CUE REACTIVITY TO APPETITIVE AND AVERSIVE CUES AMONG FEMALE
SMOKERS AND NON-SMOKERS

A Dissertation

by

AGNES SUSABDA

Submitted to the Office of Graduate Studies of
Texas A&M University
in partial fulfillment of the requirements for the degree of

DOCTOR OF PHILOSOPHY

December 2009

Major Subject: Psychology
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Approved by:

Chair of Committee, Antonio Cepeda-Benito
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December 2009

Major Subject: Psychology
ABSTRACT

Cue Reactivity to Appetitive and Aversive Cues Among Female Smokers and Non-Smokers. (December 2009)

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This study examined the motivational state associated with smoking craving specifically among women and the effect of deprivation and smoking status on the relationship between responses to appetitive cues. Utilizing both psychophysiological (startle EMG, skin conductance) and self-report measures, we compared cue reactivity to positive, neutral, aversive, smoking, and chocolate pictures among groups of 10 hr. smoking deprived smokers, non-deprived smokers, and non-smokers.

Smokers responded to smoking cues similar to pleasant affect cues with more inhibited startle and high arousal, while responses from non-smoking females indicated a neutral state. However, deprivation also significantly increased startle responses to smoking cues when compared to non-deprived smokers. Furthermore, a closer look at skin conductance responses to aversive cues suggest that smoking status (deprived and non-deprived groups) significantly inhibited one’s ability to habituate to negative affect stimuli. When responses to chocolate cues were examined, psychophysiological and
self-report data seemed to indicate that smoking deprivation influenced one’s ability to attend to other appetitive/rewarding cues. Implications of these results on female smoking craving and sensitivity to appetitive cues are discussed.
DEDICATION

For E.K.
ACKNOWLEDGEMENTS

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CHAPTER I
INTRODUCTION

In recent years, female smokers have been found to be at significantly greater risk than male smokers for heart disease and diabetes (Bolego, Poli, & Paoletti, 2002; Will, Galuska, Ford, Mokdad, & Calle, 2001). Moreover, although for both sexes risks for disease increases with intensity of smoking, the increased risk is more pronounced in females than males (Mucha, Stephenson, Morandi, & Dirani, 2006). However, greater decline of smoking has been noted among men than women in the United States (NIDA, 2002) and this has been partly attributed to greater success in smoking cessation among men (Pomerleau et al., 2005; Bohadana, Nilsson, Rasmussen, & Martinet, 2003). Studies have indicated that the narrowing gender gap in smoking rates may be due to gender differences in factors such as nicotine sensitivity, the likelihood of initiating quitting, effectiveness of nicotine replacement programs, and experience of withdrawal. For example, comparisons between male and female smokers have found that women take smaller, shorter puffs; may be more sensitive to the subjective effects of smoking (Eissenberg, Adams, Riggins, & Likness, 1999); may experience the olfactory/taste stimuli of smoking as more reinforcing (Perkins et al., 2001); report greater increases in

This dissertation follows the style of Addictive Behaviors.
negative affect during acute abstinence (Leventhal, Waters, Boyd, Moolchan, & Lerman, 2007); are more likely to report smoking to reduce negative affect and control weight (Cepeda-Benito & Reig-Ferrer, 2000); and have greater concerns about post-cessation weight gain (Pirie, Murray, & Luepker, 1991).

Most smokers will gain less than 10 pounds after quitting cigarettes, but approximately 10%, of women will gain as much as 30 pounds after quitting smoking (Froom, Melamed, & Benbassat, 1998; Williamson et al., 1991). Often, these concerns about weight gain and fears of fat are motivators for smoking initiation and the continuance of smoking (e.g., Klesges, Meyers, Klesges, & LaVasque, 1989). In turn, concerns of weight gain following smoking cessation are important obstacles towards the success of smoking abstinence (Jeffery, Hennrikus, Lando, Murray, & Liu, 2000) and are associated with a greater likelihood of smoking relapse (Borrelli & Mermelstein, 1998). Notably, female smokers are twice as likely as men to report that they expect to gain a large amount of weight upon smoking cessation, and following cessation, women are more likely than men to report weight gain and increased desire to eat (Pirie, Murray, & Luepker, 1991). Pomerleau, Garcia, Drewnowski and Pomerleau (1991) found that a comparison between female smokers and non-smokers revealed a higher preference for sweet taste among smokers that was also found to increase after acute abstinence. Furthermore, when compared to male smokers, female smokers are also more likely to have increased food intake during abstinence and report that food intake was related to and could reduce smoking cravings (Ogden, 1994). These findings suggest a relationship between smoking and food cravings.
Craving has been viewed by many researchers as playing an important role in the maintenance of addictive behaviors such as smoking (Baker, Morse, & Sherman, 1986; T. E. Robinson & Berridge, 1993; Stewart, de Witt, & Eikelboom, 1984; West & Schneider, 1987). In fact, literature also suggests that the amount of craving experienced post-cessation predict relapse into smoking addiction (Bagot, Heishman, & Moolchan, 2007; Killen & Fortmann, 1997). Two prominent theories have offered a relationship between smoking cravings and other appetitive cues. The common idea proposed in both the incentive sensitization and the dual-affect model is that drug-paired stimuli can become conditioned stimuli which can activate craving, a core motivational state.

*Incentive Sensitization Model*

The incentive sensitization theory proposes that the pursuit of food and drugs share common underlying mechanisms and motivational states. Robinson and Berridge (1993) posited that repeated drug use produced long-lasting neural adaptations in the brain, including the sensitization of the dopamine neural system or brain-pathway responsible for the processing of incentive motivation and reward. This neural sensitization translates into a sensitization towards the incentive value of the drug that results in increased wanting (craving) for the drug. In fact, the sensitization process can result in cue triggered wanting for a reward that may or may not be liked. The theory also posits that drug related stimuli (or conditioned stimuli) have profound effects on the development and expression of this sensitization (Robinson & Berridge, 1993). Berridge (1995) cites studies where investigators found that although dopamine
neurons were activated initially only when food rewards were received and tasted, with repeated practice the activity in these areas of the brain began to precede the reward. Over time, maximal activity of the dopaminergic neurons was elicited by the conditioned stimuli that consistently predicted the reward.

According to Berridge (1995), people develop cravings for certain foods because these foods were past stimuli that were already salient incentives and which become more salient with the activation of the dopaminergic system. In other words, well liked foods may induce neural sensitization of the dopaminergic system. However, neutral stimuli, such as setting the table, become paired with the activation of the system and also can act as future excitors of the neural system themselves. Repeated intake of the food and its rewarding effects lead to the dopaminergic incentive system becoming hypersensitive to the incentive value of food and activation of the this system will result in enhanced responding for a reward, regardless of the extent to which the reward is liked or possesses a positive hedonic value (Berridge, 1995). With time, this response system becomes increasingly automatic and may function out of the individual's awareness.

Robinson and Berridge (2003) further hypothesized that sensitization from one drug or food can also increase the incentive value of other rewards and the conditioned stimuli for those rewards. Thus a hypothesis of the relationship between nicotine and food craving is that nicotine craving can increase cue-triggered urges for food and vice versa. For example, Wyvell and Berridge (2001) found that drug-free rats that had been subjected to a pre-regimen of amphetamine injections worked more for sucrose in
response to food-predictive cues than control rats that had not been pre-sensitized with amphetamine. These results are congruent with the hypothesis that smokers might be sensitized to food cues because of prior exposure to nicotine.

Wack and Rodin (1982) also highlighted that smoking appears to improve information processing and performance in certain visual detection tasks. Commensurate with the incentive sensitization theory, the authors proposed that this heightened arousal of brain mechanisms caused by nicotine sensitized the brain to cues and increased “the probability that the smoker would eat if there were stimulating food cues in the environment” (Wack & Rodin, 1982, p. 371). In finding that glucose tablets can relieve smoking urges (craving), West et al. (1999) also proposed that the desire to smoke shared a common mechanism with appetite and a drive to seek out carbohydrates. Thus, satisfying one need would reduce motivation for the other. West et al. (1999) further suggested that nicotine’s ability to relieve carbohydrate craving may contribute to the relationship between nicotine and food craving. This author postulated that because nicotine can reduce both nicotine craving and hunger in some people, cravings for food/hunger can often be interpreted as cravings for nicotine. Also, to the extent that nicotine reduces hunger and food intake (Bellinger et al., 2005; Wellman et al., 2005), it is also possible that, over time, low nicotine levels overlap and become paired with hunger-like states. This would result in hunger becoming a conditioned stimulus and craving for nicotine its conditioned response.
Dual Affect Model

The dual affect model, on the other hand, proposed that craving can be understood as either a positive or negative affective state. Baker et al. (1986) posited that cravings are affect-related responses that are processed by two mutually exclusive motivational systems in the brain that respond to either appetitive or aversive stimulation and which motivate approach and avoidance responses, respectively. This motivational system has information on affect (craving) related setting events, responses, and possible consequences of various response options. The particular information that is coded into the motive system will vary with drug, drug history variables, and types of cravings. In addition, the threshold of activation of the motive system is reduced as more information is gathered through the reoccurrence of drug exposure and usage. In theory, desires to use drugs or foods (cravings) can be governed by either the appetitive (positive-affect) or avoidance (negative-affect) systems.

