CAFFEINE CUE-REACTIVITY: THE IMPACT OF CAFFEINE-RELATED STIMULI AND EXPECTANCIES ON CAFFEINE CRAVING

by

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ABSTRACT

Caffeine is consumed by approximately 90% of adults, yet its potential addictive properties have been understudied. Specifically, this study examined the impact caffeine cue reactivity and expectancy on caffeine craving and withdrawal. Following 18-hour caffeine abstinence, 65 participants, all daily caffeine consumers, had their caffeine craving and withdrawal symptoms assessed. They then received either caffeine-containing or placebo gum; some received inaccurate information regarding the gum's caffeine content. Next, participants were exposed to neutral- and caffeine-related stimuli (first visual, then auditory/olfactory), before having their craving measured again. In this study, we demonstrated the first-known example of caffeine cue reactivity. Caffeine cues elicited increased caffeine and coffee craving as well as increased heart rate. We also demonstrated brief temporary expectancy effects; caffeine withdrawal symptoms decreased for those who were told they consumed caffeine gum 30 minutes post gum administration. However, there was no impact of expectancy on caffeine or coffee craving.

LIST OF ABBREVIATIONS USED

ANCOVA	analysis of covariance
ANOVA	analysis of variance
BPM	beats per minute
CWSQ	Caffeine Withdrawal Symptom Questionnaire
DSM-5	Diagnostic and Statistical Manual of Mental Disorders, Fifth Edition
F	F-test from ANOVA or linear marginal models
M	mean
mg	milligram(s)
n	sample size
oz	ounce
p	p-value for testing significance
SD	standard deviation
SE	standard error
SPSS 25.0	Statistical Package for Social Sciences, version 25.0
TFBC	Timeline Follow Back Calendar
VAS	Visual Analogue Scale
η^2_p	partial eta squared

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CHAPTER 1: INTRODUCTION

1.1 Caffeine and Caffeine Dependence

Caffeine is the most commonly used psychoactive substance in North America, with the majority of adults (80-90%) consuming it regularly (Centre for Addictions and Mental Health, 2011; Johnson, 2012). The average Canadian adult is estimated to consume 238 mg of caffeine per day, yet there is a wide range of variation between individuals (Centre for Addictions and Mental Health, 2011; Striley, Griffiths, & Cottler, 2011).

Caffeine dependence has been shown to develop following repeated use of caffeine, such as one or two cups of coffee each morning (Juliano & Griffiths, 2004). However, the mechanisms by which this dependence develops are not thoroughly understood. The neurotransmitter adenosine is known to be involved, as repeated exposure to caffeine is believed to increase the amount of adenosine in the brain; this leads to hypersensitivity following abstinence (Johnson, 2012). Indeed, caffeine dependence has been confirmed by the appearance of a variety of withdrawal symptoms, such as sleepiness, lethargy, and headaches (Johnson, 2012). In research conducted with rats, a model based on the striatal adenosine A_{2A}-dopamine D_2 receptor heteromer was able to explain caffeine's low probability for addiction and relative weak reinforcing effects; however, this study also showed that caffeine could increase the addictive and toxic effects of other drugs (Ferré, 2016).

Indeed, many individuals who consume caffeine on a regular basis exhibit dependence-like behaviours and have difficulty in quitting or reducing caffeine intake (Hughes, Oliveto, Liguori, Carpenter, & Howard, 1998). Due to this, there have been

some questions regarding whether there should be a specific diagnosis of caffeine use disorder or caffeine dependence in the Diagnostic and Statistical Manual of Mental Disorders (DSM-5; American Psychiatric Association, 2013). Historically, caffeine use disorders have been excluded from previous versions of the DSM, due to a lack of evidence demonstrating both withdrawal and dependence to the drug (Juliano, Evatt, Richards, & Griffiths, 2012). Although more recent studies have demonstrated this evidence of caffeine dependence, the need to examine other aspects of the drug, specifically comparing the pharmacological effects of caffeine to those of other addictive substance, remains. As such, this study intends to bridge the gap in knowledge regarding caffeine cue reactivity and expectancy.

1.2 Cue Reactivity

Cue reactivity is a commonly used paradigm in addiction psychology, based on the notion that substance users are more vulnerable to craving and use when they are exposed to stimuli associated with prior use of the substance (Drummond, 2001; Siegel, 1975; Stewart, De Wit, & Eikelboom, 1984; Tiffany, 1990). A potential explanation for the phenomenon is the incentive salience hypothesis (Robinson & Berridge, 2008). This hypothesis proposes that initially neutral cues (e.g., a package of cigarettes) are paired via classical conditioning with the pharmacological effect of a drug on the brain's reward circuitry. These neutral cues gain 'incentive salience' – the cues are not only easily noticed and attended to, but they also motivate behaviour toward the drug they are connected to (Robinson & Berridge, 1993). The authors state that the incentive motivational effects of a drug and their cues become stronger (or sensitized) after repeated exposure. This is due to increased activity in the mesolimbic dopamine system

and neuroadaptations of brain circuits that mediate the classical conditioning of incentive motivational processes. This incentive motivation can last for years or even decades, even after the person has recovered from addiction (Robinson & Berridge, 2008). This begins to elucidate the importance of cues when it comes to substance use.

Cue reactivity paradigms have been shown to provoke both significant self-reported craving and physiological responses in users of tobacco, alcohol, heroin, and cocaine (Carter & Tiffany, 1999). In tobacco users, photographic cues have been shown to elicit robust cue-reactivity effects, particularly self-reported craving (Wray, Godleski, & Tiffany, 2011); when exposed to lit cigarettes, smokers showed both an increase in subjective craving and skin conductance (Carter & Tiffany, 2001). A Dutch study on alcohol cue reactivity found significant effects of craving and physiological (heart) responses to alcohol-related video clips (Witteman, et al., 2015). In former heroin users (both recent and over one-year-abstinent users), exposure to heroin-related video cues resulted in increased heroin craving, skin conductance, heart rate, and blood pressure, compared to a control group of never-users and to exposure to neutral cues (Zhao, et al., 2012). A study investigating cue reactivity in cocaine users found significant effects for craving and physiological responses (heart rate, skin conductance, and skin temperature); they also found that these effects were different depending on the cue sensory modality used to elicit the response (Johnson, Chen, Schmitz, Bordnick, & Shafer, 1998). Altogether, these studies show not only the importance of investigating both craving and physiological responses, but that sensory modalities may also play a role in cue reactivity. At the outset of the current research, no study had investigated cue reactivity in caffeine users.

1.3 CRAVING

Craving in and of itself has been a controversial topic in addiction psychology over the years. Disputes and debates have been brought forth over the clinical definition of the word, how it is and has been measured, how it relates to addiction, as well as the usefulness in studying craving in general (Tiffany & Wray, 2014). At its most basic level, craving can be defined as the subjective experience of wanting a drug (Drummond, 2001). This definition captures three distinct ideas regarding craving: that craving is conscious, is best described as an expression of desire, and that it is directed toward the use of a drug (Tiffany & Wray, 2014). This definition ties in well with the incentive sensitization theory mentioned previously (Robinson & Berridge, 2008); cue reactivity and craving are interconnected, so both should be investigated simultaneously.

Currently, craving is not listed as a withdrawal symptom of caffeine in the DSM-5 (American Psychiatric Association, 2013), however, it is important to investigate craving as a measure of motivation to consume a substance (Mills, Boakes, & Colagiuri, 2016; Sayette, et al., 2000). Following abstinence from a substance, craving is significantly increased, and poses a risk for relapse (Larimer, Palmer, & Marlatt, 1999). Because of this and the connection between craving and cue reactivity, we felt it was important to investigate caffeine craving. Craving has typically been measured via self-report questionnaires (Sayette, et al., 2000), but it has also been measured via physiological measures (Drobes & Thomas, 1999). In this study, we used a combination of both types to better capture caffeine craving.

1.4 EXPECTANCY

Whereas craving plays an influential role in substance use, it is also important to

understand the role of various pharmacological and non-pharmacological factors in motivating drug use. Indeed, previous studies have shown that craving and withdrawal related responses can be influenced by placebo effects. Placebo effects come from the administration of a drug or following a certain procedure, but not from the direct effects of the drug or procedure; instead, the effect is based on the individual's beliefs regarding the drug or procedure (Stewart-Williams & Podd, 2004). For example, in one study, 835 British women who frequently used pain medication for headaches were randomly assigned into one of four groups (Branthwaite & Cooper, 1981). The first group received aspirin labeled with a popular brand name, the second group received the same aspirin in plain packaging, the third group received a pill with no active analgesic (a placebo pill) that was labeled with the same popular brand name, and the fourth group received the pill with no analgesic in plain packaging. In this study, branded aspirin was more effective in treating headaches than unbranded aspirin, which was more effective than the branded placebo, which in turn was more effective than unbranded placebo. Not only taking an active analgesic provided headache relief, but also the mere idea or act of taking an analgesic and expecting the pill to provide relief led to headache relief.

