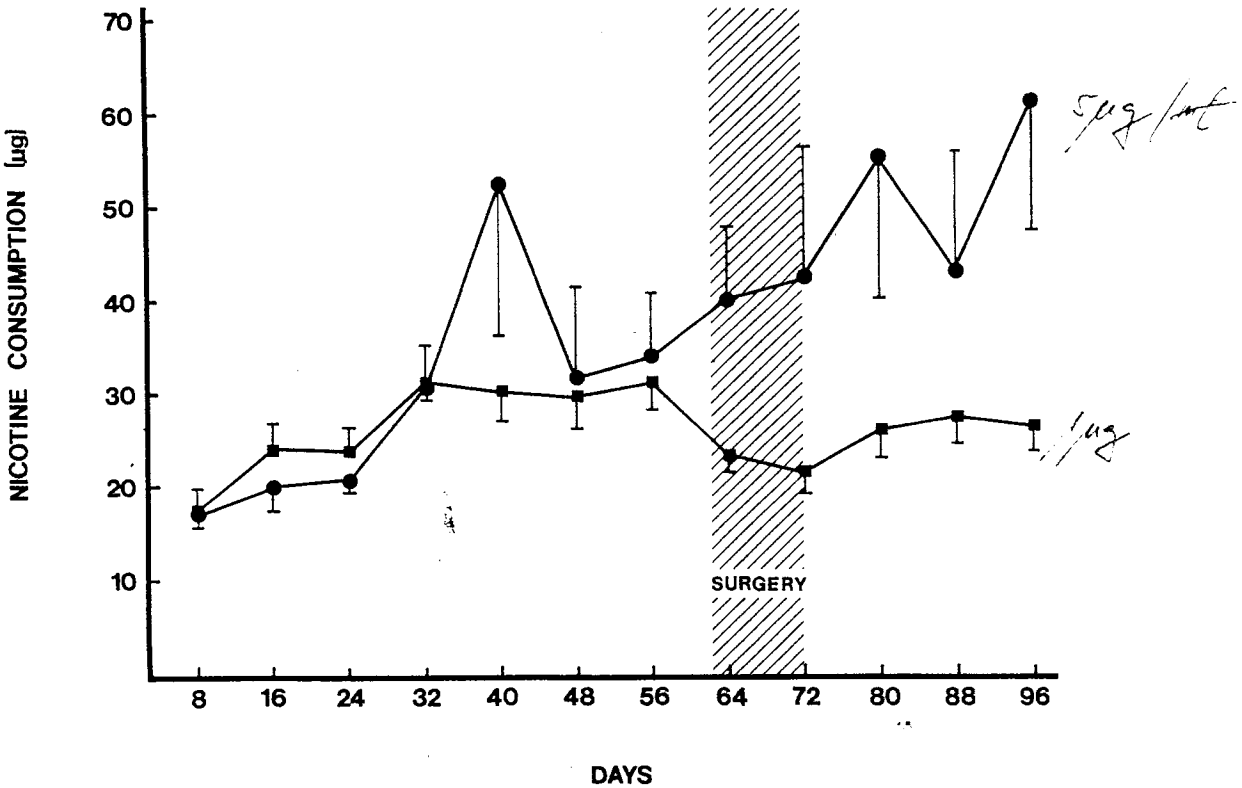

Figure 5. Mean (± *SE*) daily nicotine intake (in µg) averaged over 8-day blocks for rats given access to 1 µg/ml (solid squares) and 5 µg/ml nicotine solution (solid circles).

tine and reduce the likelihood that any taste reactivity group differences could be explained by neophobia. On days when the rats were administered nicotine, half of the rats in each group were first given an 0.8-ml intraoral infusion of 1 µg/ml nicotine, and the remaining rats an 0.8-ml infusion of 5 µg/ml nicotine. Fifteen minutes following the termination of the first infusion, the rats were administered the other nicotine solution. Following the second intraoral nicotine infusion, the intraoral cannulas were separated from the PE tubings, and the rats were returned to their home cages. The same procedure was followed on Day 5; however, during the intraoral infusions the video equipment was turned on to record the rats' oral motor responses that were elicited by the stimulus during the 1-min intraoral infusions. The videotapes were subsequently analyzed for the number of ingestive and aversive TR responses.