Baker et al. (2004) expanded this dual affect model and postulated that the negative-affect motivational system is the main but not necessarily the sole processing channel that promotes drug use. They theorized that the negative-affect motive system codes information that includes withdrawal-associated physiological and behavioral responses, cues previously associated with withdrawal, expectations regarding withdrawal, the consequences of possible response options, and stimuli that signal drug unavailability. Negative affect is a key element in drug withdrawal symptoms and repeated rehearsal of the withdrawal-drug use cycle will sensitize the drug user to preconscious external and internal signals of the negative affect which often follows.
decreasing drug levels in the body. This rehearsal process sets the stage for negative-affect states becoming conditioned stimuli capable of activating the negative-affect motivational system and associated drug use responses.

In theory, seemingly drug-neutral cues such as aversive or disagreeable stimuli that are capable of activating negative affect consciously or unconsciously can generate negative affect cravings. For the active drug user, cravings typically occur due to low levels of negative affect which are detected preconsciously. This unconscious motivational processing will in turn lead to biased attentional and automatic response selection processes which have optimally and effectively ameliorated negative affect in the past (e.g. smoking a cigarette). When drug use is unavailable, interrupted, or when negative affect is high, the addict becomes more conscious of the motivational processes behind his/her cravings. High levels of negative affect will lead to a response bias that will likely inflate the incentive value of the drug and decrease those of alternative reinforcers such as food. According to this theory, when negative affect is at moderate levels, it is at these instances that cognitive control processes are facilitated and factors such as attitudes and expectancies play a more prominent role in drug-use behavior. An important feature of Baker’s *et al.* (2004) motivational system is that activation of either the positive- or negative- affect systems makes the organism insensitive to stimuli that are incongruent with the system that is already activated. Thus, an organism in a positive-affect state would be less responsive to negative-affect stimuli, whereas an organism in a negative-affect state would be less responsive to stimuli associated with the positive-affect system.
Although both the dual affect and incentive-sensitization model would predict that a withdrawal situation would increase the incentive value of drug cues, an important difference is that one focuses mainly on negative reinforcement while the latter on the sensitization of neuronal network responsible for ‘wanting’ of the drug. In other words, the incentive-sensitization model posits that the direct rewarding actions of the drug and the incentive sensitization which occurs as a result are the main motivating factors in drug use and this can occur independent of withdrawal relief. The dual affect model, on the other hand, mainly focuses on a negative reinforcement model where the experience of and anticipation of negative affect plays a central role in drug motivational processes. In this model, drug cues signal the possibility of relief from negative affect and not necessarily its appetitive effects. In addition, the incentive sensitization model would predict that drug cues during abstinence could enhance the incentive value of non-drug rewards (e.g. food), while the dual affect model would posit that increasing negative affect during abstinence would lessen the incentive value non-drug rewards.

Startle Paradigm

In recent years many investigations into the motivational state related to craving has utilized the startle modulation paradigm (Lang, 1995). Lang (1995) proposed that emotions are organized and are driven by two main types of motivation: approach and avoidance. In this theory, it is hypothesized that when the individual’s emotional state is affectively unpleasant, the avoidance/aversive motivation system is activated and defensive reflexes such as startle would increase in amplitude. In contrast, when one’s affect is pleasant, it is suggested that the appetitive/approach motivational state is
activated and defensive reflexes such as startle would be inhibited. In other words, with respect to neutral-affect states, startle reflexes are stronger while experiencing negative affect and weaker while experiencing positive affect.

Lang (Lang, 1995) proposed that activation of the motivational state is not only defined by valence (appetitive/pleasant or aversive/unpleasant) but also by arousal (ranging from calm to aroused). Arousal is viewed as the intensity of activation of either or both motivational states (appetitive or aversive) and may be reflected in the changes of one’s skin conductance response. Cuthbert, Bradley, & Lang (1996) found that as the level of arousal heightens, the potentiation of startle during negative affect and inhibition of startle during positive affect are more pronounced. Although often overlooked or not reported in drug studies utilizing the startle paradigm, arousal as measured by skin conductance is a key factor in the study of drug motivation due to the fact that startle reflexes are also heavily influenced by attentional factors. In particular, at low levels of arousal, skin conductance often is more reflective of an attending/orienting response to novel stimuli (Bradley, Codispoti, Cuthbert, & Lang, 2001). It has been found that defensive startle will be inhibited as more cognitive effort is directed towards attending aversive stimuli lower in arousal and will potentiate at higher levels of arousal (greater activation of defensive motivation). Thus, when data on the startle reflex in response to drug and other affective stimuli are collected independent of arousal measures, there is a risk of not fully understanding whether inhibition or potentiation of startle were due to valence, attention, or arousal effects.
The importance of arousal has also been noted due to the fact that investigations into motivational states can be complicated by other factors such as the co-occurrence of more than one drive. In fact, during an aversive state such as deprivation or withdrawal, appetitive motivation can be activated by stimuli such as food or drug cues (Bradley, Codispoti, Cuthbert, & Lang, 2001). For example, a number of recent studies have found that under deprivation, startle responses to food cues were not reflective of an especially aversive or appetitive response and were more in line with a neutral response (Drobes et al., 2001; Rodriguez, Fernandez, Cepeda-Benito, & Vila, 2005). However, high arousal in response to food cues suggested that the corresponding startle responses were more indicative of frustrative non-reward (co-activation of both appetitive and aversive states) rather than a neutral state (low arousal state).

According to Lang (1995), one’s motivational state can be activated directly through sensory input such as visual stimuli. Expression of the resulting activated motivational state can be measured in a number of ways including physiological changes such as skin conductance and the magnitude of an eye blink resulting from the introduction of a startle probe. With this in mind, they developed a collection of visual stimuli grouped into pleasant, neutral, and unpleasant affective classes and varying along the dimension of arousal. This collection of pictures and the startle paradigm have been widely utilized in craving and cue reactivity research (Drobes et al., 2001; Geier, Mucha, & Pauli, 2000; J. D. Robinson et al., 2007).

Despite a number of studies on the subjective and physiological experiences of nicotine craving, the nature of smoking craving and motivation remains inconclusive.
For example, in some investigations smoking deprivation and smoking status (i.e. smoker vs. non-smokers) failed to significantly modulate baseline startle responses (Mueller, Mucha, & Pauli, 1998) or negative emotional response intensity (Piper & Curtin, 2006), while in another study deprivation decreased habituation of startle responses to negative cues (Cinciripini et al., 2006). Studies have also found that smoking cues activated appetitive motivation independent of deprivation (Geier et al., 2000), or did not modulate startle responses at all when compared to neutral cues (Orain-Pelissolo, Grillon, Perez-Diaz, & Jouvent, 2004). On the other hand, drug cues have also been found to elicit self reports of a variety of negative affect states (Sherman, Zinser, & Sideroff, 1989).

Investigations into the relationship between smoking cessation and food cravings in humans have mostly relied on self-report and limited physiological and behavioral measures (e.g. salivation and amount of food consumed). Recent findings suggest that although food deprivation can increase self-reported smoking cravings, smoking deprivation had no effects on self-reported food cravings (Alsene, Li, Chaverneff, & de Witt, 2002), or cue-elicited salivation (DiLorenzo, Walitzer, Sher, & Farha, 1991). Furthermore, despite the noted limitations of relying solely on self report measures (Sayette et al., 2000), most studies that have specifically investigated female smoking addiction have focused on self-reported subjective experiences of weight gain (Jarry, Combs, Polivy, & Herman, 1998), withdrawal symptoms (Pomerleau et al., 1993), dietary restraint and stress (Mitchell & Perkins, 1998), and body image evaluations (King, Matacin, Marcus, Bock, & Tripolone, 2000).
**Cue Exposure**

Despite recent studies indicating lower success rates of smoking cessation among women and increased sensitivity to smoking cues (Perkins et al., 1999; Field & Duka, 2004), there have been a surprisingly few studies looking specifically at female smokers vs. female non-smokers and differences in cue reactivity to various stimuli. Studies examining cue reactivity to smoking cues involving a female sample have mainly explored gender differences. For example, Field and Duka (2004) have found that deprived female smokers demonstrated a greater sensitivity to smoking cues by reports of increased craving post-cue exposure and also increased salivation to smoking cues. In addition, another study (Knott et al., 2008) indicated that female smokers responding to smoking cues had greater likelihood to experience physiological distress (exhibited through EEG reactivity) and were more likely to report increases in QSU F2 (cravings related to relief of negative affect) and negative affect than their male counterparts. Furthermore, findings of varying categories of characteristics which strongly influence withdrawal symptomatology and one’s ability to abstain from smoking (Pomerleau et al., 2005; Niaura et al., 2001) highlights the need for further investigation of cue reactivity cues among female smokers and non-smokers in developing tailored interventions for female smokers.