This placebo effect also occurs in recreational substance users. In a study investigating cigarette craving, participants were randomly assigned to one of four conditions in a balanced placebo design: half the participants received nicotine-containing lozenges and half received placebo lozenges (Schlagintweit, Good, & Barrett, 2014). Half of the participants in each of those groups were then provided with deceptive information regarding the nicotine content of the lozenge (e.g., told it contained nicotine when it did not). Participants' belief that they received nicotine, regardless of whether they actually received nicotine or not, significantly reduced their craving. Similarly, it

has been demonstrated that expectancy effects also occur in connection with the caffeine content of coffee. In a recent study, participants who were led to believe that they received caffeine had a greater reduction in craving and caffeine withdrawal symptoms than those who believed they did not receive caffeine, despite the fact that both groups had received decaffeinated coffee (Mills et al., 2016). Therefore, it appears expectancy may play a role in an individual's level of caffeine craving.

Believing instructions, that you actually received the substance you were told you received, plays an important role in expectancy. Previous studies have shown that participants who believe instructions ('believers') frequently have different findings when compared to all participants (Kelemen & Kaighobadi, 2007; Schlagintweit, Greer, Good, & Barrett, 2015). In one study, a small group of non-believers (7.5% of the total sample), produced a substantial decrease in effect size, though they did not affect statistical significance (Kelemen & Kaighobadi, 2007). As such, participants' beliefs regarding group manipulation also must be taken into consideration.

1.5 Study Aims

To date, no study has investigated the concurrent roles of expectancy and cue reactivity in caffeine. However, there is research indicating that nicotine replacement therapies are ineffective at preventing relapse in situations involving cue-induced cigarette craving (Ferguson & Shiffman, 2009). Further, there is evidence to suggest that pharmacological and non-pharmacological components of nicotine replacement therapy, regardless of nicotine content, were unable to prevent cue-induced craving (Schlagintweit et al., 2014). As such, although the belief that one has received caffeine has been shown to reduce caffeine craving, it is possible that this belief will not

completely eliminate the craving induced by exposure to caffeine-related stimuli, similar to that seen in tobacco research.

The current study aimed to investigate cue reactivity in daily caffeine consumers. Participants were all adult, daily coffee drinkers, as previous research has shown the expectancy of caffeine consumption associated with coffee is greater than that of either tea or soda (Huntley & Juliano, 2012). Cue reactivity will be investigated using different sensory modalities: visual (via pictures) and a combination of auditory and olfactory. This study also used a balanced-placebo design, to allow for participants' expectancies regarding caffeine consumption to be manipulated (via caffeinated and non-caffeinated gum). As belief of instructions plays an important role in expectancy, we aimed to compare the results of participants who believed instructions (regarding caffeine consumption) to the results of all participants.

1.6 HYPOTHESES

Based on previous cue reactivity paradigm research, we hypothesized that (1) the expectancy that one has received caffeine would reduce withdrawal-related craving, regardless of whether or not they have actually received caffeine, relative to the placebo-expectancy condition (Mills et al., 2016), and (2) that craving, withdrawal symptoms, and heartrate would increase following caffeine cues, regardless of sensory modality and whether the participant has received caffeine or not (Ferguson & Shiffman, 2009; Schlagintweit et al., 2014).

CHAPTER 2: METHOD

2.1 PARTICIPANTS

A total of 65 participants (35 male) were recruited from the Halifax Regional Municipality, Nova Scotia, via online and community bulletin boards. An initial telephone screening was conducted with all potential participants to confirm they met eligibility requirements. Specifically, participants were required to be daily coffee consumers (averaging at least 300mg of caffeine per day) for the past year, confirmed via self-report during telephone screening. Exclusion criteria was as follows: any serious medical conditions, any current DSM-5 diagnosis of psychiatric disorders or neurological disease, use of psychotropic medications, daily cigarette smoking, and prior usage of caffeine pills or gum. Participants were also required to abstain from caffeine use for 18 hours prior to their study session.

Participants were randomly assigned to receive either a caffeine-containing or caffeine-free gum, as well as congruent or incongruent instructions regarding its content. This resulted in participants being divided into four different conditions: (a) told caffeine, received caffeine (n = 16); (b) told caffeine, received placebo (n = 16); (c) told placebo, received caffeine (n = 17); and (d) told placebo, received placebo (n = 16). The average study participant was 34 years old. All participants reported daily coffee use over the past month; in-session reported values for past-week caffeine consumption varied greatly, with the average participant consuming over 400mg of caffeine per day. Most participant first tried coffee in their teenage years and had been a daily coffee drinker for around 15 years (see Table 2.1 for a further breakdown of participant characteristics). All participants provided voluntary, written consent to participate in the

study; further, they were compensated for their time at $12 per hour and an additional $10 for abstaining from caffeine for 18 hours prior to the study session. The study received ethical approval from the Nova Scotia Health Authority Research Ethics Board.

Table 2.1 Mean (standard deviation) values for participant characteristics across the four groups. No group was significantly different than the others in current age, past week caffeine consumption, age when first tried coffee, or years as a daily coffee drinker (all *p* values > .05).

Told/ Received	Placebo/ Placebo (*n* = 16; 9 male)	Placebo/ Caffeine (*n* = 17; 9 male)	Caffeine/ Placebo (*n* = 16; 8 male)	Caffeine/ Caffeine (*n* = 16; 9 male)	*p* values
Age in years	28.56 (9.54)	35.71 (15.17)	38.31 (12.98)	33.13 (11.35)	.113
Past week caffeine consumption in mg/day	364 (94)	464 (143)	358 (86)	497 (256)	.066
Age in years when first tried coffee	13.81 (3.87)	16.71 (5.42)	15.56 (5.10)	14.63 (5.66|)	.360
Years as a daily coffee drinker	12.01 (10.98)	15.79 (14.77)	18.00 (12.09)	12.68 (9.45)	.448

2.2 MATERIALS

2.2.1 Products

Gum. The caffeinated gum contained 100 mg of caffeine per piece (Military Energy Gum, MarketRight, Inc.). Caffeine-containing gum has been shown to be safe, with no adverse effects in healthy adults (Kamimori, et al., 2002). The placebo (caffeine-free) gum was obtained from the same company as the caffeinated gum; it was matched in appearance and flavour to the caffeinated gum. Neither gum is available commercially

in Canada, which decreased the likelihood of participants being familiar with the product, helping to ensure blindness to their condition.

2.2.2 Measures

Heart rate. Participant heart rates were measured using a Polar H10 Heart Rate Sensor (Polar Electro Canada, Inc.). This device was worn on a chest strap underneath participants' clothing, which allowed heart electrical pulses to be measured directly.

Timeline Follow Back Calendar (TFBC). This weeklong calendar was used to help participants recall their caffeine use over the past week. The type and amount of caffeine-containing product was recorded (e.g., a large coffee from Tim Horton's); caffeine content was calculated using online databases.

Caffeine Withdrawal. The Caffeine Withdrawal Symptom Questionnaire (CWSQ; Juliano, Huntley, Harrell, & Westerman, 2012) is a 23-item questionnaire used to assess caffeine withdrawal. The CWSQ has been shown to have high internal consistency ($\alpha = 0.90$) and to be sensitive to caffeine abstinence in both daily and non-daily caffeine users (Juliano, Kardel, Harrell, Muench, & Edwards, 2019) and has been tested in populations comparable to ours (Juliano et al., 2012).

Mood and Caffeine Craving. A Visual Analogue Scale (VAS) was used to analyze participants' mood and craving. The VAS consisted of 13 mood descriptors ('Stimulated', 'Relaxed', for example) as well as a caffeine craving ('Crave Caffeine') and coffee craving ('Crave Coffee') item. Each item was rated on a scale from 1 to 10, with the endpoints being 'Not at all' and 'Extremely'. VAS's have been demonstrated to be valid, reliable and sensitive to subjective individual experiences across a multitude of age ranges and with many different substances (Bond & Lader, 1974).

Product Liking. A single-item visual analogue scale was used to assess the degree

to which participants liked the study gum. The item was rated from 1 to 10, with the endpoints being ‘Not at all” and ‘Extremely’.

Demographics and Caffeine Use. A Demographic and Caffeine Use Questionnaire was used to collect demographic (age, sex, marital status, education, and employment) and caffeine use information (age of first use, caffeine use frequency, typical coffee consumed, and whether participants drank coffee for its taste or its stimulating properties).

Concluding Questions. These questions were used as a manipulation check to verify whether participants believed the information provided regarding the caffeine content of the gum. Participants also had the ability to provide formal feedback on their experience participating in the study.

2.3 PROCEDURE

All participants underwent a telephone screening interview, in which they were informed of the nature and purpose of the study, time commitment, compensation, etc. They were informed that an 18-hour caffeine abstinence period was required to participate in the study. In order to increase the likelihood of adherence to the abstinence requirements, participants were told (during the screening interview) that a saliva sample may be taken during the study session to ensure they had not consumed caffeine in the past 18 hours; however, no saliva samples were taken. Instead, abstinence requirements were verified by self-report.

