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The airway sensory impact of nicotine contributes to the conditioned reinforcing effects of individual puffs from cigarettes☆

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Abstract

Puffs from cigarettes are the fundamental unit of smoking reward. Here, we examined the extent to which reward from puffs can be derived from the airway sensory effect of nicotine, in the absence of a direct central nervous system effect of nicotine. We did this by assessing the self-reported reward obtained from individual puffs from nicotinized, denicotinized and unlit cigarettes within 7 s of inhalation, which is before nicotine had an opportunity to reach the brain. We also assessed the self-reported strength of airway sensations elicited by the puffs. We found that nicotinized puffs were rated as both stronger and more rewarding than denicotinized and unlit puffs. We also found that the extent to which nicotine elicited reward was directly correlated with the extent to which nicotine elicited airway sensations. This indicates that the airway sensory effects of nicotine contribute to the reward from puffs, above and beyond the reward derived from the airway sensory effects of non-nicotine constituents. These findings have implications for the interpretation of studies that use puffs as experimental units to examine nicotine reward. They also have implications for the use of denicotinized and low nicotine cigarettes as aids to smoking cessation.

Keywords: Cigarette smoking; Nicotine; Denicotinized cigarettes; Reward; Hedonic impact; Conditioned reinforcement; Airway sensation

1. Introduction

Cigarette smoking is a dependence behavior, characterized by compulsion to smoke despite awareness of its negative consequences (American Psychiatric Association, 2001). Much of the research on cigarette smoking dependence is predicated on the idea that smoking is subjectively pleasurable and behaviorally reinforcing (i.e. it is rewarding). The fundamental unit of reward from smoking is the puff, an act repeated more than a hundred times a day by a typical pack-per-day smoker. Individual puffs from cigarettes are subjectively experienced as pleasurable and desirable (Rose, 1984; Herskovic et al., 1986; Baldinger et al., 1995) and smokers will perform work to obtain them (Willner et al., 1995; Shahan et al., 1999; Perkins et al., 2002), indicating that individual puffs reproduce some of the rewarding characteristics of smoking whole cigarettes. For this reason, an increasing number of studies are using individual puffs as a way to address the psychological and neural processes that underlie smoking reward (e.g. Rose et al., 1985a,b; Herskovic et al., 1986; DeGrandpre et al., 1993; Bickel et al., 1997; Madden and Bickel, 1999; e.g. Shahan et al., 1999; Tidey et al., 1999; Perkins et al., 2002). Despite their popularity as experimental tools, the relative contributions of pharmacologic processes (i.e. direct central nervous system effects of nicotine) and sensory-motor processes (i.e. conditioned reinforcement) to the reward from individual puffs are not known.

In addition to delivering nicotine to the central nervous system, each puff from a cigarette stimulates an array of sensations within the airway that are transmitted to the

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central nervous system through peripheral nervous pathways. These sensations may be more important than the direct central nervous system effects of nicotine for smoking reward. For example, it has been shown that puffing on denicotinized cigarettes, which stimulates a component of the sensory impact of puffing without delivering an appreciable quantity of nicotine to the plasma, elicits feelings of exhilaration, euphoria and satisfaction, as well as reductions in urge, that are greater than those elicited by intravenous nicotine infusion (Westman et al., 1996; Rose et al., 2000). Conversely, blockade of airway sensations by anesthetizing the airway reduces smoking desirability and also the reduction in urge obtained from smoking (Rose et al., 1984a,b, 1985a,b). Thus, airway sensations are both necessary and sufficient to elicit smoking reward.

The reward derived from airway sensory stimulation ("sensory reward") is distinct from the reward derived from nicotine's direct central effects ("pharmacologic reward") in a number of ways. Firstly, sensory reward is elicited immediately following each puff from a cigarette, in the time it takes for airway sensation to be signaled in the brain. This makes individual puffs natural behavioral units for sensory reward, in a manner analogous to the way in which a bite of food is the natural unit of taste reward. In contrast, pharmacologic reward requires nicotine to be absorbed in the airway and transported to the central nervous system via the circulation, a process that has been estimated to take anywhere from 7 to 19 s (Russell and Feyerabend, 1978; Benowitz, 1990; Henningfield and Keenan, 1993). Furthermore, whereas pharmacologic reward is unlearned, or "primary" reward, sensory reward is learned. This learning has been proposed to be a form of conditioning, wherein airway sensations acquire reward value through repeated association with the pharmacologic reward obtained from nicotine (Rose and Levin, 1991). The role of learning in sensory reward is evidenced by the finding that the airway sensory effects of smoking are aversive to naïve smokers (Lee et al., 1993). Also, it has been shown that more heavily dependent smokers derive more reward from airway sensory stimulation than less heavily dependent smokers (Brauer et al., 2001), implying that this learning develops along with smoking dependence. Together, this suggests that sensory reward and pharmacologic reward, though both ultimately arising in the brain, may be mediated by psychological and neural processes that are at least partially distinct.