**Aims of the Study**

The main goal of the present proposal is to complement the findings of my master’s thesis, which investigated the effect of smoking deprivation on smoking cravings and food cravings in women. In the original study, female smokers were
divided into smoking deprived and a non-deprived group and presented with a series of slides depicting pleasant, aversive, neutral content. In addition, cravings were induced in smoking deprived and non-deprived women through viewing slides depicting smoking-related and chocolate images, respectively. Craving was assessed using self-report measures and an autonomic psychophysiological response, startle eye-blink. Due to research findings suggesting that nicotine administration or cessation does not seem to have a strong relationship to all types of food cravings, but specifically to sweet, high-fat foods (e.g. food intake decreased after nicotine administration Grunberg, 1982; food intake increased after cessation Perkins et al., 1990), cravings for chocolate were assessed concurrently with cravings for smoking.

The goal of the thesis was to evaluate the motivational state and affect related to craving. Based on the theory by Baker et al. (2004), smokers deprived of smoking and presented with visual smoking cues would have greater self-reported cravings to smoke and would also respond with higher startle amplitudes than nondeprived smokers across all stimuli, indicating an increase in defensive motivation. According to Baker et al. (2004), the negative affect states during deprivation would increase the incentive value of the drug and thus we would expect that smoking deprivation would cause smoking pictures to produce a response more in line with appetitive motivation. However, the overall increase in negative affect during withdrawal should potentiate startle and decrease sensitivity to positive cues among deprived smokers compared to smokers continuing to smoke. In contrast, the Incentive Sensitization theory (Robinson and Berridge, 1993) predicts that approach motivation/positive affect phenomena is the basis
of drug craving. According to this view, startle responses to smoking pictures in our abstinent group should also resemble responses to pleasant-related stimuli, but higher startle responses across all stimuli would not be observed.

Finally, we examined whether responses to chocolate pictures among deprived smokers would reflect cue triggered activation of an approach/positive affect motivational system (Zinser et al., 1999) or whether smoking deprivation would decrease the incentive value of a non-drug reward (Baker et al. (2004). Given Robinson and Berridge’s (1993) hypothesis that sensitization from one drug can also increase the incentive value and wanting of other rewards, responses to both smoking and food cues would be similar in affect (both appetitive and similar in reactivity). On the other hand, Baker et al. (2004) negative reinforcement model may theorize that higher level of negative affect during drug withdrawal would result in biased responding only towards the smoking cues and less to the chocolate cues. Thus, responses to smoking cues among the deprived group would be more appetitive (more inhibited reactivity) than responses to chocolate cues.

Results from the thesis suggested that smoking cues were appetitive for both abstinent and non-abstinent smokers (Susabda, Robinson, Cepeda-Benito, & Tamez, manuscript in preparation). However, potentiation of startle reflex responses to smoking cues among abstinent smokers relative to startle responses from non-abstinent smokers was also observed. This may indicate that, although both groups responded appetitively to the smoking cues, non-abstinent female smokers were more sensitive to the positive reinforcing value of the cues. The potentiated startle reflexes in response to smoking
cues among abstinent smokers were not significantly different than responses to positive affect cues, but could also suggest a co-activation of both appetitive states (similar startle reflex responses to smoking and positive affect cues) and aversive (potentiation due to increasing negative affect) among abstinent smokers. The results of this previous study also suggest that chocolate cravings are appetitive for female abstinent and non-abstinent smokers. Although subjective data did indicate that non-abstinent smokers were more sensitive to chocolate craving cues, the psychophysiological data suggest that startle response to chocolate cues were not dependent on smoking abstinence.

There were various limitations in the original study. First, due to the absence of physiological arousal data in this previous study, it remains unclear whether startle responses to chocolate cues and smoking cues in this study were due to increased activation of a motivational state (high arousal), increased attention (low arousal), or the activation of a frustrative non-reward (high arousal). Second, the exclusion of a non-smoking control group also limits interpretation on how smoking deprivation and its accompanying negative affect plays a role in responses to appetitive and aversive stimuli. For example, although abstinent smokers were not found to have potentiated startle responses across all stimuli, it remains unclear whether smokers (abstinent and non-abstinent) experience higher levels of negative affect. The inclusion of a non-smoking control group may reveal that smokers respond differently to affective and reward related cues. Third, we also failed to explore sensitivity to negative cues among deprived smokers by comparing habituation of startle eye blink responses to negative cues. A recent study on deprived smokers that also failed to find potentiated startle
responses across all stimuli has suggested that sensitivity to negative cues could be reflected in a lack of habituation in the startle reflex (Cinciripini et al., 2006).

Thus, this study aims to further explore cue elicited responses to appetitive and aversive stimuli among female smokers by comparing responses to that of female non-smokers and analyzing physiological arousal data. In particular, we intend to explore differences in responses to appetitive cues by comparing the arousal responses to smoking and chocolate pictures among abstinent smokers, non-abstinent smokers, and non-smokers. According to Baker et al. (2004), negative affect experienced/detected by smokers should increase the incentive salience of a drug reward, thus it would be hypothesized that arousal responses to smoking cues should be high among both abstinent and non-abstinent groups. In conjunction with the previous finding of inhibited startle response to smoking cues, high arousal to the same cues among the non-abstinent group would indicate increased activation of an appetitive motivational state; whereas low arousal could be more indicative of inhibited startle due to increased attention. When taking into consideration the moderate startle responses to smoking cues among abstinent smokers, high arousal would indicate activation of one or more motivational states rather than a neutral state. We also intend to re-analyze the startle data to explore whether negative affect is, in fact, increased among abstinent smokers by means of increasing sensitivity to negative cues. As was discussed earlier, this would be accomplished by comparing the habituation of startle eye blink responses to negative stimuli across 3 groups. Baker et al. (2004) would hypothesize that negative affect is increased among deprived smokers; while the incentive sensitization model would
predict that positive affect would be the main motivational state activated during withdrawal. By including female non-smokers, we also intend to further explore whether negative affect responding is increased among smokers in general.
CHAPTER II

METHOD

Subjects

Female cigarette smokers \((N = 55)\) and non-smokers \((N = 55)\) were recruited through the use newspaper adds and fliers posted on public bulletin boards. Individuals interested in participating were screened over the phone and those who met criteria were invited to participate in a study concerning emotional reactions to pictures. Eligibility criteria for smokers included smoking at least 10 cigarettes per day for at least the last 12 months prior to the experiment. Due to unknown but potentially confounding effects of medications, all participants who reported taking prescription medications (aside from birth-control/hormone replacement) were excluded from the study. Likewise, individuals with diabetes or other sugar metabolism problems were also excluded.

Among our sample of smokers, 3 women did not attend their second appointment, 8 were excluded from participation due to scores in the clinical range on the BULIT-R \((n=4)\) or EAT \((n=4)\). Among the non-smoking women, 2 did not attend their second test session, 6 participants were excluded from analysis due to corrupted psychophysiological data files, and 5 scored in the clinical range on the BULIT-R \((n=1)\) or EAT \((n=4)\). The participants' age ranged from 18 to 54 years \((M = 24.2, SD = 8.8)\) and smokers smoked an average of 17.2 \((SD = 7.3)\) cigarettes per day. Frequency of chocolate consumption ranged from less than once a week to daily \((35.7\% \text{ less than once a week, 27.4}\% \text{ once a week, 23.8}\% \text{ 2-3 days out of the week, 13.1}\% \text{ more than 3 days a}}\)
week), with 94% of the participants eating 2 or less servings each time they consumed chocolate.

Callers were informed that some of the smoking participants would be asked to abstain from smoking for 10 hours and all participants were asked to fast for 3 hours prior to the data collection session. Participants were told they would earn a total of $30 for their participation. Smoking subjects who agreed to participate were assigned randomly to either a 10-hour smoking deprivation group or a no deprivation group.

Materials

Visual Stimuli. A collection of 60 colored pictures were presented on a 25-inch computer monitor (Barco Multidata OCM 3346) at a distance of 1.5 m from the subject. The content of the pictures varied across 5 categories with 12 pictures per category. Three of the categories corresponded to the neutral, pleasant, and unpleasant classification of the International Affective Picture System (IAPS; Lang, 1995). These pictures were chosen according to their valence and arousal ratings (high arousal pleasant and unpleasant pictures and low arousal neutral pictures) reported in the IAPS. The fourth picture category corresponded to images depicting cigarettes, smoking-related stimuli, and women holding or smoking a cigarette. These pictures were selected from a pool of pictures according to their craving-evoking properties rated from a sample of college student smokers. The fifth picture category depicted chocolate and chocolate consumption images that were chosen also from a pool of pictures rated on the dimension of chocolate craving by a sample of college students who identified themselves as chocolate cravers.
Self-report Measures

Bulimia Test-Revised (BULIT-R; Thelen, Mitz, & Vander Wal, 1996). The BULIT-R is a 36-item questionnaire used to measure symptoms of bulimia. Only 28 of the items are scored based on responses to multiple choice questions presented in a 5-point, forced-choice format. High scores (104 or above) are indicative of a higher likelihood that the person may be diagnosed as bulimic in a clinical interview.

Eating Attitudes Test (EAT-26; Garner et al., 1982). The EAT-26 is an abbreviated 26-item version of the EAT-40 and has been found to be a reliable, valid measure of the symptoms of anorexia nervosa (Garner et al., 1982). Subjects rate each item using a 6-point scale ranging from 0 (Never) to 6 (Always). Full scale scores range from 0 to 78, with higher scores (>24) indicating a higher presence of disturbed eating patterns and eating disorder symptomatology.