Following the verification of abstinence, participants completed the TFBC, the CWSQ, the VAS, and had their heart rate measured for one minute (baseline). Once these baseline measures were taken, participants were assigned to one of the four

conditions (as per 'Chapter 2.1 Participants') and then administered a piece of gum, which may or may not have contained caffeine. Participants were told whether the gum contained caffeine, but in two of the groups, the information given was incongruent with the true caffeine content of the gum. Both the researcher and the participant were blind to the actual caffeine content of the gum; the packaging of the gum matched the information provided by the researcher. The participants then chewed the gum in a standardized manner over a 10-minute period (i.e., the gum was chewed in time with an audio recording), such that 99% of the caffeine was released (Newman, Kamimori, Wesensten, Picchioni, & Balkin, 2013). Following gum administration, there was a 30-minute waiting period, allowing for blood caffeine to reach peak levels (Syed, Kamimori, Kelly, & Eddington, 2005). During the waiting period, participants completed the single-item VAS, assessing product liking, as well as the Demographics and Caffeine Use Questionnaire.

Following the 30-minute waiting period, caffeine craving, withdrawal, and mood was reassessed using the CWSQ and VAS; heart rate was also reassessed, again over a one-minute period. Next, participants were presented with neutral and caffeine cues. Neutral cues were presented to all participants prior to caffeine cues in order to avoid carryover effects on ratings of mood and craving (Sayette, Griffin, & Sayers, 2010). Both the neutral and caffeine visual cues lasted for two minutes, each comprising 40 high resolution images. The caffeine cues consisted of coffee-related images (e.g. cups of coffee, coffee being poured into a cup), whereas the neutral cues consisted of water-related images (e.g. water bottles, water being poured into glasses). The caffeine and neutral cues were visually matched with one another and were free of imagery associated with other addictive substances (McGrath, Peloquin, Ferdinand, & Barrett, 2015).

During the first minute of both the neutral and caffeine visual cues, participants had their heart rate assessed; immediately following the presentation of the both neutral and visual cues, participants completed the CWSQ and VAS.

After the visual cues, there was a 10-minute washout period. Following the washout period, participants were presented with neutral and caffeine-related auditory and olfactory stimuli for four minutes. Auditory and olfactory cues were presented simultaneously. For the neutral auditory cues, sounds of water were played; the ambient scent of the room was used as the neutral olfactory cues. For the caffeine-related cues, a pot of coffee was brewed out of view of the participant, producing both auditory and olfactory stimuli. As with the visual cues, heart rate was assessed across the first minute of the cues while the CWSQ and VAS were completed following both the neutral and caffeine related cues.

Upon completion of the questionnaires, participants were given the opportunity to drink coffee. Participants were provided with a 4oz coffee mug (1 unit of coffee), the coffee brewed during the olfactory cues, and their preferred condiments. They could consume anywhere between no units and three units of coffee; for each unit of coffee they did not drink, participants received an extra $1. Participants were required to remain in the lab for 30 minutes following the olfactory cues, regardless of whether they consumed coffee; they could choose to consume their units of coffee at any time during this period. At the end of the session, participants completed the concluding questions, which included a manipulation check. The manipulation check inquired about the caffeine content of the gum the participant received earlier on in the session.

2.4 Statistical Analyses

Data were analyzed using either analysis of variance (ANOVA) or, in the case of the analyses of expectancy and drug effects following gum administration, an analysis of covariance (ANCOVA). Main measures included subjective ratings (e.g., mood, craving, caffeine withdrawal symptoms) and physiological responses (e.g., heart rate). For the analyses of expectancy and drug effect post gum administration, changes in these measures were calculated between groups while controlling for baseline values. Changes in these measures were also analyzed between neutral and caffeine cues for each sensory modality. Cues (neutral vs. caffeine) were analyzed as within-subjects factors; Gum Told (told caffeine vs. told placebo) and Gum Received (received caffeine vs. received placebo) were analyzed as between-subjects factors. Cue reactivity was examined by comparing the subjective and physiological responses across cues; particularly, responses following neutral visual cues were compared to responses following caffeine visual cues and responses following neutral auditory/olfactory cues were compared to responses following caffeine auditory/olfactory cues. Expectancy effects were examined by comparing the subjective and physiological responses between conditions. Secondary analyses included a comparison between conditions on gum liking and units of coffee consumed; these were completed using a 2 (Gum Told) X 2 (Gum Received) between-subjects analysis of variance (ANOVA). When an interaction was observed, a post hoc pairwise t-test was conducted.

CHAPTER 3: RESULTS

3.1 RESULTS FOR ALL PARTICIPANTS

3.1.1 Gum Liking

Ratings of gum-liking were subjected to a two-way ANOVA having two levels of Gum Told (told placebo, told caffeine) and two levels of Gum Received (received placebo, received caffeine). There was a main effect of gum received, $F(1,61) = 31.75$, $p < .001$, $\eta^2_p = .342$, indicating that gum-liking ratings for the placebo gum ($M = 6.50$, $SE = .39$) were significantly higher than gum-liking ratings for the caffeine-containing gum ($M = 3.39$, $SE = .39$). The effect for gum received was not significant, $F(1,61) = 3.62$, $p = .062$, $\eta^2_p = .056$, nor was there a significant interaction, $F(1,61) = 0.60$, $p = .443$, $\eta^2_p = .010$.

3.1.2 Expectancy and Drug Effects Post Gum Administration

Thirty-minute post gum administration measures of mood and craving were subjected to a two-way ANCOVA having two levels of Gum Told (told placebo, told caffeine) and two levels of Gum Received (received placebo, received caffeine), controlling for baseline values. There was a main effect of Gum Told for 'stimulated', $F(1,59) = 9.72$, $p = .003$, $\eta^2_p = .141$. Participants in the told caffeine condition ($M = 4.88$, $SE = .35$) reported significantly higher stimulation than those in the told placebo condition ($M = 3.33$, $SE = .34$). For 'trouble concentrating', there was both a significant main effect of Gum Told, $F(1,60) = 6.66$, $p = .012$, $\eta^2_p = .100$, and a significant Gum Told by Gum Received interaction, $F(1,60) = 4.93$, $p = .030$, $\eta^2_p = .076$. Participants who received caffeinated gum reported significantly more trouble concentrating when they were told they received placebo ($M = 4.74$, $SE = .41$) compared to those who were told

they received caffeine (M = 2.74, SE = .42), p = .001. There was no significant difference between participants that received placebo (see Figure 3.1). There was also a significant main effect of Gum Received for 'relaxed', $F(1,60) = 6.69$, $p = .012$, $\eta^2_p = .100$; this indicates that participants in the received placebo condition (M = 6.90, SE = .29) reported feeling more relaxed than those in the received caffeine condition (M = 5.83, SE = .29). There were no other significant main effects nor interactions regarding mood or craving 30-minutes post gum administration.

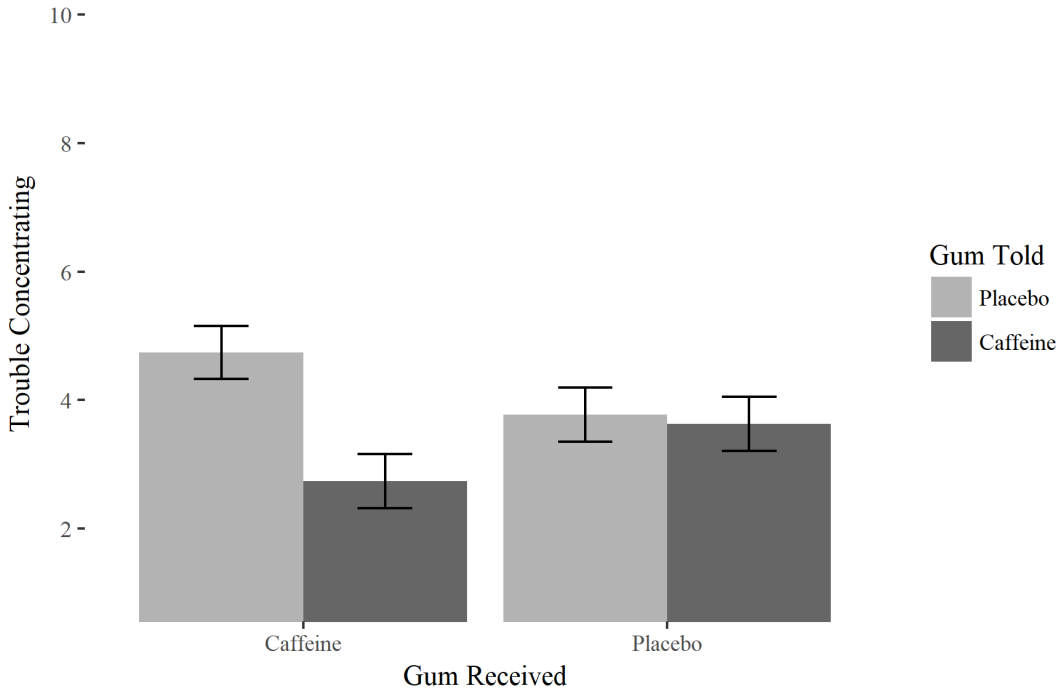

Figure 3.1 Estimated marginal means (±standard error (SE)) for subjective ratings of 'trouble concentrating' 30 minutes post gum administration, covarying for baseline ratings. Participants who received caffeinated gum reported significantly more trouble concentrating when they were told they received placebo compared to those who were told they received caffeine, p = .001. There was no significant difference between participants that received placebo.