Experiments with denicotinized cigarettes demonstrate that airway stimulation by non-nicotine constituents is sufficient to elicit rewarding effects. However, a greater component of the airway sensory effects of a puff may actually be due to nicotine. Ginzel (1975) was the first to describe the sensory effects of nicotine within the pulmonary system and distinguish these from nicotine's pharmacologic (direct central nervous system and ganglionic) effects. More recently, Lee et al. (1993) have shown that inhalation of the nicotinic receptor antagonist hexamethonium reduces the airway irritation elicited by smoking. It has also been shown that the airway visceral afferent discharge elicited by a single breath of nicotinized cigarette smoke is more than 3 times greater than the discharge elicited by a breath of denicotinized smoke (Lee et al., 1989; Kou and Lee, 1990). This latter observation implies that nicotine is the primary agent in tobacco smoke responsible for airway sensory stimulation.

The airway sensations elicited by nicotine are known to be a source of reward. For example, Pritchard et al. (1996) found that the increase in satisfaction obtained by increasing cigarette nicotine content is related as much to the perceived sensory impact of the cigarettes as it is to the plasma nicotine level. In addition, Rose et al. (1999b) found that systemic blockade of peripheral nicotinic acetylcholine receptors using the ganglionic blocker trimethaphan reduces the satisfaction obtained from smoking. This manipulation also reduced the intensity of airway sensations, implying that nicotine's airway sensory effects contribute to reward from smoking.

While previous studies have established a role for the sensory effects of both nicotine and non-nicotine tobacco constituents in smoking reward, the relative contribution of airway sensory stimulation by nicotine and non-nicotine constituents to smoking reward is not known. This is necessary to know in order to understand exactly how denicotinized cigarettes are different from nicotinized cigarettes, which has implications for the use of denicotinized cigarettes both as aids to smoking cessation and as control stimuli in experimental studies of nicotine's pharmacologic effects. Also, no study has addressed nicotine sensory reward at the level of individual puffs. This is important because individual puffs are the natural behavioral units of airway sensory stimulation by smoking. This is also necessary for interpreting the results of studies that use individual puffs to address the psychological and neural processes underlying nicotine reward.

In this study, we sought to quantify the extent to which the airway sensory effects of the nicotine delivered by individual puffs give rise to reward, above and beyond the reward derived from the airway sensory effects of non-nicotine tobacco constituents. To isolate the airway sensory effects of nicotine from the direct central nervous system effects of nicotine, we assessed self-reported reward from individual puffs within 7 s of inhalation. This interval allowed airway sensations to be elicited by the nicotine in a puff, but did not allow the nicotine in a puff to reach the brain. We compared the reward from puffs from nicotinized cigarettes to the reward from puffs from denicotinized and unlit cigarettes. We also assessed the self-reported strength of airway sensations stimulated by puffs from each type of cigarette, which allowed us to correlate the reward elicited by nicotine with the airway sensations elicited by nicotine. All of the subjects in this study smoked less than one hour before testing. This was done in order to diminish the pharmacologic effects of nicotine. This also approximated

the usual conditions of smoking for a typical one pack-per-day smoker.

2. Method

2.1. Subjects

All procedures were approved by the University of Iowa Institutional Review Board for Human Subjects Research. Twenty cigarette smokers were recruited through advertisements in the university and local community. All reported smoking more than 20 cigarettes (1 pack) per day for at least 1 year. Subjects were screened by self-report to exclude any current medical, neurological or psychiatric disorders, including a history of dependence upon substances other than tobacco. Subject characteristics are described in Table 1.

2.2. Pre- and post-procedure assessments

Subjects were instructed to smoke ad libitum in the hours before the experiment and to smoke one of their own cigarettes just before they entered the facility, a university hospital. The experiment took place in a stainless steel chamber equipped with a high-performance ventilation system. Upon arriving at the facility, subjects gave informed consent, completed the Fagerstrom Test for Nicotine Dependence (FTND) (Heatherton et al., 1991) and reported the time of smoking the last cigarette and their usual brand of cigarette.