Chocolate Craving Questionnaire-Trait (CCQ-T; Rodriguez et al., 2005). The CCQ-T (39 items) is an adaptation of the Food Craving Questionnaire-Trait (FCQ-T) to measure chocolate cravings (Rodriguez et al., 2005). The FCQ-Trait measures the intensity of 9 trait dimensions of food craving by instructing participants to think about specific foods they tend to crave (Cepeda-Benito et al., 2000). Thus, the CCQ-T instructs subjects how frequently each statement about chocolate would be generally true for them using a 6-point scale ranging from 1 (‘Never’ or ‘Not Applicable’) to 6 (‘Always’). Full scale scores range from 39 to 234, with higher scores indicating higher levels of chocolate craving trait.
Chocolate Craving Questionnaire-State (CCQ-S; Cepeda-Benito et al., 2000).

The 15-item CCQ-S is an adaptation of the Food Craving Questionnaire-State (FCQ-S). The CCQ-S measures 5 state dimensions of chocolate cravings by instructing participants to think about their current chocolate craving and indicating the extent to which they agree with each statement at that moment from "strongly agree" (1) to "strongly disagree" (5). Here, items specifying chocolate craving (vs. hunger or other states) were totalled. Full scale scores range from 9 to 45, with higher scores indicating a higher state of chocolate craving.

Questionnaire of Smoking Urges (QSU; Tiffany & Drobes, 1991). The QSU is a 32-item questionnaire used to assess current craving for smoking (Tiffany & Drobes, 1991). The QSU has 2 factors. The first factor (F1) reflects intention to smoke and the anticipation of pleasure from smoking. The second factor (F2) reflects the anticipation of relief from negative affect and smoking withdrawal. In our sample, Cronbach’s alpha for F1 was 0.79 and F2 was 0.85.

Fagerström Tolerance Questionnaire (FTQ; Fagerström & Schneider, 1989).

The FTQ includes three multiple-choice items (0 to 2 scale) and five two-choice (0 to 1 scale) that are added to compute a total nicotine dependence score ranging from 0 to 11, with high scores indicating higher levels of dependence. (Fagerström & Schneider, 1989).

The Hopkins Symptom Checklist-21 (HSCL-21; Green, Walkey, McCormik, Ross, & Taylor, 1988). The HSCL-21 is a 21-item version of the 58-item Hopkins Symptom Checklist (Derogatis, Lipman, Rickels, Uhlenhuth & Covi, 1974) that is frequently used
to assess symptoms of distress. The HSCL-21 asks subjects to endorse items on a 1 to 4 Likert scale to indicate how they have felt in the previous seven days. Total score scores range from 21 to 84.

*Self-Assessment Manikin (SAM; Lang, 1980).* The SAM is a self-report rating form that consists of figures which represent the dimensions of valence (happy to sad affect), arousal (low to high activation), and dominance (feeling very small to feeling in control). We adapted this ratings form to also include a fourth dimension, craving. Each dimension has 5 figures which represent varying intensity level. Subjects are instructed to rate pictures by selecting the figure that best represents their state for each of the dimensions.

*Demographic, Food, and Smoking History Forms.* These questionnaires collected information on age, race, 3-hour food/caffeine recall data, and smoking history.

*Physiological Measures*

Startle (eye blink) response was used as an index of affective responding to the visual stimuli. Startle responses to food and smoking pictures are conceptualized as reflecting craving and the motivational processes underlying responses to these cues (Geier, 2000; Drobos, 2001; Hawk Jr. *et al.*, 2004). The eyeblink response was assessed as EMG activity using the MP100 System (Biopac, Goleta, CA) data recorder. Two 4mm Biopac Ag-AgCl electrodes filled with electrode gel (Signa Gel) were secured on the orbicularis oculi region below the left eye. Impedance was checked using the UFI 1089 mk III Checktrode. The raw EMG signal was amplified, filtered (bandpass = 10-500Hz), and integrated using EMG100 and the AcqKnowledge 3.5 software (Biopac,
Goleta, CA). The data were edited off-line to detect any clear movement artifact. Scoring of startle responses was accomplished by taking the peak amplitude of EMG integrated signal from 20ms until 120ms after probe onset.

To index arousal, skin conductance response (SCR) was acquired by two Ag/AgCl finger electrodes (Grass Technologies, West Warwick, RI) filled with skin conductance electrode paste (EC33, Grass Technologies, West Warwick, RI) placed in the index and middle finger of the left hand. The SCR data was recorded using BIOPAC System’s GSR100 electrodermal response amplifier. SCR data were also edited off line and scored in microsiemens.

*Procedures*

Participants who met the inclusion criteria and agreed to participate were scheduled for two appointment sessions. Upon arrival to the laboratory for their first session, participants were asked to complete a series of questionnaires (BULIT-R, EAT, CCQ-T, QSU, Fargerstrom, Hopkins Checklist-21, and the Smoking History form). Participants who scored in the clinical range in the BULIT-R (score >104) or the EAT (score > 24) were excluded from participation. Participants who did not qualify or declined participation in the second part of the study were compensated with $10 for their time.

Qualified smoking participants were randomly assigned to either a 10-hour smoking deprivation group or a no-deprivation group and asked to return the next day. All participants were asked to not eat and to drink their usual amounts of caffeine for 3 hours prior to the testing session. On the day of the second testing session, a blood
sample was obtained to measure glucose level (Bayer Dex Meter Glucometer) and then a CO-level test was performed. Smoking subjects assigned to the non-abstinent group were asked to smoke one of their own cigarettes shortly after their arrival to their second session; while subjects in the deprived and control group were asked to chew sugar-free mint gum for 5 minutes. After smoking the cigarette, or chewing the gum, all participants were then asked to fill out a food log form and the CCQ-S. Smoking subjects assigned to both the abstinent and non-abstinent groups also filled out the QSU.

After the questionnaires were completed, all subjects were asked to rinse and dry their hands and sit in a comfortable recliner. Their face was then prepared for electrode placement and the electrodes were attached according to established guidelines (Blumenthal, et al., 2005). The light in the room was dimmed, headphones were put in place, baseline physiological data were collected for 10 minutes while the participant relaxed, and then physiological reactivity (eye blink startle response and skin conductance) to neutral, positive, negative, chocolate and smoking pictures were monitored. Each subject were instructed to watch each picture for the entire time it was on the screen and to ignore the noises that could come from the headphones.

At the end of the visual presentation, the electrodes were removed and the participants were asked to fill out the CCQ-S and the QSU. The pictures were then shown again in groups of three, with all the pictures in each group corresponding to the same type of picture (i.e., aversive, pleasant, neutral, smoking, or chocolate). Participants were asked to rate all the pictures using the SAM figures. Each picture was shown for 6 seconds and, after each block presentation, participants had 15 seconds to
rate each picture type along dimensions of valence, arousal, dominance, and craving. All participants were then debriefed and paid $30. The second testing session lasted approximately 1½ hours.

*Stimuli Presentation*

The pictures were presented in two pseudorandomised orders, where each picture was shown for 6 s, followed by a blank (white background) monitor for 10 seconds. The acoustic startle stimulus consisted of a 100dB (A) white noise burst presented for 50 ms over Sennheiser EH2270 headphones. The noise was produced by Cool Edit 2002 (Syntrillium, Phoenix, AZ) with instantaneous rise time. To reduce anticipation of the startling noise, the noise was presented at three random intervals from 2.5 to 5 s after picture onset (2.5, 4, and 5) and only during nine of twelve pictures per picture category. Additionally, nine startle probes were presented randomly during inter-trial intervals (ITI). The presentation and timing of the pictures and startle probes was controlled by Superlab software (Cedrus Corporation, San Pedro, CA).

*Data Reduction*

Startle responses were scored off line by extracting the peak amplitude of startle responses for each trial (falling within a 21-120 ms window following the acoustic stimuli) (Blumenthal, *et al.*, 2005). A difference score was then obtained for each startle response by subtracting the mean baseline EMG activity (1 second before onset of acoustic stimuli) for that particular trial. Trials where the waveform suggested too much baseline activity or clear movement artifact in the startle response were considered a zero-response trial and not included in the analyses. To correct for individual
differences in startle response magnitudes, each startle response was converted to a $z$ score (using the mean and sd of that particular subject’s startle response), and then transformed to a $T$ score ($[z \times 10] + 50$) (Drobès et al., 2001).

Skin conductance responses will also be scored offline (in microsiemens) by calculating deviations of every second (in the 7 seconds following onset of each picture) from a 1 second baseline period before picture onset. The maximum deviation of responses which occurs between 1 and 4 seconds after picture onset will be scored and a log transformation performed to normalize the data (log[SCR]) (Bradley, Codispoti, Sabatinelli, & Lang, 2001).

**Statistical Analysis**

All analyses will be performed using the SPSS 16.0 statistical package. The subjective data will be first analyzed using either multivariate between group ANOVAs (Fagerström, CCQ-T, Hopkins, BULIT-R, EAT-26) or mixed between group with repeated measures ANOVAs (QSU, CCQ-S, SAM). Between group comparisons will explore differences between abstinent, non-abstinent, and non-smokers on their responses to these measures.