Caffeine withdrawal symptoms at 30-minutes post gum administration were analyzed the same as mood and craving, controlling for baseline. There was a main effect of Gum Told, $F(1,60) = 6.62$, $p = .013$, $\eta^2_p = .099$, indicating that caffeine withdrawal symptoms in the told placebo condition ($M = 30.23$, $SE = 1.13$) were significantly higher than caffeine withdrawal symptoms for those who were told they had caffeine-containing gum ($M = 26.10$, $SE = 1.14$; see Figure 3.2). The effect for Gum Received was not significant, $F(1,60) = .05$, $p = .829$, $\eta^2_p = .001$, nor was there a significant interaction, $F(1,61) = 1.56$, $p = .217$, $\eta^2_p = .025$.

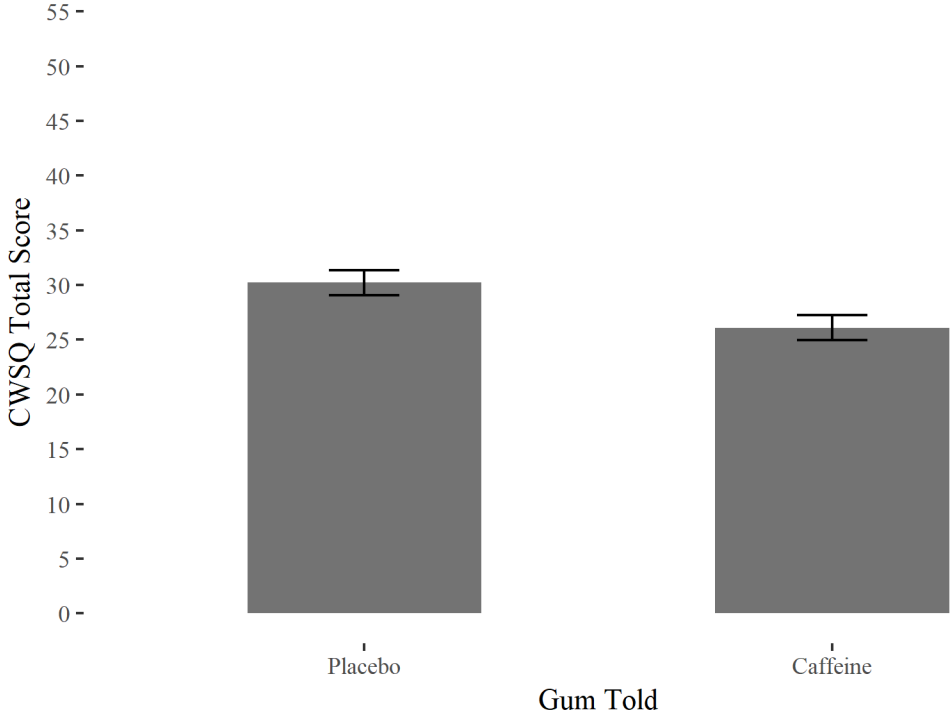

Figure 3.2 Estimated marginal mean total scores (±standard error (SE)) for Caffeine Withdrawal Symptom Questionnaire (CWSQ) 30 minutes post gum administration, covarying for baseline scores. Participants who were told they received caffeinated gum reported significantly less caffeine withdrawal symptoms than those who were told placebo, $p = .013$.

Maximum and average heart rates at 30-minutes post gum administration were analyzed in a similar fashion. There were no significant main effects nor interactions for heart rates.

3.1.3 Visual Cues

Mixed 2 x 2 x 2 ANOVAs were conducted to examine the effects of the visual cues on mood, craving, withdrawal symptoms, and heart rate. The ANOVAs had two between-subjects factors, Gum Told (told caffeine, told placebo) and Gum Received (received caffeine, received placebo), and one within-subjects factor, Cues (caffeine visual cues, neutral visual cues). There were significant main effects of Cues for both 'relaxed', $F(1,61) = 8.18$, $p = .006$, $\eta^2_p = .118$, and 'pleasant', $F(1,61) = 8.24$, $p = .006$, $\eta^2_p = .119$. Participants reported feeling more relaxed after viewing the neutral visual cues ($M = 6.11$, $SE = .25$) when compared to viewing the caffeine visual cues ($M = 5.51$, $SE = .29$). Similarly, participants reported feeling more pleasant after viewing the neutral cues ($M = 5.96$, $SE = .26$) when compared to viewing the caffeine cues ($M = 5.44$, $SE = .28$).

A couple interactions were also found for subjective ratings of mood. There was a significant interaction of Gum Told by Cues for 'stimulated', $F(1,61) = 8.30$, $p = .005$, $\eta^2_p = .120$, and 'jittery', $F(1,61) = 4.55$, $p = .037$, $\eta^2_p = .069$. When participants were told they consumed placebo gum, they felt more stimulated following the caffeine visual cues ($M = 3.42$, SE = .38) relative to the neutral visual cues ($M = 3.04$, $SE = .36$); when participants were told they consumed caffeinated gum, the felt more stimulated following the neutral visual cues ($M = 4.06$, $SE = .37$) relative to the caffeine visual cues ($M = 3.72$, $SE = .39$). Regarding the second interaction, participants in the told placebo

condition reported feeling more jittery after viewing the neutral cues (M = 1.70, SE = .30) compared to the caffeine cues (M = 1.49, SE = .24); participants in the told caffeine condition felt more jittery following the caffeine visual cues (M = 2.47, SE = .25) as opposed to the neutral visual cues (M = 2.25, SE = .30).

Caffeine craving had both a main effect of Cues, $F(1,61) = 11.78$, $p = .001$, $\eta^2_p = .162$ (see Figure 3.3), and a Gum Received by Cues interaction, $F(1,61) = 6.16$, $p = .016$, $\eta^2_p = .092$. For the main effect of Cues, participants reported higher caffeine craving following the caffeine visual cues (M = 6.31, SE = .34) compared to the neutral visual cues (M = 5.84, SE = .35). This main effect was further broken down in the Gum Received by Cues interaction: participants who received caffeinated gum had little change in caffeine craving between the neutral visual cues (M = 6.21, SE = .49) and caffeine visual cues (M = 6.34, SE = .48); however, those who received placebo gum had an increase in caffeine cravings from the neutral visual cues (M = 5.47, SE = .50) to the caffeine visual cues (M = 6.28, SE = .48). Similarly, coffee craving had both a main effect of Cues, $F(1,61) = 18.02$, $p < .001$, $\eta^2_p = .228$ (see Figure 3.3), and a Gum Received by Cues interaction, $F(1,61) = 4.68$, $p = .034$, $\eta^2_p = .071$. Just as with caffeine craving, participants reported higher coffee craving after viewing the caffeine cues (M = 7.60, SE = .31) as compared to the neutral cues (M = 7.04, SE = .31). Similarly, this main effect was further explained by the Gum Received by Cues interaction: participants who received caffeinated gum had little change in coffee craving from the neutral visual cues (M = 7.28, SE = .44) to the caffeine visual cues (M = 7.55, SE = .43); participants who received placebo gum had an increase in coffee craving from neutral visual cues (M = 6.81, SE = .44) to caffeine visual cues (M = 7.66, SE = .43).

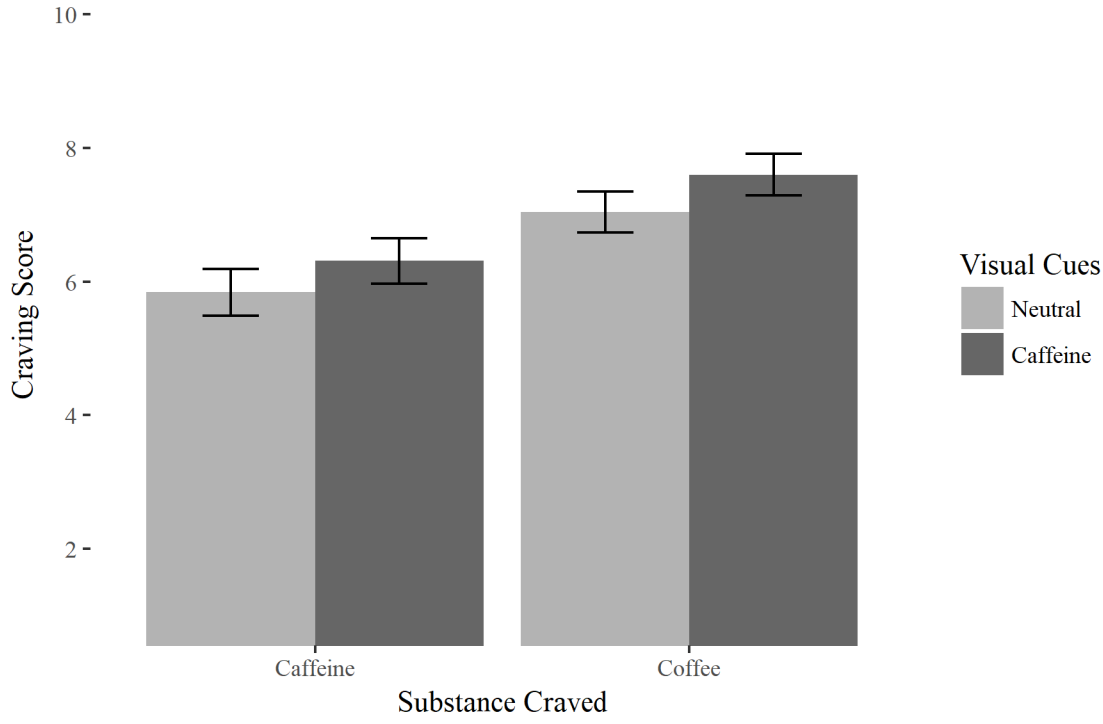

Figure 3.3 Estimated marginal means (±standard error (SE)) for subjective ratings of craving. Following caffeine visual cues, participants rated both caffeine craving and coffee craving higher when compared to ratings post neutral visual cues, *ps* ≤ .001.