Heart rate, withdrawal symptoms and self-reported urge were measured both before and after the controlled smoking procedure in order to determine subjects' response to the pharmacological effects of the nicotine delivered during the procedure. Heart rate was measured over a two-minute interval at rest, with eyes closed. Subjects were administered the Minnesota Withdrawal Form (MWF) (Hughes, 1992) and the Brief Questionnaire of Smoking Urges (QSU-B) (Cox et al., 2001). Exhaled carbon dioxide (CO) was also measured before and after the procedure using a Micro-smokerlyzer (Bedfont, Kent, UK).

Table 1
Subject characteristics

	N	Mean	Range	S.D.
Females	9			
Males	11			
Menthol smokers	2			
Hand-rolled smokers	1			
Age		29	19–60	12.72
Cigarettes/day		25.5	20–40	7.05
Years smoking this much		16.93	1–38	12.49
Minutes since last cigarette		18.6	8–30	0.16
FTND score		5.45	2–9	2.06
Usual brand nicotine*		0.91	0.1–1.2	0.23
Usual brand tar*		12.24	5–16	3.39

* Hand-rolled cigarettes excluded from calculation of nicotine and tar content.

2.3. Stimuli

All cigarettes were Quest cigarettes (Vector Tobacco, Inc., New York, NY). Quest cigarettes are made from tobacco in which a key enzyme for nicotine synthesis (quinolinate phosphoribosyl transferase) has been inactivated through gene duplication. Varying nicotine levels are achieved by blending of modified and wild-type strains. This method of denicotinization leads to a nearly complete removal of nicotine and is presumed to spare more of the sensory properties of the non-nicotine constituents of tobacco smoke than chemical denicotinization. Nicotinized cigarettes were Quest 1: Low Nicotine (0.6 mg nicotine; 10 mg tar; FTC method). Denicotinized cigarettes were Quest 3: Nicotine Free (less than 0.06 mg nicotine; 10 mg tar; FTC method). Unlit cigarettes were either Quest 1 or Quest 3. All identifying marks were covered so that the cigarettes appeared identical. Cigarettes were handed to the subject by the experimenter, who sat directly across from the subject, separated by a small table. A screen obscured the view of the lit cigarettes, ashtrays and lighter.

2.4. Controlled smoking procedure

Eighteen individual puffs were presented in six blocks. Each block consisted of three trials, with each puff type (nicotinized, denicotinized and unlit) being presented once per block in pseudo-random order. The subject was not aware of the sequence. On each trial, the subject was handed a cigarette and instructed to hold the cigarette close to his or her face, without touching it to the lips. After 5 s, the subject was instructed to take a single puff from the cigarette, inhaling and exhaling as if smoking one of his or her own cigarettes. Four seconds after the instruction to puff, the subject was prompted to rate the puff as follows: "Rate pleasure" [rating], "Rate strength" [rating], "Rate desire for more" [rating]. A 4-s interval was chosen to allow the subjects time to inhale and exhale fully before rating the puffs. The onset of inhalation usually followed this instruction by 2–3 s, during which the subject brought the cigarette to the lips and drew smoke into his or her cheeks. Pleasantness ratings were nearly always completed within 7 s of the instruction to puff. This means that most pleasantness ratings actually occurred within 4–5 s following the onset of inhalation. Ten seconds after the desirability rating, the next cigarette was handed to the subject. Subjects took two practice puffs from unlit cigarettes before the beginning of data collection, in order to gain familiarity with the procedure. New cigarettes were lit after every 6 trials, such that no more than two puffs were taken from any one cigarette and the char lines never reached more than 2/3 of the way to the filter. During lighting of the new cigarettes,

subjects were allowed to sip water and to communicate with the experimenter.

2.5. Self-report ratings of puffs

Ratings were made verbally using 7-point (1–7), Likert-type scales. Before the procedure, subjects were instructed on the use of the rating scales, which were displayed before the subject throughout the procedure. The first item was "How pleasant was the puff?" Subjects were instructed that this referred to how much they liked the puff, or found it pleasurable. The second item was "How strong was the puff?" Subjects were instructed that this item referred to the strength of the sensations in the throat and chest caused by the puff. The third item was "How much do you desire another puff from this cigarette?" Subjects were instructed that this referred specifically to the desire for another puff from the cigarette from which a puff had just been taken, and not a desire for puffs from other cigarettes or and overall urge to smoke. Subjects were instructed that they would be prompted to rate the 3 items after each puff and were told to refer to the scales as necessary throughout the procedure.