A mixed design ANOVA will be performed using startle probe times (2.5, 4, 5 seconds) as the within subjects factors and group as the between. Researchers have also reported that activation of attentional processes during earlier parts of picture viewing may inhibit startle, and that affective modulation of the startle response may be more likely to occur during the second half of a 6-second picture presentation (Bradley, Cuthbert, & Lang, 1999; Codispoti, Bradley, & Lang, 2001). Thus, repeated measures
ANOVA’s will be conducted using time as the within subjects variable to test for differences in startle responses between startles introduced at 2.5 and 4 seconds, and between 4 and 5 seconds within each type of picture presentation.

In our assessment of whether female smokers and non-smokers presented modulation of startle in response to aversive, pleasant, and neutral stimuli, a repeated measures ANOVA compared blink startles in response to Positive, Negative, and Neutral pictures. This analysis was conducted to test whether we have replicated significant differences between Positive, Negative, and Neutral pictures, as found by other researchers (Vrana, Spence, & Lang, 1988; Geier, Mucha, & Pauli, 2000, Drobes et al., 2001). Mixed repeated measures ANOVA’s will also be used to compare skin conductance responses to positive, neutral, and negative stimuli among the three groups.

Repeated measures ANOVAs will be conducted to explore whether startle responses and skin conductance for smoking and chocolate pictures will be significantly different than positive, neutral or negative startle responses. Furthermore, to determine a significant difference between Group 1 (abstainers), Group 2 (non-abstinent), Group 3 (non-smokers) on their startle response to Chocolate and Smoking pictures, a mixed design ANOVA will be conducted using Group as the between subjects factor and Picture Type as the repeated measures factor. This difference will test whether smoking abstinence or smoking status can increase the motivation for chocolate and smoking. We will also examine how smoking status influence startle responding in the presence of smoking and chocolate cues after controlling for Trait Chocolate craving (CCQ-T).
In addition, the effect of smoking abstinence on habituation in startle and skin conductance responses across time to the negative affect pictures will be explored by comparing changes in responses across 4 blocks of presentation time by group in mixed repeated measures ANOVA’s.

To control for deviations from the sphericity assumption, the degrees of freedom associated with the within factor were adjusted using the Greenhouse-Geisser correction for all of our repeated measures analysis. Interaction effects were further explored using either repeated or univariate ANOVAs. Statistical significance was set at $\alpha = .05$, which was adjusted using the Bonferroni method for post hoc comparisons.
CHAPTER III
RESULTS

Subjective Variables

Table 1 summarizes the univariate analysis of variance (ANOVA) results that tested for baseline between group differences across theoretically relevant variables. Smokers in the abstinent (GRP 1) and control (GRP 2) groups reported similar levels of nicotine dependence (FTND), chocolate craving traits (CCQ-T), symptoms of eating disorders (BULIT-R and EAT-26), and levels of psychological distress (HSCL-21). That is, the randomization of smokers into the abstinent and control conditions created two comparable groups of participants. Compliance to study instructions was high as all individuals in the deprived group met the CO level requirement (cutoff of 6); whereas none of the ongoing smokers yielded values at or below the cutoff.

When compared to our group of female smokers, the non-smoking control group (GRP 3) reported similar levels of chocolate craving traits (CCQ-T) and eating disorder symptoms (EAT-26); however, the non-smoking group reported significantly less psychological distress (HSCL-21), F(1, 84) = 4.62, p<.05 and bulimia-specific (BULIT-R), F(1, 75) = 5.90, p<.01.

To monitor smoking cravings among the female smokers, we conducted two repeated measures ANOVAs with time of assessment (baseline, pre-cue exposure, post-cue exposure) as the repeated measures factor and group (abstinent, control) as the between subjects factors. There are two main factors on the QSU, one which involves the anticipation of positive outcomes from smoking (F1) and another which highlights
the anticipation of relief from nicotine withdrawal and/or negative affect associated with withdrawal (F2). From the F1 factor on the QSU, the results yielded a significant within subjects effect, $F(2, 74) = 16.27, p < .001$, a significant group effect, $F(1, 37) = 23.26, p < .001$, and a time by group interaction effect, $F(2, 74) = 18.52, p < .001$. The interaction effect reflected a significant increase in F1 craving report from baseline to pre-cue exposure for GRP 1, $F(1, 18) = 19.15, p < .001$, but a significant decrease in F1 craving for the GRP 2, $F(1, 20) = 12.03, p < .005$. Following cue exposure, change in report of F1 craving significantly increased only in GRP 2, $F(1, 20) = 36.29, p < .001$ (see Table 2 and Figure 1); however, GRP 1 still maintained significantly higher cravings.

Reports of cravings that anticipate relief from nicotine withdrawal and/or negative affect (F2) had a similar trend across time as reports of the anticipations of positive outcomes (F1) (see Figure 2). The results yielded a significant within subjects effect, $F(2, 78) = 10.04, p < .001$, a significant group effect, $F(1, 39) = 18.44, p < .001$, and a time by group interaction effect, $F(2, 78) = 15.44, p < .001$. The interaction effect indicated that reports of cravings that anticipate the negative reinforcement qualities of smoking declined from baseline to the beginning of the second session (pre-cue exposure) for GRP 2, $F(1, 21) = 19.95, p < .001$, but increased for GRP 1, $F(1, 19) = 20.23, p < .001$. In comparing reports at pre-cue exposure and post-cue exposure, the results revealed a significant increase in F2 craving only for GRP 2, $F(1, 20) = 15.76, p < .01$. 
To assess chocolate cravings (CCQ-S) over time, we also conducted repeated measures ANOVAs with time of assessment (baseline, pre-cue exposure, post-cue exposure) as the repeated measures factor and group (GRP 1, 2, 3) as the between subjects factors. The results yielded a significant effect for time, $F(1.83, 146.69) = 38.03$, $p < .001$, a time by group interaction effect, $F(3.67, 146.69) = 3.43$, $p < .05$, but the between group effect was not statistically significant (see Table 2, Figure 3). The time effect indicated that chocolate cravings did not change from baseline to pre-cue exposure for all 3 groups, but increased from pre- to post-cue exposure. To interpret the interaction, we conducted pre- to post-cue exposure changes within each group and found significantly higher chocolate cravings at post-cue vs. pre-cue exposure in GRP 2, $F (1, 20) = 35.17$, $p < .001$, and GRP 3, $F (1, 41) = 26.94$, $p < .001$. The absence of significant increases in chocolate cravings in GRP 1 could indicate that smoking deprivation decreased appetitive reactivity to other rewarding cues in the environment.

Overall, the results suggest that the experimental manipulation, smoking deprivation vs. non-deprivation, was effective in that from baseline to pre-cue exposure deprived smokers reported higher augmentation of smoking cravings than non-deprived smokers. Conversely, chocolate craving remained unchanged from baseline to pre-cue exposure in all groups. Non-deprived smokers were the most responsive to the cue-exposure procedure with the highest report of smoking and chocolate cravings. Non-smokers reported a significant increase in chocolate craving from pre to post-cue exposure, indicating they reacted to the exposure of chocolate pictures. This seems to
suggest that non-deprived smokers and non-smokers were more reactive to reward-related cues than smoking-deprived smokers.

SAM Ratings

Each SAM variable (affective valence, arousal, dominance, and craving) was analyzed separately using a mixed, repeated measures ANOVA, with the five types of pictures as the repeated factor and group as the between subjects factor (see Table 3). For each ANOVA, we specified four a priori planned contrasts to compare the means of each picture type to the mean of the neutral picture category.

The significant repeated measures effects were as follows (see Table 3, Figures 4-7). Results for valence indicated a significant effect for picture type, $F (2.79, 234.05) = 28.39, p < .001$, and a significant picture type and group interaction effect, $F (5.57, 234.05) = 5.45, p < .001$. The within subjects effect indicated that participants rated aversive pictures as negative in affect; and pleasant and chocolate pictures as positive in affect. To interpret the interaction effect we conducted separate between subject ANOVAs for each picture type. We found that GRP 2 and GRP 3 rated chocolate pictures as more positive in affect than GRP 1, $F (1, 86) = 4.59, p < .05$, and GRP 3 rated smoking pictures as more negative in affect than GRP 1 and GRP 2, $F (2, 86) = 19.98, p < .001$.

In terms of arousal, there was a significant within subjects effect for picture type, $F (3.25, 269.50) = 9.74, p < .001$, a significant between group effect, $F (2, 83) = 5.19, p < .01$, and a significant picture by group interaction, $F (6.50, 269.50) = 4.54, p < .001$. Across the 3 groups, participants reported greater arousal reactivity to negative, positive
and chocolate pictures than to neutral pictures. The interaction effect was due to a
significant between subjects effect on arousal responding to smoking pictures. That is,
GRP 1 and GRP 2 reported significantly higher arousal to these cues than GRP 3 F (1, 86) = 45.00, p < .001.

