Smoking Cues Decrease Prepulse Inhibition of the Startle Response and Increase Subjective Craving in Humans

Kent E. Hutchison
University of Colorado at Boulder

Raymond Niaura
Miriam Hospital

Robert Swift
Brown University and Veterans Affairs Medical Center, Providence

The present study investigated whether exposure to smoking cues would attenuate prepulse inhibition (PPI) of the startle reflex and increase craving among smokers across 2 experimental sessions. It was hypothesized that exposure to smoking cues would result in a decrease in PPI. Twenty-six smokers were exposed to smoking cues and control cues in 2 experimental sessions 1 week apart. Results indicate that smoking cues reliably attenuated PPI in both the 1st and 2nd sessions as compared with control cues. Findings also suggest that smoking cues reliably increased craving, increased negative affect, and reduced positive affect relative to baseline measures in both sessions. Results are consistent with the premise that exposure to smoking cues precipitates increases in dopamine activation or changes in information processing that cause a disruption of PPI.

Smoking remains the leading preventable cause of death in the United States, causing as many as 419,000 deaths a year. Despite recent advances in the treatment of nicotine dependence, such as nicotine replacement therapy, long-term abstinence rates are only 10%-30% (e.g., Rose, 1996). Basic research on the mechanisms that mediate nicotine dependence, and drug dependence in general, has been instrumental in the development of new prevention and treatment efforts. Preclinical studies have demonstrated that one of the biological mechanisms that contribute to the addictive liability of nicotine and other drugs is the generation of psychomotor stimulation and positive reinforcement through intensification of mesolimbic dopamine (DA) activity (see Robinson & Berridge, 1993; Wise & Bozarth, 1987). Nicotine as well as alcohol, cocaine, amphetamines, and opiates are known to activate mesolimbic DA (Balfour, 1994; Bozarth, 1994; Di Chiara & North, 1992; Koob, 1992; Pich et al., 1997; Pontieri, Tanda, Orzi, & Di Chiara, 1996; Woolverton & Johnson, 1992). Furthermore, it has been suggested that the motivational and appetitive properties of drug-seeking behavior are enhanced as the dopaminergic pathways become sensitized through repeated use (Robinson & Berridge, 1993).

Activation of dopaminergic pathways is moderated by conditioned stimuli in the environment that have been previously paired with drug consumption, such that these stimuli enhance the incentive, motivational properties of drug use behavior (Robinson & Berridge, 1993; Stewart, 1992; Wise, 1988). Several preclinical studies have shown that exposure to drug-related stimuli in the absence of the drug produces psychomotor stimulation and DA activity (for reviews, see Robinson & Berridge, 1993; Stewart, 1992). For example, exposure to alcohol cues during a 15-min waiting period before self-administration significantly increased DA activity in the nucleus accumbens, which was interpreted as evidence that prior learning and anticipation of reinforcement activates a DA response (Weiss, Lorang, Bloom, & Koob, 1993). These observations are also consistent with the conditioned appetitive model of drug use, which suggests that conditioned stimuli that are repeatedly paired with drug administration become associated with the appetitive and rewarding aspects of the drug, such that these cues or small doses of the drug itself are able to prime the DA system (Stewart, De Wit, & Eikelboom, 1984).

Clinical research has also demonstrated that exposure to conditioned stimuli associated with drug use lead to conditioned appetitive responses and changes in information processing. There is extensive research documenting that exposure to drug-related stimuli increases craving for a variety of drugs (for reviews, see Glautier & Remington, 1995; Niaura et al., 1998). Likewise, exposure to a variety of interoceptive and exteroceptive smoking cues reliably elicits conditioned appetitive responses, including increases in craving and changes in physiological measures, such as heart rate and blood pressure (e.g., Niaura et al., 1998).
Studies have demonstrated that reactivity to drug-related stimuli predicts relapse among alcoholics and smokers, suggesting that reactions to conditioned stimuli are clinically relevant (e.g., Niaura et al., 1988; Rohsenow et al., 1994).