2.6. Heart rate

EKG was measured using the standard configuration for lead II. Signals were acquired at 1 KHz. The transducers, amplifiers, analog to digital converter (MP100) and data acquisition and analysis software were all from Biopac, Inc. (Santa Barbara, CA). The mean heart rate was measured pre- and post-procedure over the two-minute interval during which subjects rested with eyes closed. Heart rate for each beat was measured using a peak detection algorithm implemented within the Biopac software. The mean heart rate was the average of the series of individual heart beat measurements.

2.7. Data analysis

Responses from the first block were excluded in order to reduce novelty effects. Only trials on which pleasantness ratings were completed within 7 s of the instruction to puff were included. The remaining puff-related responses were averaged within each subject for each puff type. The dependent measures for each puff type were thus: mean pleasantness rating (pleasantness), mean desirability rating (desirability) and mean strength rating (strength).

Repeated measures ANOVA was used to examine the overall effect of puff type on each of the dependent measures. The degrees of freedom were Hyunh–Feldt corrected as deemed necessary by Mauchly's test of sphericity. Post-hoc *t*-tests were used to compare puff types pair-wise. The critical *t*-values (2-tailed) for significant effects were adjusted for multiple comparisons using Tukey's correction. This led to a critical $t(19)=3.59$ for $p<0.05$ and $t(19)=4.67$ for $p<0.01$, which applied to all comparisons made between puff types. In addition, an effect size estimate (d) was calculated for each comparison, taking into account the correlation between dependent variables due to a repeated measures design (Dunlap et al., 1996).

For each subject, the difference in the mean rating for nicotinized puffs and the mean rating for denicotinized puffs was calculated for the pleasantness, desirability and strength reports. This provided, for each subject, measures of the degree to which nicotinized puffs were reported as stronger, more pleasant and more desirable than denicotinized puffs, respectively. The difference scores for pleasantness and desirability were respectively correlated with the difference score for strength using Pearson product moment correlation.

3. Results

Fig. 1 shows the mean self-report responses for each puff type. The ANOVA revealed a main effect of puff type on ratings of pleasantness [$F(2,38)=57.42$, $p<0.0001$], desirability [$F(2,39)=43.19$, $p<0.0001$] and strength [$F(1.5,29.6)=61.83$, $p<0.0001$, Hyunh–Feldt corrected]. Post-hoc *t*-tests revealed that, compared to denicotinized puffs, nicotinized puffs were rated as more pleasant ($t=5.06$, $p<0.01$, Tukey-corrected), more desirable ($t=5.24$, $p<0.01$, Tukey-corrected) and stronger ($t=5.70$, $p<0.01$, Tukey-corrected). Pleasantness, desirability and strength were all greater for both denicotinized and nicotinized puffs than they were for unlit puffs (all t's >5.0, all p's <0.01, Tukey-corrected). Table 2 shows the effect size calculations for each of these comparisons.

As shown in Fig. 2, the degree to which smokers reported nicotinized puffs as stronger than denicotinized puffs was significantly positively correlated with the degree to which smokers found nicotinized puffs more pleasurable than denicotinized puffs ($r=0.70$, $p=0.001$) and the degree to which they found nicotinized puffs more desirable than denicotinized puffs ($r=0.71$, $p=0.001$).

There were no pre- to post-procedure changes in heart rate ($t=0.79$, $p=0.44$), MWF score ($t=0.5$, $p=0.31$) or QSU-B score ($t=0.79$, $p=0.44$), indicating that, as a group, subjects' responsiveness to nicotine's pharmacological effects was diminished. Exhaled CO increased significantly from pre- to post procedure ($t=2.61$, $p=0.02$). These data are summarized in Table 3.

4. Discussion

In this study we show that individual puffs from nicotinized cigarettes are subjectively appreciated as being more pleasurable and desirable and as eliciting stronger airway sensations than puffs from denicotinized cigarettes when these are rated immediately after inhalation. We find this in the context of a diminished pharmacological effect of