For maximum heart rate, there was a main effect of Cues, $F(1,61) = 10.99$, $p = .002$, $\eta^2_p = .153$ (see Figure 3.4); this indicates that participants maximum heart rate was significantly higher when viewing the caffeine cues ($M = 82.89$, $SE = 1.43$) as compared to when they were viewing the neutral cues ($M = 79.88$, $SE = 1.46$). For average heart rate, there was both a significant main effect of Cues, $F(1,61) = 10.70$, $p = .002$, $\eta^2_p = .149$, as well as a Gum Told by Gum Received by Cues interaction, $F(1,61) = 4.76$, $p = .033$, $\eta^2_p = .072$. The average heart rate increased from the neutral visual cues to the caffeine visual cues across all conditions, however, the change was only significant in participants that had matching Gum Told and Gum Received conditions (see Table 3.1).

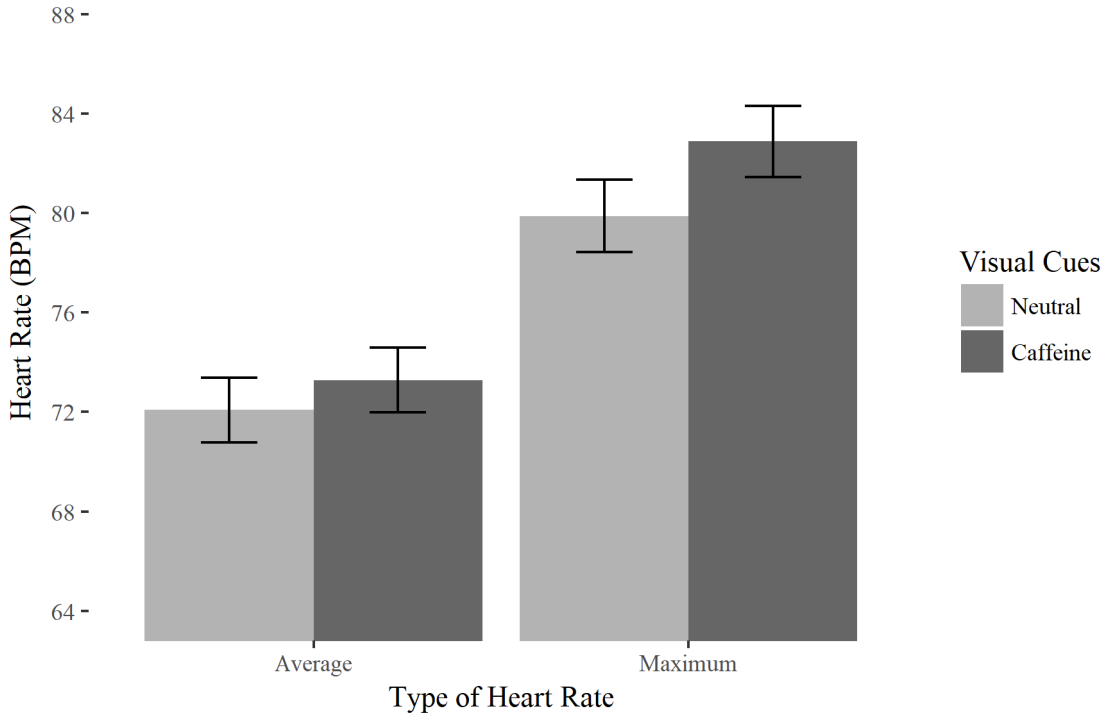

Figure 3.4 Estimated marginal means (±standard error (SE)) for average and maximum heart rates in beats per minute (BPM). During caffeine visual cues, participants had elevated average and maximum heart rates when compared to their heart rates during neutral visual cues, *ps* = .002.

Table 3.1 Average heart rate (in BPM), standard error (*SE*; in parentheses), and significance of mean difference for participants during neutral and caffeine visual cues.

Condition	Neutral cues	Caffeine cues	Significance
Told caffeine/received caffeine	71.81 (2.64)	73.81 (2.61)	.009
Told caffeine/received placebo	72.56 (2.64)	73.13 (2.61)	.449
Told placebo/received caffeine	70.71 (2.56)	70.94 (2.54)	.744
Told placebo/received placebo	73.25 (2.64)	75.25 (2.61)	.009

3.1.4 Auditory/Olfactory Cues

Mixed 2 x 2 x 2 ANOVAs were also used to examine the effects of the combined auditory and olfactory cues on mood, craving, withdrawal symptoms, and heart rate. The ANOVAs had two between-subjects factors, Gum Told (told caffeine, told placebo) and Gum Received (received caffeine, received placebo), and one within-subjects factor, Cues (caffeine visual cues, neutral visual cues). There were main effects of Gum Told for 'head rush', $F(1,60) = 5.51$, $p = .022$, $\eta^2_p = .084$, and 'jittery', $F(1,60) = 4.12$, $p = .047$, $\eta^2_p = .064$ (see Figure 3.5). Participants in the told caffeine condition ($M = 2.97$, $SE = .37$) reported more head rush than those in the told placebo condition ($M = 1.73$, $SE = .37$). Similarly, participants in the told caffeine condition ($M = 2.72$, $SE = .30$) reported more jitteriness than those in the told placebo condition ($M = 1.85$, $SE = .30$).

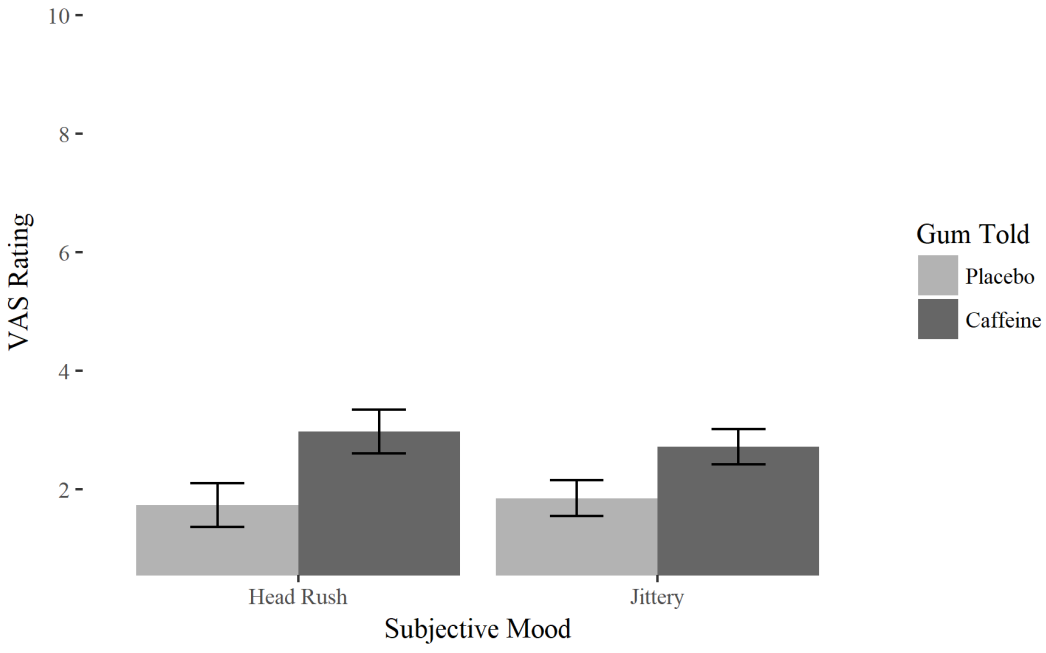

Figure 3.5 Estimated marginal means (±standard error (SE)) for subjective ratings of 'head rush' and 'jittery' during auditory/olfactory cues. Participants who were told they received caffeine rated feelings of head rush and jitteriness higher than those who were told placebo, *ps* < .05.