Recently, researchers have applied the startle reflex paradigm to investigate the affective processes that underlie the effects of alcohol and drugs in humans (Elash, Tiffany, & Vrana, 1995; Patrick, Berthot, & Moore, 1996; Stritzke, Patrick, & Lang, 1995). The startle reflex is a defensive response, such as an eyelink, to a startling stimulus, such as a loud noise, and is influenced by a number of well-documented physiological and psychological factors (for reviews, see Davis, Falls, Campeau, & Kim, 1993; Lang, 1995). One aspect of the startle reflex that has important implications for research on alcohol and drug abuse is prepulse inhibition (PPI) of the startle reflex. PPI of the startle reflex refers to the reflex-suppressing effect of a nonstartling lead stimulus that precedes the startle-eliciting stimulus (for reviews, see Graham, 1992; Graham & Hackley, 1991). The putative function of this mechanism is to protect the information processing of the prepulse from interruption by the startle-eliciting stimulus (Graham, 1992; Norris & Blumenthal, 1996). Although PPI is predominantly a measure of involuntary, automatic information processing, a few researchers have shown that PPI may be increased slightly by instructing participants to attend to the pulses (e.g., Filion, Dawson, & Schell, 1993). More important, the neural circuitry that mediates the effect of a prepulse on the startle reflex overlaps considerably with the mesolimbic circuitry that mediates the effects of drugs and drug cues. Although this overlap is complex and involves several neurotransmitter systems, pharmacological manipulations that facilitate mesolimbic DA activity clearly decrease PPI (Ott & Mandel, 1995; Swerdlow, Braff, Taaid, & Geyer, 1994; Swerdlow, Caine, Braff, & Geyer, 1992; Swerdlow et al., 1990; Swerdlow, Vaccarino, Amalric, & Koob, 1986; Zhang, Engel, Hjorth, & Svensson, 1995).

Recently, PPI has been applied to investigations of alcohol and drugs in humans. For example, consumption of d-amphetamine attenuated PPI and increased subjective stimulation in humans 90 min after consumption (Hutchison & Swift, 1999). With respect to alcohol, results of one study showed that alcohol did not affect PPI (Grillon, Sinha, & O'Malley, 1994), whereas another study showed that the effects of alcohol on PPI were moderated by baseline PPI (Hutchison, Rohsenow, Monti, Palfai, & Swift, 1997). Investigations of the effects of nicotine have produced some contradictory findings. One study showed that smoking increases PPI under limited conditions (Kumari, Checkley, & Gray, 1996). However, another study demonstrated that controlled smoking of cigarettes with a high dose of nicotine immediately attenuated PPI compared with controlled smoking of cigarettes with a low dose of nicotine (Hutchison, Niaura, & Swift, 1999). Although we know of no reports of the effects of smoking cues on PPI, one study showed that imaginal exposure to smoking cues enhanced the magnitude of the startle response, particularly in the context of positive emotional stimuli (Elash et al., 1995).

The objective of the present research was to determine whether exposure to smoking cues would precipitate an attenuation of PPI and increase in craving and whether this effect would be reliable and robust enough to be replicated in a second session 1 week later. On the basis of the preclinical literature suggesting that exposure to drug cues produces mesolimbic DA activation, we hypothesized that exposure to smoking cues would result in a decrease in PPI. We also expected reliable increases in craving and negative affect after exposure to smoking cues across sessions, replicating previous studies on smoking cue reactivity. Results that support reliable decreases in PPI and increases in craving over multiple sessions would suggest that PPI may be useful as a pre- to posttreatment outcome indicator.

Method

Participants

This study was approved by the Brown University Institutional Review Board (IRB) and the Roger William’s Medical Center IRB. Participants were recruited from the greater Providence, Rhode Island, community and gave their written informed consent before participating. Participants were excluded if they had any history of cardiac illness, reported any hearing loss, or were taking any psychotropic medications or illicit drugs. Participants received $50 for completing the study.