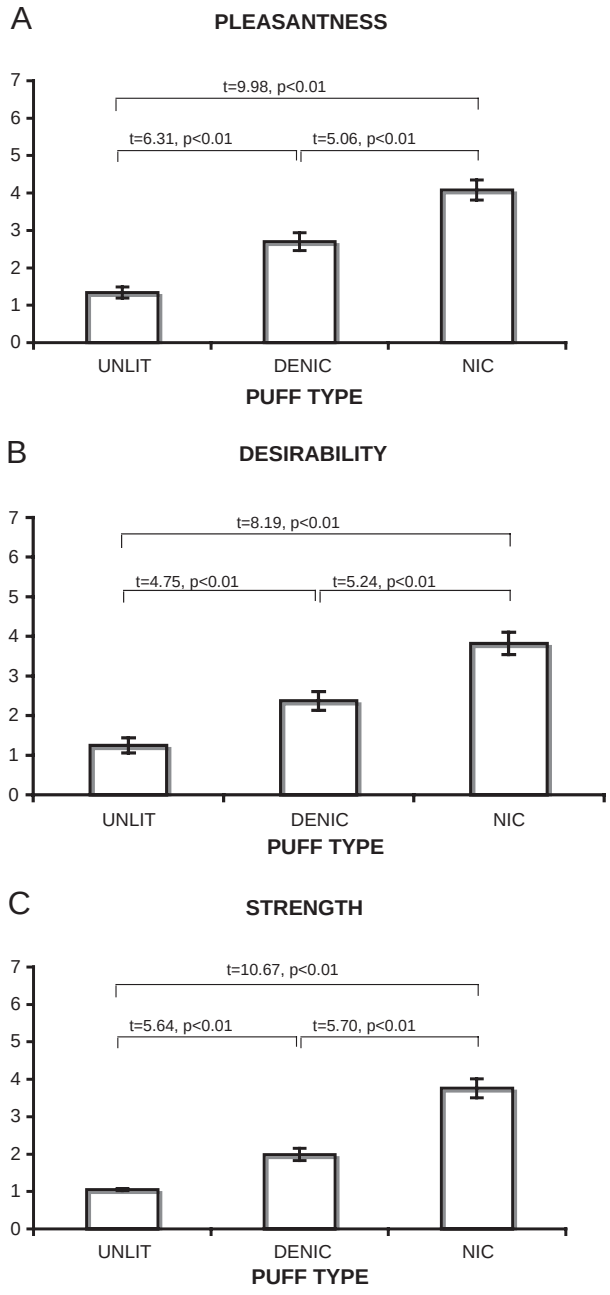

Fig. 1. Self-report pleasantness (A), desirability (B) and strength (C) of unlit, denicotinized and nicotinized puffs. Error bars represent 1 standard error of the mean. All *t*-tests are 2-tailed and all *p*-values are Tukey-corrected for multiple comparisons.