In terms of SAM ratings of dominance, there was statistically significant effect
for picture type, F (3.19, 265.00) = 18.92, p < .001 and a significant picture type by
group interaction, F (6.39, 265.00) = 3.03, p < .01. Here, there were significant
differences between neutral and both unpleasant and smoking pictures in GRP 1 and
GRP 2, with both groups reporting less control in reaction to unpleasant, F (1, 43) =
27.33, p < .001, and smoking pictures, F (1, 43) = 21.16, p < .001, than in response to
neutral pictures. GRP 3 reported significantly less dominance to unpleasant vs. neutral
pictures, F (1, 42) = 9.36, p < .01, but significantly higher dominance to smoking vs.
neutral pictures.

Reports of craving for chocolate and smoking on the SAM indicated a significant
picture type by group interaction. In GRP 1, smoking craving was significantly greater
than chocolate craving, F (1,19) = 17.26, p < .01, while in GRP 2 there were no
differences between smoking and chocolate cravings. Among GRP 3, they reported
greater chocolate than smoking cravings, F (1, 42) = 63.99, p < .001. Between subject
comparisons revealed that GRP 2 reported greater chocolate craving than GRP 1, F (1,
42) = 7.95, p < .01, and that GRP 1 and GRP 2 reported higher smoking cravings than
GRP 3, F (1,85) = 149.26, p < .001.
Startle Cue Reactivity

Given that attentional processes may inhibit startle during early segments of picture presentation (Bradley, Cuthbert, & Lang, 1999; Codispoti, Bradley, & Lang, 2001), it was important to examine whether startle probes presented earlier in the trial (i.e. at 2.5 seconds) resulted in startle amplitudes that were significantly different than those presented later in the trial (i.e. at 4 and 5 seconds). Thus, we conducted a mixed repeated measures ANOVA using the IAPS cues (i.e. pleasant, neutral, and aversive pictures) and the three startle probes in our study (i.e. 2.5, 4, and 5 seconds) as two within group factors and group (GRP 1, GRP 2, and GRP 3) as the between subjects factor. The analysis revealed a significant interaction of startle probe and picture type (See Figure 8). Planned comparisons involved 2 mixed repeated measures ANOVA’s comparing startle probe presentations (2.5 vs. 4 and 4 vs. 5 seconds) by picture type by group. For the ANOVA testing startle amplitude differences between 2.5 and 4 second probes, there was a time by picture effect, $F(1.9, 145.42) = 5.76, p < .05$. Follow up analyses revealed lower startle amplitude at 2.5 vs. 4 seconds for aversive pictures, $F(1, 79) = 8.69, p < .01$, and higher amplitude at 2.5 sec than 4 sec probes for neutral pictures, $F(1, 78) = 6.83, p < .05$. Although a trend revealed decreasing startle across tim, the follow-up comparison for the pleasant pictures was not significant. For the ANOVA testing startle amplitude differences between the 4 and 5 sec probes there was no time nor time by picture interaction effect, all $Fs < 3$. Thus, heretofore, emotional modulation of the startle response will be tested using the average of the startle response across the 4 and 5 second probes.
To test the extent to which our lab could assess emotional modulation of the startle response, we tested the startle modulation to positive, neutral, and aversive pictures. We conducted a mixed repeated measures ANOVA, with picture type as the repeated measure (pleasant, neutral, and aversive), and group (GRP 1, GRP 2, GRP 3) as the between subjects factor. This analysis revealed an effect for picture type, F (1.84, 141.46) = 11.81, p < .001, $\eta^2 = .13$), and for group, F(2, 77) = 5.46, p < .01, $\eta^2 = .12$), but no picture by group interaction effect. Post hoc analysis of the between group effect, comparing 2 groups at a time, revealed a significant between subjects effect for the GRP 1 vs. GRP 2 comparison, p < .01. A priori planned comparisons comparing startles to aversive and pleasant pictures to neutral revealed that startles to aversive pictures were potentiated with respect to the startles to neutral pictures, F (1, 77) = 19.13, p < .001, $\eta^2 = .19$, with no differences between neutral and pleasant pictures.

Three separate mixed repeated ANOVAs examined startle modulation to chocolate pictures. Each ANOVA compared the startles to chocolate pictures with the startle to the three IAPS pictures (pleasant, neutral, or aversive) between the three groups. The comparison of chocolate and aversive pictures between groups revealed a significant main effect for picture type F (1, 77) = 33.43, p < .001, $\eta^2 = .28$, but no significant between subject or interaction effects. The analysis revealed that the three groups responded to chocolate pictures with significantly inhibited startle when compared to aversive pictures. A priori planned comparisons revealed that startle to chocolate pictures in all 3 groups were similar to that of neutral pictures and, for GRP 1 and 3, similar to startle to positive pictures. However, for GRP 2, their response to
chocolate pictures were significantly more inhibited than that of positive pictures, $F(1,19) = 6.48, p < .05, \eta^2 = .25$.

An additional set of three separate mixed repeated ANOVAs examined startle modulation to smoking pictures. Each ANOVA compared the startles to smoking pictures with the startle to the three IAPS pictures (pleasant, neutral, or aversive) between the three groups. As with chocolate pictures, startles to smoking pictures were inhibited with respect to aversive pictures $F(1, 76) = 47.50, p < .001$, but there were no group or picture by group interaction effect. The analysis comparing smoking and neutral pictures resulted in a significant main effect for picture type, $F (1, 76) = 8.06, p < .001$, and a significant interaction of group and picture type, $F (2, 76) = 3.12, p < .05$. The interaction indicated that while for GRP 1 and GRP 3, startles to smoking and neutral pictures were similar; GRP 2 startles to smoking pictures were significantly inhibited compared to neutral pictures, $F (1, 19) = 15.03, p < .005, \eta^2 = .44$. A similar trend was found in comparing startle to positive and smoking pictures, with GRP 2 startles to smoking pictures significantly more inhibited compared to positive pictures, $F (1, 19) = 15.96, p < .01$. The data suggests that smoking pictures presented to Group 2 subjects (non-deprived smokers) resulted in further inhibition of startle responses when compared to neutral and positive affect pictures; while in GRP 1 and GRP 3 the pattern was not observed. The significantly lower startle inhibition to smoking pictures in GRP 1 than GRP 2 suggests that smoking deprivation lowers the positive-affect value of smoking pictures.
Skin Conductance

Analysis of arousal responses to all 5 pictures across the 3 groups (see Figure 8, Table 4) revealed a significant main effect for picture type, $F(3.81, 289.58) = 8.11$, $p < .001$ and, although GRP 1 had a notably higher trend of arousal across the 5 pictures than GRP 2 and GRP 3, there was no significant group or interaction effects. A priori planned comparisons confirmed a similar level of arousal for positive and aversive pictures across the 3 groups. Arousal for neutral pictures were significantly lower than arousal to aversive pictures in GRP 2, $F(1, 19) = 5.18$, $p < .05$ and GRP 3, $F(1, 38) = 8.60$, $p < .01$; but was not significant for GRP 1. Thus, for GRP 2 and GRP 3, the trend in arousal to positive/aversive/neutral pictures was as expected with higher arousal to positive and negative pictures than to neutral pictures; whereas for GRP 1 this trend in skin conductance was less pronounced.

A priori planned comparisons for skin conductance to chocolate pictures compared them to positive, aversive, and neutral affect pictures within each group. For all 3 groups, arousal to chocolate pictures was significantly lower than that of positive and negative pictures; but similar to skin conductance levels for neutral pictures. Furthermore, for smokers in GRP 1 and GRP 2, arousal to chocolate pictures was lower, but not significant, than arousal to smoking pictures. In GRP 3, skin conductance levels to chocolate and smoking were notably similar. When taking into consideration the inhibited startle response in all 3 groups to chocolate pictures, the skin conductance data to chocolate pictures may suggest a response that is more similar to neutral rather than appetitive. However, significant appetitive and craving self-report responses to the
chocolate images, especially in GRP 2 and GRP 3 indicate that inhibited arousal and startle to the cues may be more of an orienting response.

Analysis of skin conductance responses to smoking pictures compared to each of the other 4 pictures found that among smokers in GRP 1 and GRP 2, arousal to smoking pictures were similar to that of positive and aversive pictures. However, for non-smokers (GRP 3), arousal to smoking pictures were significantly less than positive pictures, $F(1, 39) = 4.03, p < .05$ and to aversive pictures, $F(1, 39) = 8.12, p < .005$, but similar to neutral. Here, the trend in skin conductance for neutral, chocolate, and smoking pictures among non-smokers was lower levels of arousal when compared to positive and negative pictures. On the other hand, in GRP 2, arousal to smoking pictures was higher (although not significant) when compared to neutral pictures and more similar to arousal for positive and negative pictures. Among smoking deprived subjects (GRP 1), there was a higher (yet non-significant) level of arousal across the pictures when compared to deprived smokers, but also a lack of skin conductance changes across positive, negative, neutral, and smoking pictures. This may indicate a state of frustration due to non-reward in response to smoking pictures. In GRP 2, a higher arousal response in conjunction with inhibited startle response to smoking pictures seem to have indicated appetitive motivation; whereas in GRP 3, inhibited startle response to the smoking cues along with low arousal indicated a neutral state.

**Covariates**

The CCQT did not significantly correlate to startle or skin conductance responses to the 5 picture types. We also tested significance of other possible covariates such as
nicotine dependence (FQT) and number of cigarettes smoked to physiological responses, but did not find any significant correlations.