Of the 36 participants who completed the screening process, 4 did not show up for the experimental sessions and 2 were excluded when it was revealed that they were taking psychotropic medications. Of the remaining 30 participants, 1 did not comply with the study instructions and 3 failed to bring their own cigarettes (required for the cue exposure procedure) to the experimental sessions. Thus, the final sample included 26 participants (14 men and 12 women) who were primarily Caucasian (23 Caucasian, 2 African American, and 1 Asian). The mean age of the sample was 32 years (ranging from 18 to 47; SD = 8.8), and the mean number of years of education was 13.6 (SD = 1.6). The average Fagerstrom Tolerance Questionnaire (Fagerstrom & Schneider, 1989) score for the sample was 6.9 (SD = 2.2). The participants had been smoking for an average of 13.8 years (SD = 2.7) and smoked an average of 20.6 cigarettes per day (SD = 7.2). The average expired CO reading at baseline was 15.5 ppm (SD = 8.8) for the first session and 15.1 ppm (SD = 9.1) for the second session.

Procedure

Participants were scheduled for two experimental sessions that were 1 week apart. Participants were exposed to both the control cues and the smoking cues in each session, such that they were exposed to the control cues and smoking cues twice. Participants were instructed not to smoke after midnight so that they would be deprived for at least 8 hr before arriving at the laboratory on the morning of each experimental session. Participants completed baseline demographic and smoking history measures at the beginning of the first session. In each of the experimental sessions, participants were seated at a desk and instructed to relax. Participants were then told that they would hear a series of loud and abrupt noises over the headphones. The participants were instructed to ignore the noises and to look straight ahead at the wall in front of them. A 5-min baseline block of startle trials was conducted at the beginning of each session and consisted of six pulse-alone and six prepulse trials (see the description of pulse-alone and
prepulse trials below). The purpose of the baseline block of startle trials was to allow the participants to become accustomed to the startle probe procedures.

After the baseline block of startle trials was completed, the participants completed baseline measures of craving and affect (see the description that follows). Participants were then given another relaxation period, after which they completed measures of craving and affect. Participants were then exposed to either control cues or smoking cues. The order of presentation of the control cues and smoking cues was counterbalanced such that half the participants received control cues first and the other half received smoking cues first. The order of the presentation was the same for each experimental session for each participant. The exposure to control cues consisted of instructing the participant to hold a pencil with his or her smoking hand and focus attention on the pencil. During this period (190 s), participants received a block of four prepulse and four pulse-alone startle trials. Immediately after the startle trials, participants completed measures of craving and affect. Following the cue exposure procedures outlined by Sayette and Hufford (1994), the smoking cue condition consisted of instructing the participants to remove one of their own cigarettes from the pack and light it without putting it in their mouths by holding it in the flame for several seconds. After lighting the cigarette, participants were asked to focus on the cigarette. Participants also received a 190-s block of startle trials during the smoking cue exposure and completed measures of craving and affect immediately after the startle trials.

**Measures**

**Craving measure.** The craving measure consisted of five items that were rated on a scale of 0–100 and that were combined to form a craving scale (Shiffman et al., 1998). The five items were “I crave a cigarette right now,” “I have an urge for a cigarette,” “I have a desire for a cigarette right now,” “If it were possible, I would smoke now,” and “All I want right now is a cigarette.” Cronbach’s alpha for the scale was .97, suggesting good internal consistency.

**Positive Affect/Negative Affect Scale (PANAS).** The PANAS is a 20-item measure with subscales measuring positive affect and negative affect. The PANAS is a reliable and valid measure of both positive and negative affect with alphas of .84–.90 (Watson, Clark, & Tellegen, 1988).

**Startle and cardiac reactivity.** The Biopac Systems Model MP-100 (Biopac Systems, Inc., Santa Barbara, CA) was used to record heart rate and elicit and record the startle response during the exposure procedures. Each block of startle trials consisted of four prepulse and four pulse-alone trials. Each prepulse and pulse-alone trial was distributed such that one prepulse and one pulse-alone trial occurred every 50 s with at least 20 s in between trials with random, low-intensity background noise in between trials. The order of the prepulse trials and pulse-alone trials was varied in a predetermined, pseudorandom order such that participants would be unable to anticipate the next trial. A prepulse trial consisted of a tone (frequency = 800 Hz, intensity = 58 dB, duration = 30 ms, rise time = 3 ms) that was presented 120 ms before the startle-eliciting stimulus (white noise with instantaneous rise time, intensity = 105 dB, duration = 50 ms). A pulse-alone trial consisted of only the startle-eliciting stimulus.