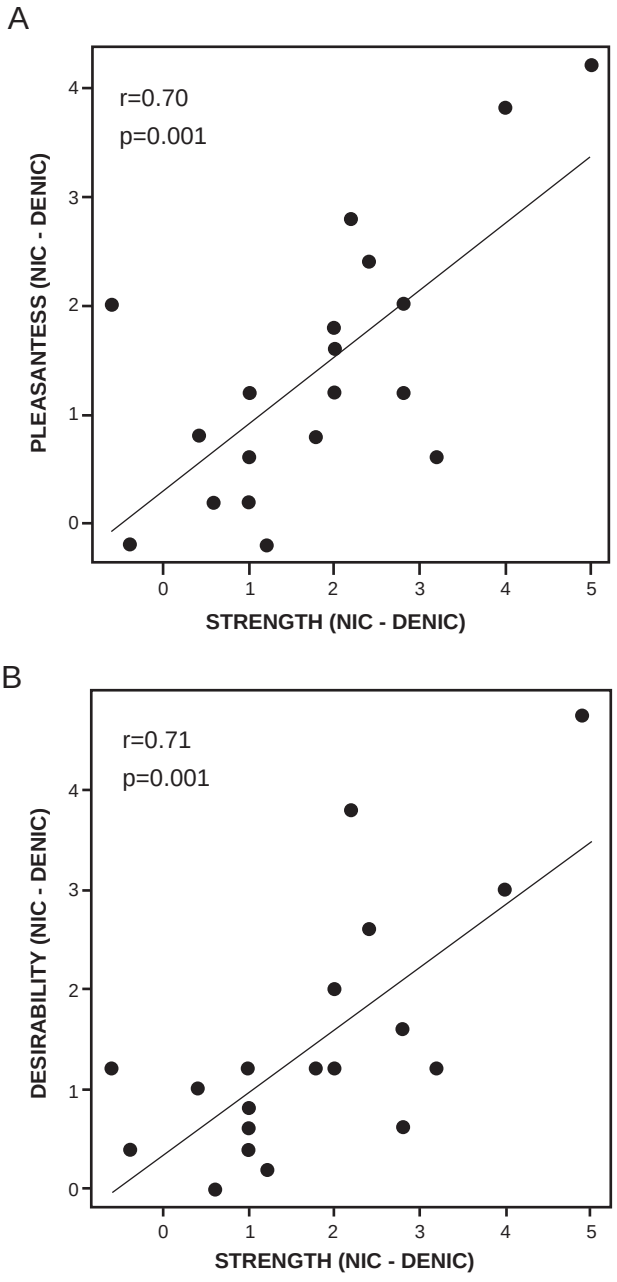

Fig. 2. Correlation of the difference in strength between nicotinized and denicotinized puffs and (A) the difference in pleasantness between nicotinized and denicotinized puffs and (B) the difference in desirability between nicotinized and denicotinized puffs. Points represent difference scores for individual subjects.

nicotine, as shown by a failure of the procedure as a whole to increase heart rate and decrease withdrawal symptoms or smoking urges. Furthermore, we show that the extent to which nicotinized puffs are rated as more pleasant and more desirable than denicotinized puffs are respectively correlated with the extent to which nicotinized puffs are rated as eliciting stronger airway sensations than denicotinized puffs.

Table 2
Effect size (*d*) estimates comparing different puff types

	Unlit vs. Denic	Unlit vs. Nic	Denic vs. Nic
Pleasantness	1.49	2.85	1.24
Desirablity	1.18	2.45	1.27
Strength	1.75	3.23	1.88

Previous studies (Herskovic et al., 1986; Baldinger et al., 1995; Perkins et al., 1996, 2002; Shahan et al., 1999) have shown that increasing the nicotine content of individual puffs increases their reward value, but these studies did not make an effort to distinguish between the reward derived from nicotine's airway sensory effects and the reward

Table 3
Changes from pre- to post-procedure

	Pre-procedure	Post-procedure	SD of change	t	p
Heart rate	81.63	81.16	4.85	0.79	0.44
QSU-B score	12.20	12.90	8.62	0.79	0.44
MWF score	2.90	3.50	2.19	1.05	0.31
Exhaled CO	22.40	28.75	4.00	2.61	0.02

derived from nicotine's direct central and ganglionic effects. The present results provide direct evidence that nicotine delivered by individual puffs can give rise to reward through airway sensory effects alone. These results are consistent with previous findings showing that nicotine exerts sensory effects upon the airway (Ginzel, 1975; Lee et al., 1993) and with findings that have provided correlative evidence that nicotine's airway sensory effects contribute to smoking reward (Pritchard et al., 1996; Rose et al., 1999a,b). This study provides new information by completely isolating nicotine sensory reward from nicotine pharmacologic reward. This allowed for a quantification of the relative contributions of nicotine and non-nicotine sensory stimulation to smoking reward at the level of individual puffs.

Subjects were asked to rate the pleasantness of puffs beginning 4 s after the instruction to puff, and always completed this rating within 7 s of inhalation. Seven seconds is the minimum estimate of the time required for nicotine to reach the brain following inhalation (Benowitz, 1990). This estimate is based upon the lung–brain transit time of inhalation anesthetics (Mapleson, 1973). Unlike inhalation anesthetics, which are lipophilic, nicotine is positively charged at the pH of cigarette smoke, and is therefore likely to be exchanged more slowly across the alveolar surface. Indeed, the absorption kinetics of nicotine have been found to be significantly slower than previously assumed (Rose et al., 1999a,b). Studies in rodents (Brewer et al., 2004) indicate that the delivery of nicotine to the brain may be slowed significantly by binding within the lung. This means that 7 s may actually be an underestimation of the minimum inhalation–brain delay for nicotine. This makes it extremely likely that, in the present study, pleasantness ratings were always completed before nicotine had a chance to bind to receptors within the brain, which would mean that differences in pleasantness ratings between nicotinized and denicotinized puffs cannot be attributed to nicotine's direct central nervous system effects.

Because ratings of desirability were usually made after 7 s following inhalation, it is possible that these ratings were influenced by nicotine's direct central effects. However, if this were the case, then the effect size for the difference between nicotinized and denicotinized puffs should have been greater for ratings of desirability than for ratings of pleasantness, since this delay would have allowed time for nicotine pharmacologic reward to add onto nicotine sensory reward. However, these effect sizes differed by only a very small amount. One possibility is that ratings of desirability, like ratings of pleasantness, also occurred before nicotine reached the brain. The maximum estimate of the inhalation–brain transit time for nicotine has been reported to be 19 s (Henningfield and Keenan, 1993). Though the interval between inhalation and desirability rating was not measured in the present study, this interval is likely to have been less than 19 s for a large proportion of the trials.