Results

Taste reactivity analyses are based on the data collected from 17 rats (1 µg/ml group, $n = 6$; 5 µg/ml group, $n = 5$; 0 µg/ml group, $n = 6$). During the days that the TR responses were videotaped, analysis of the two-bottle intake showed that the rats' preference ratio for 1 µg/ml nicotine (66.1% ± 4.7%) was significantly higher than that for 5 µg/ml nicotine (26.0% ± 4.9%) and water (47.9% ± 3.5%; $ps < .05$).

1 µg/ml nicotine. The total number of ingestive TR responses (MM + TP + LTP) and aversive responses (G + HS + FF + CR) elicited by the nicotine infusions in the groups were analyzed by separate one-way analyses of variance. All rats ingested the 0.8-ml intraoral infusion of 1 µg/ml nicotine, and there were virtually no aversive TR responses elicited by the stimulus. As shown in Figure 6, the total number of ingestive responses elicited by intraoral infusions of 1 µg/ml nicotine differed significantly between the groups, $F(2, 14) = 4.1$, $p < .05$. Newman-Keuls tests revealed that intraoral infusions of 1 µg/ml elicited significantly more ingestive TR responses in rats having access to 1 µg/ml nicotine during the two-bottle tests than in control rats (0 µg/ml nicotine group; $p < .05$). Intraoral infusions of 1 µg/ml nicotine elicited the fewest number of ingestive TR responses in control rats. The number of ingestive TR responses elicited by 1 µg/ml nicotine in rats given access to 5 µg/ml was not significantly different from that of control rats.

5 µg/ml nicotine. All rats, with the exception of 1 control rat, ingested the entire intraoral infusion of 5 µg/ml nicotine. Groups differed, however, in the total number of ingestive TR responses elicited by intraoral infusions of 5 µg/ml, $F(2, 14) = 3.9$, $p < .05$. Rats given access to 1 µg/ml during the two-bottle test showed significantly more ingestive TR responses to intraoral infusions of 5 µg/ml nicotine than did control rats ($ps < .05$). There was no significant difference between the rats given two-bottle access to 0 µg/ml and 5 µg/ml nicotine.

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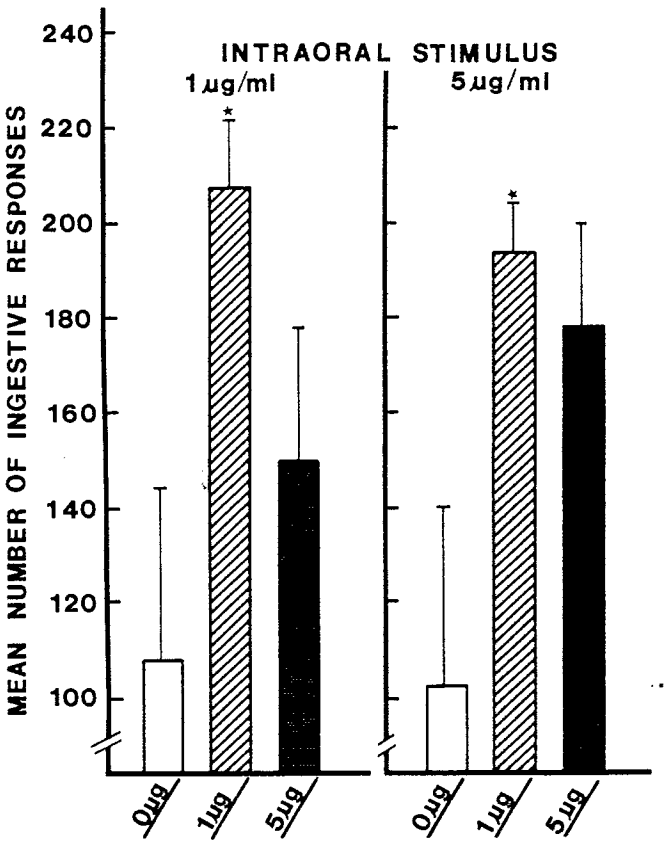

Figure 6. Mean (± *SE*) number of ingestive taste reactivity responses elicited by 0.8-ml intraoral infusions of 1 µg/ml nicotine and 5 µg/ml nicotine. (Histograms show the response of rats given access to 1 µg/ml nicotine, 5 µg/ml nicotine, and 0 µg/ml [water] during the two-bottle preference tests [Experiment 2].)