**Habituation**

Habituation to aversive cues was examined by separately comparing startle and skin conductance responses across 4 blocks of time in the visual presentation. That is, we conducted separate group by block repeated ANOVAS for skin conductance and startle responses to aversive pictures. When comparing habituation of the startle response to negative affect pictures, there was no significant group or interaction effects. However, habituation in arousal response to the negative affect cues indicated a significant decrease in arousal over time only among non-smokers (GRP 3), $F(1, 37) = 25.57, p < .001$, and maintenance of high arousal to aversive pictures across time among all smokers (GRP 1 and GRP 2). This seems to suggest that smokers, deprived and non-deprived, are more sensitive to negative affect cues in their environment.
CHAPTER IV

SUMMARY

This investigation explored cue elicited responses to appetitive and aversive stimuli among female smokers by comparing responses from psychophysiological and self-report data to a control group of female non-smokers. In particular, we explored the relationship of responses to appetitive cues by observing valence and arousal responses to smoking and chocolate pictures among abstinent smokers with a 10 hr. deprivation, non-abstinent smokers, and non-smokers. Results suggest that although all smokers responded to smoking cues with inhibited startle and high arousal indicating appetitive motivation, smoking deprivation among female smokers heightened startle to smoking cues when compared to non-deprived smokers. A trend in higher arousal responses among deprived vs. non-deprived subjects also point to an increase in sympathetic activation and anxiety. The potentiated startle among deprived smokers, along with increased skin conductance response across all affective stimuli may indicate a state of frustrative non-reward and a decreased ability to respond to appetitive cues. Non-smoking controls, on the other hand, responded to the smoking cues in a neutral manner.

Self report measures of smoking craving were commensurate with the psychophysiological data. The smoking cues were effective in that participants in GRP 1 and GRP 2 reported appetitive motivation, lower dominance, and high level of craving in SAM ratings to the pictures, while non-smokers rated them to be aversive with low arousal, low craving, and higher dominance. In addition, subjective data of craving (QSU) from deprived smokers suggests co-activation of appetitive and aversive states in
that they rated significantly higher smoking cravings related to both positive and negative reinforcement. Due to possible ceiling effects of high smoking craving reported by GRP 1 participants undergoing withdrawal at pre-cue exposure, exposure to smoking cues significantly increased smoking craving only among GRP 2.

All groups responded to chocolate cues with inhibited startle and low arousal suggesting a neutral state. However, self report ratings of chocolate cues and chocolate cravings were somewhat at odds with the psychophysiological data in that all 3 groups rated chocolate cues to be appetitive and highly arousing. Furthermore, for non-deprived smokers and non-smokers, their higher craving rating for chocolate on the SAM and greater increases in craving on the CCQ-S seem to indicate that their psychophysiological response to the chocolate cues were more of an orienting rather than a neutral state. For deprived smokers, the cue exposure did not increase their cravings for chocolate. In fact, abstinent smokers rated their smoking craving to be significantly higher than chocolate cravings, while non-deprived smokers reported similar cravings for smoking and chocolate. The implication is that smoking deprivation seems to decrease sensitivity to rewarding cues in the environment outside of smoking.

As suggested in previous research (Sutton, 1997; Bradley, Cuthbert, & Lang, 1999; Codispoti, Bradley, & Lang, 2001), affective modulation of the startle response was restricted to the latter part of the 6-second picture presentation. Specifically, startle responses to affective cues elicited at 2.5 seconds significantly differed from startles produced at 4 and 5 seconds. Here, we found that across the 3 groups modulation
occurred among cues presented with 4 and 5 second startle probes, with no significant differences between time.

Commensurate with self-report, we found evidence of emotional modulation of the startle reflex to positive and aversive affect cues. Specifically, the startle to aversive pictures was potentiated with respect to the startle response to positive and neutral pictures. Furthermore, as predicted, a trend across the 3 groups indicated that skin conductance responses to positive and negative affect pictures were higher than that of neutral. Although we did not find significant differences between startle responses to positive and neutral affect pictures, some researchers have argued that viewing positive affect pictures rated significantly higher in arousal than neutral cues may lead to higher than expected startle responses (Dillon & LaBar, 2005).

It was surprising to find decreased modulation of skin conductance responses among abstinent smokers (i.e. no significant differences in skin conductance responses between affective cues); however, other researchers have also found a similar trend among food deprived subjects (Drobes, et al., 2001). A trend of higher arousal in conjunction with decreased modulation of skin conductance responses among abstinent smokers seems to reflect a state of higher activation and anxiety. Thus, among smokers, deprivation seem to increase anxiety such that they may have a more difficult time regulating their physiological response to varying cues.

Although a comparison of habituation to the startle probe across time did not reveal any significant group differences as has been indicated by other researchers (Cinciripini et al., 2006), analysis of adaptation of arousal responses to negative affect
pictures across time indicated a lack of habituation to the cues among all female smokers and implied sensitivity to aversive cues among smokers. Commensurate with our finding of higher psychophysiological arousal among deprived smokers and also a lack of adaptation to aversive cues over time among all smokers, there is evidence from other researchers (Kassel and Unrod, 2000; Gilbert et al., 2008a; Gilbert et al., 2008b) that nicotine interacts with distracting cues in the environment to lower anxiety levels. In line with our finding of a lack of adaptation to aversive cues among all smokers, Gilbert (1995) and Kassel and Unrod (2000) proposed that even smokers who continue to smoke will not experience decreased anxiety without the presence of distracting or more positive cues in the environment. This along with significantly higher reports of distress among our smoking subjects strongly suggests that female smokers experience a higher level of overall anxiety. Nicotine’s proposed ability to direct attention to distracting or benign cues can also be seen in that non-deprived smokers displayed modulation of arousal across picture types and were be able to attend and to report a higher increase in chocolate craving to the cues than deprived smokers.

Baker et al. (2004) proposed that negative affect is increased among deprived smokers; while the incentive sensitization model suggested that positive affect would be the main motivational state activated during withdrawal. This investigation confirmed through psychophysiological and self report measures that although female smokers responded to smoking cues in a manner similar to appetitive motivation (Geier et al., 2000; Mucha et al., 1999), smoking deprivation increased reports of withdrawal symptoms related to negative affect and resulted in potentiated startle responses to
smoking pictures when compared to non-deprived smokers. Although we did not find (as Baker’s theory would suggest) that nicotine significantly increased positive affect responding among non-deprived smokers (as would be indicated by inhibited startle responses across affective cues when compared to deprived smokers), this finding is in line with Gilbert’s (1995) proposal that nicotine’s ability to increase positive affect may not be observed when there is a lack of situational factors which allow attention to be reallocated. However, we did find some evidence of increased negative affect among deprived smokers in that they were not as sensitive to other reinforcing cues in their environment such as chocolate, had increased skin conductance responses to all affective cues, and decreased modulation of arousal between the cues.

Although we did find significantly potentiated startle to smoking cues among deprived vs. non-deprived smokers, commensurate with other studies (Orain-Pelissolo, Grillon, Perez-Diaz, & Jouvent, 2004) there was a lack of startle modulation when comparing neutral and smoking cues among deprived smokers. However, our finding of decreased startle modulation to smoking/neural cues among deprived female smokers reporting high distress (on HSCL-21) is in line with recent findings (Lang and McTeague, 2009) suggesting that chronic anxiety/negative affectivity decreases defensive reactivity to personalized negative affect cues. Overall, results indicating appetitive motivation and yet also increased arousal across affective cues and potentiated startle to smoking cues suggest that female smoking craving during an acute state of withdrawal is experienced as an appetitive and a co-occurring aversive/anxious state. This co-occurrence of both appetitive and aversive motivation during withdrawal based
craving was also reflected in QSU reports of significantly higher urges related to both the anticipation of pleasure/positive outcomes and relief from negative affect. Other studies have also found that deprivation of smoking (Payne, Smith, Sturges, & Holleran, 1996) and other substances such as certain types of food among food cravers (Drobes et al., 2001; Rodriguez et al., 2005) may result in ambivalent responses indicative of a frustrative non-reward state.

Both psychophysiological and self-report data from non-smokers in this study consistently suggest that, unlike previous research indicating an aversive response (Geier et al., 2000), female subjects without any smoking history experience responded to smoking cues with a neutral response. The inclusion of a non-smoking group was important in that we were able to confirm that female smokers do report higher levels of negative affect (HSCL-21) along with higher physiological arousal responses to aversive cues across time. Although results indicated higher chocolate craving responses from non-deprived smokers compared to the other 2 groups, this was not significant when comparing GRP 2 and GRP 3. However, given differences in group size, along with a lack of high chocolate cravers in our sample, nicotine may in fact be able to further increase sensitivity to rewarding cues in the environment among female smokers above that of a non-smoking sample.

In this study, we were able to utilize a startle paradigm to demonstrate smoking cue reactivity among women and explore motivational/affective state of smoking craving. Future directions of this research would include increasing the deprivation manipulation and recruitment of female smokers reporting higher chocolate craving in
order to more closely reflect cravings among women motivated to abstain from smoking and the possible relationship between chocolate craving traits and smoking craving. Research findings (Dempsey, Cohen, Hobson, & Randall, 2007) suggesting that stage of change in smoking cessation can also influence physiological (including startle) responsiveness to smoking cues also highlights the importance of recruitment of smokers motivated to quit in studies attempting to characterize craving among women. In addition, including subsets of female smokers who report higher (clinical range) eating disorder, anxiety, or depressive symptoms in future studies, we may be able to further explore how different subsets of female smokers respond to affective/craving cues.