Another possibility is that the dose of nicotine delivered to the brain by each puff was too low to exert an effect on any of the self-report measures, regardless of the inhalation-rating interval. It has been shown that intravenous nicotine delivered in doses obtained from smoking whole cigarettes does not lead to a subjectively appreciable acute reward above that obtained from smoking denicotinized cigarettes (Rose et al., 2000). Also, a recent meta-analysis of studies examining nicotine self-administration in humans (Dar and Frenk, 2004) has shown that dependent smokers do not self-administer nicotine in the absence of the sensory-motor process of smoking. Together, these findings suggest that the pharmacologic effects of nicotine, even in doses delivered by whole cigarettes, is not a source of acute reward from smoking. Though controversial (see Perkins, 2004), these findings would imply that the nicotine delivered to the brain by individual puffs from nicotinized cigarettes would not be enough to elicit a subjectively rewarding effect above the reward obtained from the airway sensory effects of non-nicotine constituents. This would suggest that differences in reward value between nicotinized and denicotinized puffs are due primarily to the airway sensory impact of nicotine. However, this interpretation requires some caution, since the precise contribution of nicotine's pharmacologic effects to the reward from individual puffs is not known.

In the present study, the ability of nicotine's pharmacologic effects to contribute to the reward from individual puffs was further diminished by acute tolerance to the pharmacologic effects of nicotine. Subjects possessed a diminished response to nicotine's pharmacologic effects, as indicated by a lack of effect of the procedure on heart rate, withdrawal symptoms or urge to smoke. If the procedure as a whole, which delivered 6 puffs from nicotinized cigarettes, did not exert a significant pharmacologic effect, then individual puffs from nicotinized cigarettes were unlikely to have exerted a pharmacologic effect, including pharmacologic reward. Acute tolerance to nicotine's pharmacologic effects has been shown to last for approximately 2 h after nicotine exposure (Perkins et al., 1995). This means that for a typical one pack-per-day smoker, who smokes on average every hour, most cigarettes are smoked under conditions of acute tolerance to nicotine's pharmacologic effects. For them, like for the subjects in the present study, the majority of reward obtained from nicotine may actually be due to its airway sensory effects, rather than to its pharmacologic effects.

Despite tolerance to nicotine's pharmacologic effects, subjects were still able to appreciate and derive reward from nicotine's airway sensory effects. This suggests that the

airway sensory effects of nicotine are less susceptible to acute tolerance than the pharmacologic effects of nicotine are. One explanation for this may be that, unlike nicotine's pharmacologic effects, nicotine's airway sensory effects are not mediated by nicotinic acetylcholine receptors; rapid desensitization of nicotinic acetylcholine receptors may underlie acute tolerance, so a non-receptor-mediated process could potentially circumvent acute tolerance. This is unlikely, however, since the airway expresses an abundance of nicotinic acetylcholine receptors (Wang et al., 2001; Keiger et al., 2003; Proskocil et al., 2004) and it has been shown that blockade of nicotinic acetylcholine receptors significantly reduces the airway sensory effects of smoking (Lee et al., 1993; Rose et al., 1999a,b). An alternative explanation is that the airway sensations stimulated by nicotine may be transduced by an isoform of the nicotinic acetylcholine receptor that is not susceptible to rapid desensitization, or does not undergo as complete a desensitization as the receptors that mediate nicotine's pharmacologic effects.

Though the primary difference between nicotinized and denicotinized cigarettes in the present study was assumed to be the airway sensory impact of nicotine, these may have also differed with respect to some non-nicotine sensory factor. This may have occurred either as a result of the genetic denicotinization process or, more likely, through the manipulation of the denicotinized tobacco by the manufacturer in order to increase its sensory impact. Tobacco industry documents (Bates, 1983, 1995) show that cigarette manufacturers have a long history of manipulating the pH of mainstream tobacco smoke in order to modify its sensory impact. One way that pH may affect sensory impact is by modulating the absorption of nicotine; a higher pH of mainstream smoke has been shown to lead to a greater absorption of nicotine in the airway (Armitage and Turner, 1970). In this case, however, increasing pH would only increase the sensory impact of nicotinized cigarettes. Another possibility is that pH acts upon sensory impact of non-nicotine constituents independently of its effects on nicotine absorption. However, evidence for such effects is lacking. Apart from pH, it is possible that any number of non-nicotine constituents may have been introduced into the tobacco that may have modulated the sensory impact of the denicotinized and nicotinized cigarettes used in the present study. The extent of such manipulations cannot be known without a full chemical analysis of the mainstream smoke of each of the cigarettes, which is beyond the scope of this study. Though the potential effects of such manipulations are important to consider in any study that compares nicotinized and denicotinized cigarettes, they are likely to be small in relation to the airway sensory effects of nicotine.