General Discussion

Previous reports indicate that nicotine is not preferred over water and is avoided at concentrations greater than 3 µg/ml (Naquira & Argueros, 1978). This was consistent with the results of Experiment 2 in which 5 µg/ml nicotine was avoided relative to water during two-bottle tests. Intake test results suggest that nicotine is an aversive taste stimulus. Analysis of the rats' TR responses elicited by nicotine, however, leads to the opposite conclusion. Nicotine was shown to elicit ingestive TR responses up to a concentration of 50 µg/ml. Only in response to 50 µg/ml and greater did nicotine elicit aversive TR responses. The comparison of intake and TR results suggests that postoral effects are responsible for the avoidance of nicotine at concentrations less than 50 µg/ml.

When the rats were given voluntary access to nicotine during the two-bottle test, the present data show that the rats initially ingest equal volumes of 1 µg/ml nicotine and water and consume only small amounts of 5 µg/ml nicotine. This pattern is consistent with those of previous reports (Naquira & Argueros, 1978). Over days, however, the preference ratios were found to increase for both 5 µg/ml nicotine and 1 µg/ml nicotine. The preference for 1 µg/ml nicotine exceeded that for water. This is the first demonstration, to our knowledge, that during voluntary intake tests, rats develop a preference for a nicotine solution.

As indicated above, the increased consumption of the nicotine may or may not reflect a change in its palatability. In order to address this issue, taste reactivity responses elicited by nicotine solutions were measured. Nicotine solutions (both 1 µg/ml and 5 µg/ml) elicited significantly more ingestive TR responses in rats showing the two-bottle preference for 1 µg/ml nicotine than in control rats. This indicates that a change in the rat's immediate response to nicotine accompanies the increase in the long-term preference for nicotine. Because control rats were given access to water during the two-bottle tests, one might argue that the fewer number of ingestive TR responses seen in the control group reflects a neophobic response to nicotine. This explanation is unlikely because control rats had been given multiple exposures to nicotine (during adaptation) prior to the taste reactivity test.

What accounts for the increase in preference and the number of ingestive TR responses elicited by nicotine solutions? Under conditions in which a "neutral" taste is paired with

positive postoral reinforcement, the preference for the taste increases (Fanselow & Birk, 1982; Mehiel & Bolles, 1984), as are ratings of the taste's pleasantness (Booth et al., 1982). An associative mechanism may similarly explain the increase in ingestive TR responses and preference for nicotine in rats given access to nicotine; that is, nicotine's palatability is enhanced because of its pairings with positive postoral reinforcement.

Exposure to low doses of injected nicotine once per day for 5 days or more causes an increase in the number of nicotinic receptor sites in the brains of rats (Ksir, Hakan, Hall, & Kellar, 1985). Thus, the nicotine-consuming rats in the present study may demonstrate altered behavioral reactions to nicotine because of similar increases in receptor number.

In summary, nicotine elicits ingestive TR responses over a range of concentrations, and only at higher concentrations (50 μg/ml and 100 μg/ml) are the oral stimulating properties of nicotine aversive. This contrasts with long-term intake test results in which nicotine is avoided at concentrations of 5 μg/ml and greater. A conclusion reached when comparing brief-access and long-term test results is that, in *naive* rats, factors other than taste can inhibit nicotine intake. Last, preferences for low concentrations of nicotine develop after long-term voluntary intake of nicotine by rats.

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Received December 23, 1987
Revision received March 29, 1988
Accepted April 11, 1988 ■