The startle reflex was recorded with two 4-mm surface electrodes that were placed on the surface of the skin over the orbicularis oculi of the left eye 20 mm apart (i.e., just below the eye in the orbital region). A ground electrode was placed on the forehead. The electromyographic (EMG) signal of this muscle was sampled at 1000 Hz before and after the startle-eliciting probes and was filtered with a bandwidth of 28–500 Hz. This particular bandwidth was chosen because a 28–512 Hz bandwidth provides the optimal balance for filtration of low-frequency artifacts and signal retention (VanBoxtel, Boelhouwer, & Bos, 1998). The signal was rectified digitally off-line but was not integrated because, in general, the raw signal and integrated signal are highly correlated (Blumenthal, 1996) and because the process of integration attenuates the raw signal. The primary variable of interest in this study was the percentage of PPI, which was calculated as the difference of the average startle response magnitude on pulse-alone trials minus the magnitude of the average corresponding prepulse trials, divided by the magnitude on the pulse-alone trials (i.e., \%PPI = [pulse – prepulse] / pulse · 100). Two secondary variables were the magnitude of the startle response on pulse-alone trials and the latency to peak response on pulse-alone trials. Latency to peak was used in this study because it is more easily scored than latency to onset. Latency to onset of the startle response would not have provided unique information and could not have been scored using change of slope algorithms given an unintegrated EMG signal. Heart rate was estimated as the average number of R-waves per minute in each of the 5-min blocks. The electrocardiographic signal was recorded with standard disposable electrodes placed on either side of the heart.

**Design and Analysis**

A 2 × 2 within-subjects design was used in this study. The two independent variables included cue (control cue vs. smoking cue) and time (Session 1 vs. Session 2). Before analysis, the distributions of all of the dependent variables were checked to determine whether transformations would be necessary. All the variables used in the analyses reported here were normally distributed. Because it is possible that the order of presentation (e.g., smoking cues first vs. control cues first) may confound the analysis and interpretation of results in within-subjects designs (e.g., Keppel, 1982), we conducted a 2 × 2 × 2 (Order × Cue × Session) analysis of variance (ANOVA) for each dependent variable before testing the hypotheses. Unless otherwise indicated, no significant effects involving order were found, and hypotheses concerning the effects of smoking cues and session were tested with 2 (cue) × 2 (session) repeated measures ANOVAs.

**Results**

**Smoking Cues and Psychophysiological Reactivity**

The startle data from 1 participant were not included in this set of analyses because the participant did not exhibit a measurable startle response across trials. For the remaining 25 participants, a 2 × 2 repeated measures ANOVA on the primary dependent variable, PPI, revealed a significant main effect for cue, F(1, 24) = 4.89, p < .05, indicating that smoking cues attenuated PPI. There was no main effect for session or a significant interaction (p > .10). A second 2 × 2 repeated measures ANOVA was conducted on the magnitude of the startle response and did not show any significant main or interaction effects (p > .10). Repeated measures ANOVAs on the latency to peak response and on heart rate also did not suggest any main or interaction effects (p > .10). Figure IA shows the means and standard errors for the effects of cue and session on PPI.

**Smoking Cues and Subjective Reactivity**

A 2 × 2 × 2 (Order × Cue × Session) repeated measures ANOVA with cigarette craving as the dependent variable
revealed a significant Order × Cue interaction, $F(1, 25) = 4.73, p < .05$, as well as significant main effects for cue, $F(1, 25) = 5.53, p < .05$, and session, $F(1, 25) = 6.76, p < .05$. The interaction indicated that craving increased after exposure to smoking cues among participants who received control cues first and smoking cues second but that it was not greater after smoking cues relative to control cues when participants received the smoking cue first. A similar pattern was found in an analysis of negative affect with a marginally significant Order × Cue interaction ($p = .06$). Thus, the interaction suggested a differential carryover effect, such that participants who received the smoking cues condition first were still reacting to the smoking cues with increased craving and negative affect throughout the presentation of the control cues (see Figures 2A and 2B).