In this study, the estimates of effect size allowed for a quantitative comparison between the airway sensory impact of nicotine and the airway sensory impact of non-nicotine tobacco constituents. For each of the self-report measures, the effect size of the difference between nicotinized puffs and denicotinized puffs was approximately equal the effect size of the difference between unlit puffs and denicotinized puffs. This indicates that, for cigarettes with a nicotine content of 0.6 mg and a tar content of 10 mg, the airway sensory impact of nicotine is approximately equal to the airway sensory impact of non-nicotine constituents. This also indicates that the reward derived from the airway sensory impact of nicotine is approximately equal to the reward derived from the combined effects of the airway sensory impact of non-nicotine constituents and the motor act of puffing. The nicotinized cigarettes used in this study, which may be considered "light", have a relatively low nicotine/tar ratio: 0.06. For full-flavored cigarettes, which have higher nicotine/tar ratios, the proportion of sensory impact that can be explained by nicotine would be even higher.

This means that any study that quantifies the reward obtained from the nicotine in individual puffs will include a significant component of reward derived from airway sensation. This includes studies that use puffs from denicotinized cigarettes as "control" stimuli. For instance, Shahan et al. (1999) have shown that subjects will perform more work to obtain puffs from nicotinized cigarettes than they will to obtain puffs from denicotinized cigarettes. If one assumes that denicotinized cigarettes control completely for the sensory-motor process of smoking, then this result can be taken as evidence that reward is derived from the pharmacological effects of nicotine. However, if, as the present results suggests, a significant proportion of the reward from individual puffs can be attributable to the sensory impact of nicotine, the difference in reinforcement value between nicotinized and denicotinized puffs may be due to differences in sensory impact. Perkins et al. (2002) used a similar reinforcement paradigm to address sex differences in the reward from nicotine. In this study, it was found that the reinforcement value of puffs is more sensitive to nicotine content in males than in females. This was taken as evidence supporting the hypothesis that males are more sensitive than females to the primary reinforcing effects of nicotine, in contrast to females, who are more sensitive to conditioned reinforcement from non-nicotine stimuli (reviewed in Perkins et al., 1999). The present results indicate that nicotine in puffs is in itself is a source of conditioned reinforcement. This points to the possibility that males may actually be more sensitive to the conditioned reinforcement derived from the airway sensory effects of smoking than females are. A number of studies have also shown that smokers' preference for the nicotine content of individual puffs increases as a result of nicotine deprivation (Rose et al., 1984a,b; Herskovic et al., 1986; Perkins et al., 1996; Madden and Bickel, 1999). The present results suggest that what is modulated by deprivation is the reward derived from the airway sensory effects of nicotine. This is similar to the way in which hunger modulates the reward obtained from the sensory impact of food, a phenomenon known as alliesthesia (Cabanac, 1971). Because they are temporally

discrete units of smoking reward, individual puffs may also soon be used in combination with event-related functional imaging techniques, such as functional magnetic resonance imaging (fMRI), to address the neural substrates of nicotine reward. The present results imply that any study that compares the neural response to nicotinized puffs to the neural response to denicotinized puffs will reveal neural correlates of reward derived from nicotine's airway sensory effects. Indeed, the methods presented in the present study are ideally suited to address the neural substrates of sensory reward from smoking.

A clinical implication of these results is that smokers who switch to low nicotine and denicotinized cigarettes as a way to wean themselves off of nicotine will find that they receive less reward from each puff than they are used to, due to a reduced sensory impact. This may reduce the efficacy of this smoking cessation strategy. It has been suggested that switching to reduced nicotine cigarettes may be more acceptable to smokers if their sensory impact could be somehow increased (Rose and Behm, 2004). These manipulations, which may include increasing the pH of the denicotinized tobacco smoke, would increase the efficacy of denicotinized cigarettes for reducing smoking urges and would also increase compliance, both of which would tend to reduce the rate of relapse to smoking the usual brand.

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