Future work need to also explore the use of other physiological and self-report measures of affective state in studies exploring smoking craving. Particularly, a recent study of smoking cue reactivity utilizing the startle paradigm have suggested that voluntary emotion regulation may not be compromised by deprivation/withdrawal (Piper & Curtin, 2006). However, other researchers have noted that blink startle responses may have low test-retest reliability in studies voluntary emotion regulation and that corrugator EMG is a more stable measure of trait-ability for emotion regulation (Lee, Shackman, Jackson & Davidson, 2009). Furthermore, recent studies have also suggested that postauricular reflex is another index of motivation/affect that is sensitive to valence and may be useful in conjunction with startle EMG in studies examining ambivalent cues (Hess, Sabourin & Kleck, 2007). The inclusion of a state measure of overall negative affect/depressive symptoms at pre and post-cue exposure could have
also examined the consistency between self report and physiological measures given our findings of a more ambivalent response in smoking craving.
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APPENDIX A

FIGURES
Figure 1. QSU factor 1 cravings (positive reinforcement effects) as measured between group and across time

**a = p < .001, within group
**b = p < .001, between group
Figure 2. QSU factor 2 cravings (negative reinforcement effects) as measured between group and across time

**a = p < .001, within group
**b = p < .001, between group
*a = p < .01, within group
*b = p < .01, between group
Figure 3. CCQ-state as measured between group and across time

**a = p < .001, within group
**b = p < .001, between group
*a = p < .05, within group
*b = p < .05, between group
Figure 4. SAM valence ratings
Figure 5. SAM arousal ratings
Figure 6. SAM dominance ratings
Figure 7. SAM craving ratings
Figure 8. Startle EMG response between affective picture types and across startle probes

Picture 1 = Pleasant
Picture 2 = Neutral
Picture 3 = Aversive
Figure 9. Startle EMG responses between group and across 5 picture types
Figure 10. Skin conductance responses between group and 5 picture types
Table 1. Measures on day 1

<table>
<thead>
<tr>
<th>Measure</th>
<th>Group 1 (abstinent Day 2)</th>
<th>Group 2 (non-abstinent Day 2)</th>
<th>Group 3 (non-smokers)</th>
</tr>
</thead>
<tbody>
<tr>
<td>FTQ</td>
<td>4.0 (2.4)</td>
<td>4.12 (1.9)</td>
<td>NA</td>
</tr>
<tr>
<td>CCQ-T</td>
<td>82.86 (36.90)</td>
<td>89.77 (29.60)</td>
<td>82.46 (29.14)</td>
</tr>
<tr>
<td>BULIT-R</td>
<td>60.18 (16.12)</td>
<td>54.50 (17.75)</td>
<td>46.21 (13.77)**b</td>
</tr>
<tr>
<td>EAT-26</td>
<td>7.60 (5.15)</td>
<td>10.74 (8.20)</td>
<td>7.66 (5.33)</td>
</tr>
<tr>
<td>HSCL-21</td>
<td>39.00 (13.45)</td>
<td>39.35 (11.96)</td>
<td>34.26 (8.98)**b</td>
</tr>
</tbody>
</table>

mean (SD)

* (p<.05) comparing GRP 1 vs. GRP 2
*a (p<.05) comparing SMOKING vs. NON-SMOKING
*** (p<.01) comparing GRP 1 vs. GRP 2
**b (p<.01) comparing SMOKING vs. NON-SMOKING

Table 2. Measures of smoking and chocolate craving as a function of cue exposure and deprivation

<table>
<thead>
<tr>
<th>Measure</th>
<th>Group 1</th>
<th>Group 2</th>
<th>Group 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>QSU F1</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>time 1</td>
<td>81.40 (21.02)</td>
<td>75.41 (18.52)</td>
<td>NA</td>
</tr>
<tr>
<td>time 2</td>
<td>97.85 (9.35)**a</td>
<td>61.14 (21.56) **a,b</td>
<td>NA</td>
</tr>
<tr>
<td>time 3</td>
<td>99.25 (7.07)</td>
<td>79.55 (19.91) **a,b</td>
<td>NA</td>
</tr>
<tr>
<td>QSU F2</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>time 1</td>
<td>41.95 (16.00)</td>
<td>35.30 (11.75)</td>
<td>NA</td>
</tr>
<tr>
<td>time 2</td>
<td>54.95 (14.21)**a</td>
<td>28.45(11.49) **a,b</td>
<td>NA</td>
</tr>
<tr>
<td>time 3</td>
<td>55.05 (15.34)</td>
<td>36.23(15.88) *a,b</td>
<td>NA</td>
</tr>
<tr>
<td>CCQ-S</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>time 1</td>
<td>15.10 (5.85)</td>
<td>16.19 (6.77)</td>
<td>15.88 (7.24)</td>
</tr>
<tr>
<td>time 2</td>
<td>14.55 (5.88)</td>
<td>14.57(5.11)</td>
<td>14.86 (5.82)</td>
</tr>
<tr>
<td>time 3</td>
<td>18.00 (9.43) *b</td>
<td>24.33(9.77) **a</td>
<td>19.57 (8.19) **a</td>
</tr>
</tbody>
</table>

mean (SD)

* a p < .05 (within group)
*** a p < .001 (within group)
* b p < .05 (between group)
** b p < .01 (between group)
Table 3. SAM ratings across different picture types within each dimension

<table>
<thead>
<tr>
<th>Picture</th>
<th>Valence</th>
<th>Arousal</th>
<th>Dominance</th>
<th>Craving</th>
</tr>
</thead>
<tbody>
<tr>
<td>Positive</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>GRP1</td>
<td>4.03 (1.87)</td>
<td>5.43 (2.07)</td>
<td>5.83 (1.98)</td>
<td>na</td>
</tr>
<tr>
<td>GRP2</td>
<td>4.14 (2.25)</td>
<td>5.24 (1.94)</td>
<td>6.02 (1.29)</td>
<td>na</td>
</tr>
<tr>
<td>GRP3</td>
<td>4.34 (1.94)</td>
<td>4.81 (2.07)</td>
<td>5.95 (1.41)</td>
<td></td>
</tr>
<tr>
<td>Neutral</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>GRP1</td>
<td>5.09 (1.08)</td>
<td>3.96 (1.69)</td>
<td>5.95 (1.84)</td>
<td>na</td>
</tr>
<tr>
<td>GRP2</td>
<td>5.02 (1.05)</td>
<td>4.09 (1.89)</td>
<td>5.85 (1.88)</td>
<td>na</td>
</tr>
<tr>
<td>GRP3</td>
<td>4.88 (0.75)</td>
<td>3.73 (1.90)</td>
<td>5.82 (1.72)</td>
<td></td>
</tr>
<tr>
<td>Negative</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>GRP1</td>
<td>5.64 (1.35)</td>
<td>5.74 (1.73) **a</td>
<td>3.76 (1.22) *a</td>
<td>na</td>
</tr>
<tr>
<td>GRP2</td>
<td>5.51 (1.70)</td>
<td>5.57 (1.88) **a</td>
<td>3.99 (1.93) **a</td>
<td>na</td>
</tr>
<tr>
<td>GRP3</td>
<td>6.08 (1.07) **a</td>
<td>4.59 (2.81) **a</td>
<td>4.59 (2.41) **a</td>
<td></td>
</tr>
<tr>
<td>Chocolate</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>GRP1</td>
<td>4.05 (1.71) *b</td>
<td>4.57 (2.05) **a</td>
<td>5.55 (1.99)</td>
<td>4.38 (2.27) *a</td>
</tr>
<tr>
<td>GRP2</td>
<td>3.12 (1.27) *b</td>
<td>5.38 (1.66) **a</td>
<td>5.47 (2.06)</td>
<td>6.03 (1.57)</td>
</tr>
<tr>
<td>GRP3</td>
<td>2.99 (1.12) *b</td>
<td>5.30 (1.31) **a</td>
<td>5.75 (1.75)</td>
<td>5.23 (2.17) **a</td>
</tr>
<tr>
<td>Smoking</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>GRP1</td>
<td>3.98 (1.68) *a</td>
<td>6.83 (1.50) **a</td>
<td>4.14 (1.81) *a</td>
<td>6.75 (1.72)</td>
</tr>
<tr>
<td>GRP2</td>
<td>3.65 (1.31) *a</td>
<td>5.78 (1.86) **a</td>
<td>4.65 (1.55) *a</td>
<td>6.24 (2.03)</td>
</tr>
<tr>
<td>GRP3</td>
<td>5.85 (1.86) *a</td>
<td>3.80 (1.86) **a,b</td>
<td>6.26 (1.80) **a</td>
<td>2.21 (1.30) **b</td>
</tr>
</tbody>
</table>

mean (SD)
* a p < .05 (within group)
** a p < .001 (within group)
* b p < .05 (between group)
** b p < .001 (between group)
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