There was a significant Gum Told by Gum Received by Cues interaction for subjective ratings of 'relaxed', $F(1,60) = 6.01$, $p = .017$, $\eta^2_p = .091$. Participants reported no significant differences in ratings of relaxedness between the neutral and caffeine auditory/olfactory cues except in the told caffeine/received placebo condition: participants in this condition rated their relaxedness significantly lower in following the caffeine auditory/olfactory cues ($M = 6.00$, $SE = .59$) when compared to the neutral auditory/olfactory cues ($M = 6.75$, $SE = .53$). For participant ratings of 'stimulated', the was both a main effect of Cues, $F(1,60) = 5.02$, $p = .029$, $\eta^2_p = .077$, and a Gum Told by Cues interaction, $F(1,60) = 4.06$, $p = .048$, $\eta^2_p = .063$. Overall, participants reported feeling more stimulated following the caffeine auditory/olfactory cues ($M = 4.07$, $SE = .29$) when compared to the neutral auditory/olfactory cues ($M = 3.53$, $SE = .26$). However, upon review of the interaction, this effect appears mainly for participants in the told caffeine group. Participants in the told placebo condition reported no difference in stimulation between the neutral auditory/olfactory cues ($M = 3.34$, $SE = .37$) and the caffeine auditory/ olfactory cues ($M = 3.40$, $SE = .41$), whereas participants in the told caffeine condition reported significantly more stimulation following the caffeine auditory/olfactory cues ($M = 4.75$, $SE = .41$) when compared to the neutral auditory/ olfactory cues ($M = 3.72$, $SE = .37$).

Coffee craving had a significant main effect of Cues, $F(1,60) = 8.42$, $p = .005$, $\eta^2_p = .123$; participants reported higher levels of coffee craving following the caffeine auditory/olfactory cues ($M = 7.93$, $SE = .31$) when compared to the neutral auditory/ olfactory cues ($M = 7.59$, $SE = .32$). For caffeine craving, there were no main effects; however, there was a significant Gum Received by Cues interaction, $F(1,60) = 4.90$, $p = .031$, $\eta^2_p = .075$. Participants in the received placebo condition showed an increase in

caffeine craving from neutral auditory/olfactory cues (M = 6.33, SE = .48) to caffeine auditory/olfactory cues (M = 6.78, SE = .48); conversely, participants in the received caffeine condition showed a non-significant decrease in caffeine craving from neutral auditory/olfactory cues (M = 6.58, SE = .46) to caffeine auditory/olfactory cues (M = 6.40, SE = .47).

For maximum heart rate, there was again a main effect of Cues, $F(1,60) = 25.72$, $p < .001$, $\eta^2_p = .300$; this indicates that participants maximum heart rate was significantly higher during the caffeine auditory/olfactory cues (M = 82.68, SE = 1.43) than during the neutral auditory/olfactory cues (M = 77.43, SE = 1.53). For average heart rate, there was both a significant main effect of Cues, $F(1,60) = 10.07$, $p = .002$, $\eta^2_p = .144$, as well as a Gum Told by Gum Received by Cues interaction, $F(1,60) = 5.35$, $p = .024$, $\eta^2_p = .082$. Overall, there was an increase in heart rate from the neutral auditory/olfactory cues (M = 70.87, SE = 1.37) to the caffeine auditory/olfactory cues (M = 72.68, SE = 1.29). However, after analyzing the interaction, this increase is only significant in the told placebo/received caffeine condition; in all other conditions, there no differences in average heart rate (see Figure 3.6).

3.1.5 Coffee Self-Administration

Coffee self-administration was compared between groups via a two-way ANOVA having two levels of Gum Told (told placebo, told caffeine) and two levels of Gum Received (received placebo, received caffeine). There were no significant main effects nor an interaction when analyzing the number of coffee units consumed. Follow-up analyses were conducted, investigating whether there was a relationship between group condition and participants' choice to consume any versus no units of coffee. There was a significant difference in choice to drink between participants who received

caffeine and those that received placebo, $X^2(1, N = 64) = 8.45$, $p = .004$. For participants in the received caffeine condition, 15 of 33 (45.5%) chose to consume at least one unit of coffee; 25 of 31 (80.6%) of participants in the received placebo condition chose to consume at least one unit of coffee.

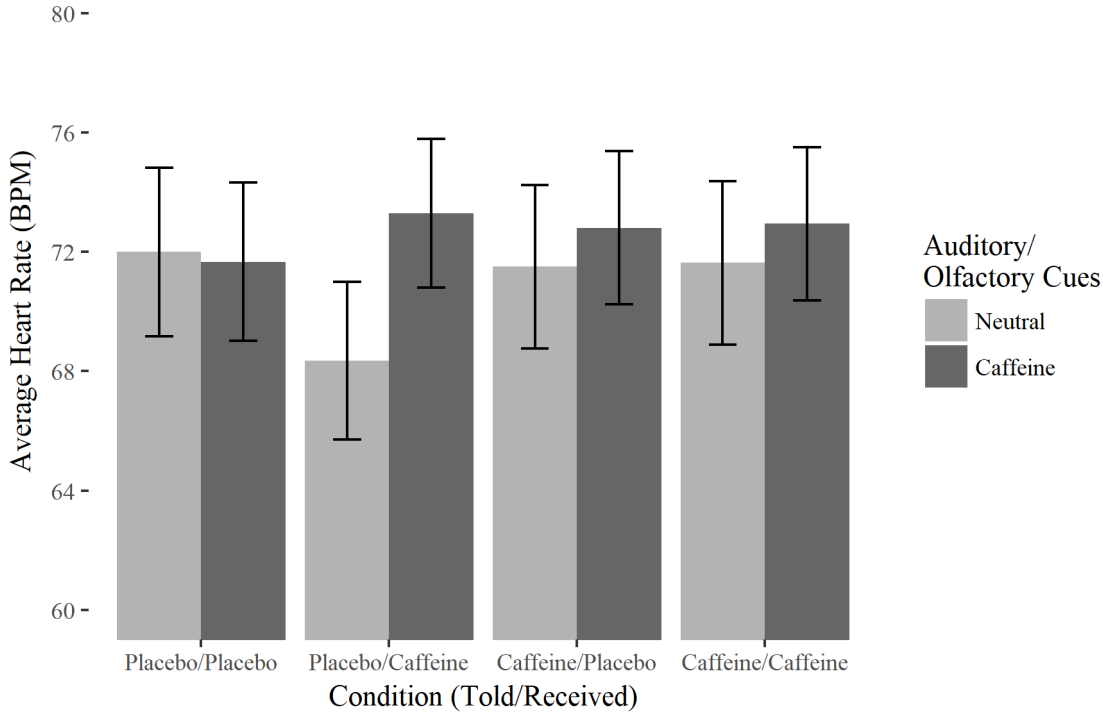

Figure 3.6 Estimated marginal means (±standard error (SE)) for average heart rates during auditory/olfactory cues. There was an overall increase in average heart rate between neutral and caffeine auditory/olfactory cues, however, this increase was only significant for those in the told placebo/received caffeine condition, $p < .001$.

3.2 Results for Believers

3.2.1 Instruction Manipulation Check

At the conclusion of the study sessions, 11 of 65 participants (16.9%) were found

to not believe or were unsure of the information they received pertaining to the caffeine content of the gum. As previous studies have shown that participants who believe instructions can have different findings when compared to all participants, the previous tests were run again with only believers included in the analyses. All four conditions had at least one participant removed; notably, the told caffeine/received placebo had the most participants removed for non-belief, at 5 participants. For a breakdown of believers by group, see Table 3.2.

Table 3.2 Number of participants that believed the information they received pertaining to the caffeine content of their gum.

Condition	Original *n*	Believer *n*	Believer %
Told caffeine/received caffeine	16	15	93.8
Told caffeine/received placebo	16	11	68.8
Told placebo/received caffeine	17	14	82.4
Told placebo/received placebo	16	14	87.5

3.2.2 Expectancy and Drug Effects Post Gum Administration

With regard to expectancy and drug effects post gum administration, all of the significant results that were found for all participants remained when only the believers were examined. There was, however, an additional main effect found: believers reported a significant main effect of Gum Told for 'head rush', $F(1,49) = 5.24$, $p = .026$, $\eta^2_p = .097$ (see Figure 3.7). Believers in the told caffeine condition ($M = 2.89$, $SE = .34$) had more head rush 30-minutes post gum administration when compared to believers in the told placebo condition ($M = 1.80$, $SE = .33$). No other differences between believers and all participants were found at this time point.

3.2.3. Visual Cues

The effects of neutral and visual cues were re-examined after removal of non-believers. When focusing only on believers, all the main effects of Cues remained, as did the Gum Told by Cues interaction for 'stimulated' and the Gum Received by Time interaction for 'crave caffeine'. The interactions for 'jittery' (Gum Told x Cues; $F(1,50) = 3.10$, $p = .085$, $\eta^2_p = .058$), 'crave coffee' (Gum Received x Cues; $F(1,50) = 2.31$, $p = .135$, $\eta^2_p = .044$), and average heart rate (Gum Told x Gum Received x Cues; $F(1,50) = 3.91$, $p = .054$, $\eta^2_p = .073$) no longer achieved significance. There were no new results when the tests were conducted using only the believers' data.