Because the differential carryover effect was clearly confounding the control cue data, the baseline measures of craving and affect were used instead of the control cue data. Thus, the analysis proceeded with 2 (baseline vs. smoking cues) × 2 (Session 1 vs. Session 2) repeated measures ANOVAs for craving, positive affect, and negative affect. Significant main effects on craving were observed for cue, $F(1, 25) = 24.56, p < .01$, and session, $F(1, 25) = 7.29, p < .05$, suggesting that exposure to smoking cues increased craving relative to a relaxation baseline and that craving at baseline and after exposure to smoking cues was lower in the Session 1 condition.
second session (see Figure 1B). There was no significant interaction.

A similar analysis was conducted on positive affect and negative affect. A significant main effect on positive affect was noted for cue, \( F(1, 25) = 5.81, p < .05 \), and session, \( F(1, 25) = 6.52, p < .05 \), suggesting that positive affect was lower after exposure to smoking cues relative to a relaxation baseline and that positive affect was greater in Session 1 than Session 2 (see Figure 1C). The interaction effect was not significant \( (p > .10) \). Likewise, the analysis of changes in negative affect revealed significant main effects for cue, \( F(1, 25) = 4.79, p < .05 \), and for session, \( F(1, 25) = 7.35, p < .05 \), indicating that negative affect was greater after smoking cue exposure as compared with a relaxation baseline and that negative affect was greater in Session 1 than in Session 2 (see Figure 1D). The interaction effect was not significant \( (p > .10) \).

**Temporal Stability and Intercorrelations of the Cue Reactivity Measures**

The temporal stability of the cue reactivity measures was assessed by computing Spearman correlations between the measures after exposure to the smoking cues on Week 1 and the same measures after exposure to the smoking cues on Week 2. The correlation between PPI on Week 1 and Week 2 was .57 \( (p < .01) \), and the correlation between the subjective craving score on Week 1 and Week 2 was .53 \( (p < .01) \). The correlation between positive affect after exposure on Week 1 and after exposure on Week 2 was .64 \( (p < .01) \), whereas the correlation for negative affect was .65 \( (p < .01) \). Thus, all the measures showed significant correlations between Week 1 and Week 2, suggesting good temporal stability. These findings were likely due to the simple fact that participants were generally consistent in their responses across sessions.

The intercorrelations (Spearman’s) of the dependent measures after the Week 1 cue exposure were also calculated. PPI was not significantly correlated with any of the subjective measures \( (p > .10) \). Negative affect after smoking cue exposure was significantly associated with craving after exposure \( (r = .71, p < .01) \). None of the other subjective measures was significantly correlated \( (p > .10) \). In addition, the correlation between baseline negative affect and craving after exposure to smoking cues was also significant \( (r = .62, p < .01) \). This was likely attributable to the dysphoria that was produced by the requirement of deprivation and suggests that a large portion of the variance in craving after exposure to cues may be explained by negative affect at baseline.

**Discussion**

Consistent with the hypotheses, the findings of the present study demonstrate that exposure to smoking cues attenuated PPI of the startle reflex relative to control cues among deprived smokers. The findings also demonstrate that exposure to smoking cues increased craving, increased negative affect, and decreased positive affect relative to baseline measures among deprived smokers. These findings are consistent with the literature on cue reactivity (e.g., Niaura et al., 1988; Niaura et al., 1998). Of methodological importance, the study also suggested that the counterbalancing of smoking cues and control cues may introduce the confound of differential carryover effects, especially with regard to the analyses of the subjective measures of craving and affect. Thus, smoking cues increased craving and negative affect, and these increases did not dissipate when control cues were presented. Conversely, PPI appeared to be relatively immune to carryover effects because smoking cues attenuated PPI regardless of whether smoking cues or control cues were presented first. The finding that craving was significantly lower during Session 2 than Session 1 was also of methodological importance. It is possible that smokers may have habituated to the laboratory session and to the presentation of smoking cues such that craving decreased in general across multiple sessions.

The finding that smoking cues attenuated PPI is consistent with preclinical studies demonstrating that exposure to drug cues elicits mesolimbic DA activation (Robinson & Berridge, 1993; Stewart et al., 1984; Weiss et al., 1993) and with research demonstrating that mesolimbic DA activation attenuates PPI (e.g., Swerdlow et al., 1994). Although we know of no reports of nicotine cues precipitating increases in mesolimbic activation, it has been established that nicotine increases mesolimbic DA activation, and it is a logical extension that nicotine cues may also induce DA activation. Thus, the attenuation of PPI in the present study may reflect the contribution of an important biological mechanism that has been implicated as one of the primary substrates involved in cue-elicited craving as well as the reinforcement derived from alcohol and drugs. However, this interpretation should be viewed with caution until it can be more thoroughly tested.