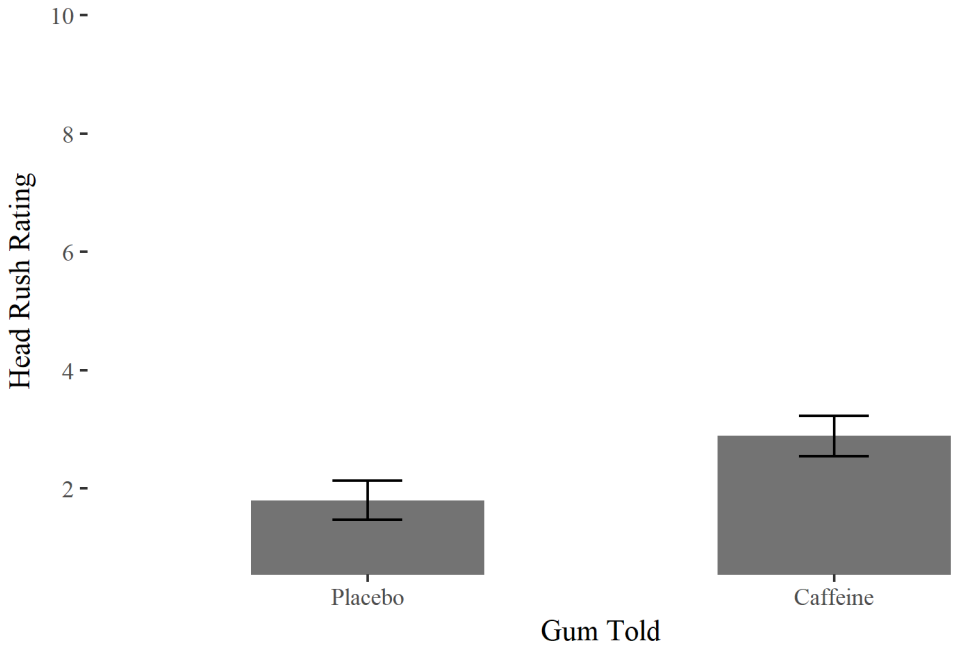

Figure 3.7 Estimated marginal means (±standard error (SE)) for subjective ratings of 'head rush' 30 minutes post gum administration, focusing only on believers (participants who believed the instructions regarding gum content) and covarying for baseline ratings. Participants who were told they received caffeine felt significantly more head rush those who were told placebo, $p = .026$.

3.2.4. Auditory/Olfactory Cues

Finally, the combined auditory and olfactory cue effects were re-investigated, using responses from believers only. All main effects remained significant, as did the Gum Received by Cues interaction for 'crave caffeine' and the Gum Told by Gum Received by Cues interaction for 'relaxed' and average heart rate. The Gum Told by Cues interaction for 'stimulated' ($F(1,50) = 2.54$, $p = .117$, $\eta^2_p = .048$) no longer achieved significance. However, there were two new findings when focusing only on the believers. Believers reported a significant main effect of Cues for 'jittery', $F(1,50) = 4.37$, $p = .042$, $\eta^2_p = .080$; this meant that believers felt slightly more jittery in following the caffeine auditory/olfactory cues ($M = 2.47$, SE = .22) when compared to the neutral auditory/olfactory cues ($M = 2.14$, $SE = .27$). Lastly, believers reported a Gum Told by Gum Received interaction for 'pleasant', $F(1,49) = 7.99$, $p = .007$, $\eta^2_p = .140$. Believers in the told placebo condition rated pleasantness higher when they actually received caffeinated gum ($M = 6.81$, $SE = .49$) than when they received placebo gum ($M = 5.91$, $SE = .47$); contrarily, believers in the told caffeine condition rated pleasantness higher when they actually received placebo gum ($M = 6.32$, $SE = .53$) as opposed to caffeinated gum ($M = 4.77$, $SE = .46$).

3.2.5 Gum Liking and Coffee Self-Administration

For both gum liking and coffee self-administration, removing non-believers from the sample had no effect on the results. A significant main effect of Gum Received remained for gum liking, $F(1,50) = 21.60$, $p < .001$, $\eta^2_p = .302$, as did the significant difference of choice to consume any coffee between participants who received caffeine versus placebo, $X^2(1, N = 54) = 6.99$, $p = .008$.

CHAPTER 4: DISCUSSION

4.1 Overview of Main Findings

The current study aimed to investigate the concurrent roles of cue reactivity and expectancy on caffeine craving and withdrawal. Based on the literature, we hypothesized that expectancy effects would lessen, but not eliminate, caffeine craving and withdrawal. We also hypothesized that we would find evidence of caffeine cue reactivity; we expected subjective caffeine craving, withdrawal, and heart rate would increase following the presentation of caffeine cues.

We found partial support for our first hypothesis. Following gum administration, total caffeine withdrawal scores were significantly lower for participants who were told they received caffeine, compared to participants who were told they received placebo gum. However, this was the only significant finding for withdrawal symptoms throughout the entire study – there were no difference between groups after any of the cues. This suggests that expectancy removed negative aspects of caffeine withdrawal, but this effect was temporary. With regard to caffeine and coffee craving, there were no significant effects of expectancy – in this aspect, the data failed to support the first hypothesis. These results contrast with findings of a recently published report. In that study, individuals who were told they consumed caffeinated coffee, whether they received caffeine or not, showed significant decreases in caffeine craving but not in withdrawal symptoms (Juliano et al., 2019). However, an earlier study with a longer abstinence period (24 hours) found that expectancy decreased both caffeine withdrawal symptoms and caffeine craving (Mills et al., 2016). It's possible that a longer abstinence period would have produced the same effects in all three studies. Additionally, caffeine

was administered via caffeinated gum, which was not the typical route of administration for any participant; all participants administered their daily caffeine via coffee. Expectancy is based on a combination of criteria: verbal instructions regarding active drug content and dose, anticipated effects of the substance, and also past experience. Though the gum and verbal instructions given at the time of administration adequately addressed the first two criteria, the lack of participant experience with caffeinated gum may have muted the expectancy effect.

There were other findings related to expectancy, specifically the addition of feelings related to consuming caffeine whether or not caffeine was consumed. After gum administration, participants reported feeling stimulated and having more head rush when they were told they consumed caffeinated gum, compared to those who were told they received placebo. Later in the study, during the auditory/olfactory cues, this addition of subjective responses continued: participants in the told caffeinated gum condition felt more head rush and more jittery than those who were in the told placebo condition. To our knowledge, we are the first study to examine these specific feelings related to caffeine use. Also, of note, participants felt more stimulated following the presentation of the combined auditory/olfactory cues; it is probable that this is due to the scent of the coffee. A recent study has shown that simply smelling a coffee-like scent increased participants expected physiological arousal (Madzharov, Ye, Morrin, & Block, 2018). In our study, feeling stimulated would be a type of physiological arousal, showing support for the idea put forth in the previous study.

Regarding our second hypothesis, we also found support for caffeine cue reactivity in both sensory modalities. Craving (both caffeine and coffee) and heart rate both increased from neutral to caffeine visual cues. For auditory/olfactory cues, coffee

craving and heart rate again increased from neutral to caffeine cues; for those who received placebo gum, caffeine craving also increased from neutral to caffeine cues. Both of these findings lend support to our second hypothesis. However, the cues had no effect on caffeine withdrawal symptoms. As this is the first study to show the existence of caffeine cue reactivity, replication in future studies is essential. It would also be important to investigate the combined effect of visual, auditory, and olfactory modalities to see if there is an additive effect.

4.2 Implications

It has long been reported that caffeine is only mildly reinforcing in humans (Ferré, 2016; Nehlig, 1999). Caffeine appears to be reinforcing at only low or moderate amounts – at high doses, caffeine becomes aversive (Nehlig, 1999). As such, studies on rats have shown that typical daily amounts of caffeine are not enough to activate the dopamine mesolimbic circuit in the brain, also known as the reward pathway of the brain (Acquas, Tanda, & Di Chiara, 2002; De Luca, Bassareo, Bauer, & Di Chiara, 2007). Similarly, this has also been shown in humans – one such study showed that a moderate amount of caffeine (3mg/kg of body weight) resulted in activation of brain regions involved in attention, vigilance, and anxiety, but not in areas of reinforcement and reward (Nehlig, Armspach, & Namer, 2010). If it is true that caffeine consumption does not lead to increased dopamine in the mesolimbic reward circuit, then this is the first study that shows cue reactivity for a substance that does not activate this circuit.