Alternatively, this particular finding may represent an information-processing mechanism. The function of the inhibitory mechanism involved in PPI is to facilitate processing of the prepulse by protecting this processing from interference from the startle-eliciting stimulus. It is possible that smoking cues attenuate PPI because smokers automatically allocate more attentional resources to the smoking cues, as compared with control cues, leaving fewer resources available for processing the prepulse. Indeed, results of previous studies have suggested that exposure to smoking cues decreases the availability of cognitive resources (Cepeda-Benito & Tiffany, 1996; Sayette & Hufford, 1994). However, it should be noted that, although directing cognitive resources to the prepulse stimuli may enhance PPI (e.g., Dawson, Hazlett, Filion, Nuechterlein, & Schell, 1993; Filion et al., 1993), there is no evidence that directing cognitive resources to a salient external stimulus, such as smoking cues, decreases PPI. In fact, the results of at least one study suggest that PPI is enhanced both when cognitive resources are directed to the prepulse stimuli and when cognitive resources are directed away to external stimuli (Blumenthal & Flaten, 1994), which contradicts this alternative explanation of the present findings. Nonetheless, this explanation represents an attractive and viable alternative...
hypothesis that could be tested in future studies that use more interesting control cues and nonsmoker controls.

The correlation between cue-induced craving and cue-induced changes in PPI in the current study were nonsignificant, suggesting that the effects of smoking cues on PPI did not mediate the effects of smoking cues on subjective craving. The lack of a significant correlation is not surprising given that different neural substrates are likely to underlie different response domains (i.e., physiological vs. self-report). Other investigations of smoking cue reactivity typically do not show significant correlations between physiological and subjective measures (e.g., Cepeda-Benito & Tiffany, 1996; Drobes & Tiffany, 1997; Tiffany, 1990). The findings from the present study suggest that withdrawal-induced negative affect was the predominant variable associated with subjective craving.

The results of this research have implications for future prevention and treatment efforts. If the effects of smoking cues on PPI reflect the activity of the underlying neurobiological substrates of conditioned appetitive craving or if PPI reflects cue-induced changes in information processing, PPI may be a physiological marker for smokers who are most susceptible to the effects of smoking cues and who may be most vulnerable to relapse. Moreover, the finding that the effects of smoking cues on PPI are evident in a follow-up session 1 week later suggests that this measure may be useful in future studies that involve multiple assessments over time. In addition, PPI and subjective craving may be useful as targets for pharmacological interventions designed to modify cue or drug-elicited mesolimbic activation or cognitive interventions designed to modify cue-induced changes in information processing. Behavioral and pharmacological treatments that prevent smoking-cue induced changes in PPI and increases in subjective craving may prove to be useful in the treatment of nicotine dependence.

Several limitations of this study should be noted. This study was not designed to test the effect of smoking cues on the affective modulation of the startle response. Affective modulation is typically found in the first few seconds after multiple presentations of discrete stimuli with affective content (i.e., the presentation of slides depicting negative affective content; for a review, see Lang, 1995). The procedures used in the present study are not comparable to the procedures used in investigations of affect and startle; instead, the procedures used here were designed specifically to test PPI. Furthermore, the conclusions of the present study are based on a relatively small sample and should be replicated in a larger study. It is possible that the findings are due to biological mechanisms distinct from the activation of mesolimbic DA because 5-hydroxydopamine agonists and noncompetitive N-methyl-D-aspartate antagonists are also known to attenuate PPI (e.g., Bakshi & Geyer, 1995; Varty & Higgins, 1995). Finally, we examined reactivity to cues only after a significant period of deprivation to maximize the salience of the smoking cues. As a result, it is unclear how withdrawal may have influenced the findings. Future researchers should examine the effects of smoking cues on PPI, craving, and mood while manipulating deprivation levels. Researchers should also examine medications that block mesolimbic DA activation to extend the findings of the present study.

References


Revision received December 23, 1998
Accepted January 6, 1999