Although there were significant findings related to caffeine cue reactivity, the changes from neutral to caffeine cues were quite small. This could be due to a number of factors. First, as mentioned above, we examined cue reactivity via different sensory

modalities. To our knowledge, most (if not all) studies examining drug cue reactivity utilize one sensory modality at a time. It is possible that combining multiple sensory modalities would have a greater impact on cue reactivity. Recent studies have shown that it is possible to have greater cue reactivity with other combination of cues, such as combining pictures of objects and environments (Conklin, et al., 2019); this would most likely work with combinations of senses. This ties into the second factor: although the cues are eliciting craving and increased heart rate, they would most likely be more effective if they were more similar to real life. Typically, when cues are encountered in real life, there are numerous cues working in concert. If an individual were to walk by a coffee shop, they would be impacted by the sights, sounds, and smells of coffee, even the other people in the coffee shop. This atmosphere is more likely to produce a stronger reaction than cues presented in a laboratory (Shiffman, et al., 2015). Thus, though we were able to elicit cue reactivity in our study, albeit smaller in magnitude, responses to cues in every day life may be stronger. For the coffee drinker that wants to cut back, the ubiquity of coffee sights and smells may make it difficult to curb the habit. Finally, the pharmacokinetics of caffeine could yield a third possible explanation for the subdued cue reactivity findings. Typically, caffeine (when administered via coffee or gum) reaches peak blood plasma concentration after approximately one hour (Kamimori et al., 2002). This is leads to a delayed onset of the effects of caffeine, when compared to other drugs of abuse; for example, nicotine, delivered via cigarettes, reaches peak blood plasma concentration after just four minutes (Hajek, Przulj, Phillips, Anderson, & McRobbie, 2017). Because of this delayed onset, caffeine-related cues may not form a strong connection with the physiological effects of caffeine. For example, if an individual orders their coffee from a coffee shop and drinks it on the way to work, the physiological

effects of the caffeine will take affect after the individual is at work; the stimuli in the coffee shop are no longer present, so they do not become strong, salient cues for the effects of caffeine. In other drugs of abuse, the effects are more immediate, which result in stronger cues.

In this study, there was an unanticipated lack of pharmacological effects of caffeine. One possible explanation for this is the amount of caffeine administered to participants. The average participant consumed over 400mg of caffeine per day; frequently, participants consumed their daily caffeine before noon, via successive cups of coffee. The amount of caffeine in each piece of gum (100mg) was approximately one-quarter of a participant's typical intake. It is possible that the small amount of caffeine administered led to a lack of findings regarding the pharmacological effects of caffeine. Participants presumably had some level of caffeine tolerance, so 100mg of caffeine would have had a muted effect on their system. However, this could also be because the pharmacological effects of caffeine are much more subtle when compared to other drugs (especially other stimulants); this is even more true at a small amount like 100mg (Juliano et al., 2019). Nevertheless, caffeine administration did appear to have an impact on whether participants consumed any coffee in the final portion of the study. This shows that even a small amount of caffeine is able to affect further consumption, whether the individual was aware of the caffeine or not.

4.3 Limitations

This study is not without its limitations. First, it is possible that the 18-hour abstinence period was too short, which in turn led to a lack of findings regarding caffeine withdrawal. Other studies that used the CWSQ to measure caffeine withdrawal

symptoms had abstinence periods similar to ours, at 16 hours (Juliano et al., 2012; Juliano et al, 2019). Also, the half-life of caffeine is estimated to be approximately 6 hours (White et al., 2016), so allowing three half-lives to pass should have eliminated most caffeine from participants' systems. However, peak withdrawal symptoms typically occur in the 20-51 hour range (Juliano & Griffiths, 2004). It is possible that we missed the critical period with at least some of our participants. Secondly, participants in this study all reported above-average daily caffeine use. As such, the findings may not generalize to individuals who consume only a small amount of caffeine per day.

Thirdly, caffeine abstinence was confirmed by self-report only. Although we informed participants during telephone that we may or may not take a saliva sample to confirm abstinence (to increase the chance of compliance), we had no way to ensure compliance. This could have resulted in self-report bias when reporting withdrawal symptoms if a participant wanted us to believe they had complied with instructions. Finally, craving was assessed via two single-item questions. Research has shown that craving is better captured by multi-item questionnaires that examine multiple aspects of craving (Tiffany & Wray, 2014); however, at this point in time, no valid and reliable questionnaire exists for caffeine. Future studies should examine a more robust measure of caffeine craving.

4.4 Conclusions

In conclusion, our research demonstrates the first known example of caffeine cue reactivity. As such, this study also shows the first example of cue reactivity by a substance that does not have a major affect on the mesolimbic dopamine pathway. This study also confirms the expectancy effects of caffeine, although there is not a strong

connection between caffeine expectancy effects and cue reactivity.

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APPENDIX A: EXPLORATORY ANALYSES

We also wanted to examine sex differences for each of these tests; however, as the group size is greatly reduced when adding in sex as a variable, we included these analyses as exploratory only. See Table A.1 for a breakdown of groups. This appendix is a summary of the main results after re-running the initial tests while including sex as a variable.

Table A.1 Group sizes when including Gum Told, Gum Received, and Sex as variables.

	Females		Males	
Condition	Told Caffeine	Told Placebo	Told Caffeine	Told Placebo
Received Caffeine	7	8	9	9
Received Placebo	8	7	8	9

First, there were no key findings that differed from our main analyses when examining gum liking or any of the analyses related to the visual cues. Ratings of mood and craving, withdrawal symptoms, and heart rate 30 minutes post gum administration were analyzed using a factorial ANCOVA, having two levels of Gum Told (caffeine, placebo), two levels of Gum Received (caffeine, placebo), and two levels of Sex (male, female), controlling for baseline ratings. For ratings of 'stimulated', there was both a significant main effect of Sex, $F(1,55) = 6.50$, $p = .014$, $\eta^2_p = .106$, and a significant Sex by Gum Received interaction, $F(1,55) = 6.98$, $p = .011$, $\eta^2_p = .113$. Overall, males reported feeling more stimulated 30 minutes post gum administration ($M = 4.59$, $SE = .30$) when compared to females ($M = 3.43$, $SE = .34$). When examining this relationship in the interaction, this difference only occurs in the received caffeine condition – there is

no difference between males and female in feelings of stimulation in the received placebo condition (see Figure A.1). For maximum heart rate, there was a significant Gum Told by Sex interaction, $F(1,56) = 4.96$, $p = .030$, $\eta^2_p = .081$. For males, maximum heart rates were significantly lower when they were told they received caffeine ($M = 78.60$, $SE = 1.59$) as opposed to placebo ($M = 84.06$, $SE = 1.55$), $p = .016$; for females, there was no difference between conditions. There were no other key sex differences 30 minutes post gum administration.

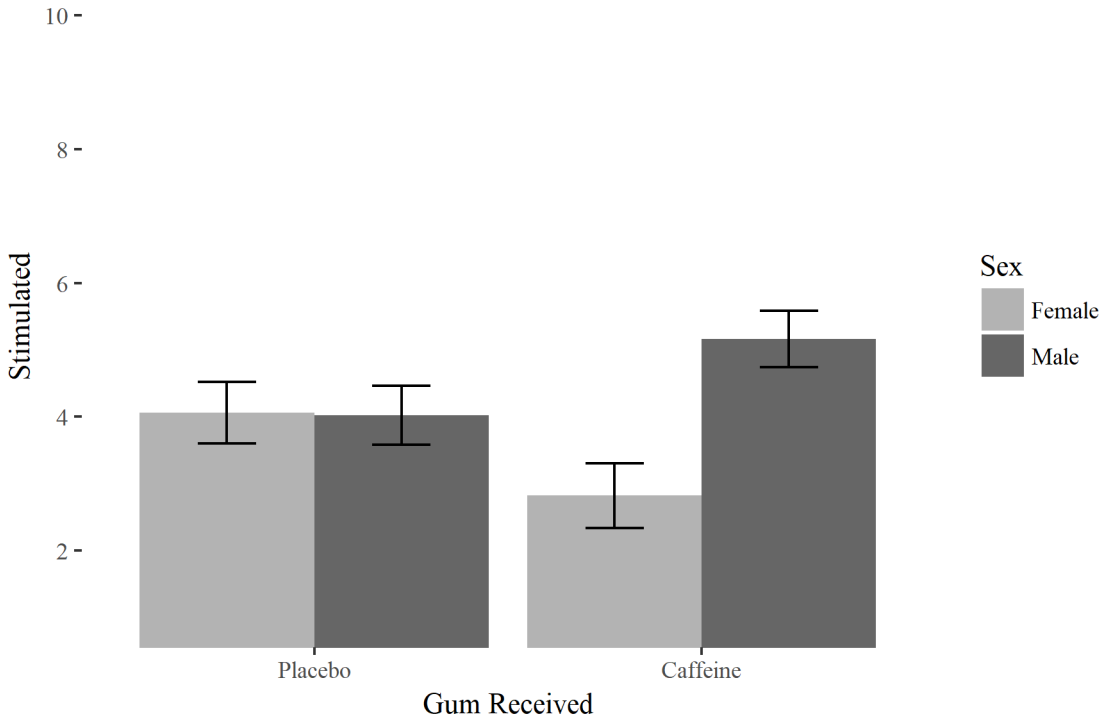

Figure A.1 Estimated marginal means (±standard error (SE)) for subjective ratings of 'stimulated' 30 minutes post gum administration, covarying for baseline ratings. Males who received caffeinated gum reported feeling significantly more stimulated than females who received caffeinated gum, $p = .001$. There was no significant difference between males and females who received placebo.

For mood, craving, withdrawal, and heart rate differences between auditory/ olfactory cues, a mixed 2 X 2 X 2 X 2 ANOVA was conducted. Cues (neutral, caffeine) was included as a within-subjects factor, whereas Gum Told (placebo, caffeine), Gum Received (placebo, caffeine), and Sex (male, female) were included as between-subjects factors. Out of these analyses, there were two key findings regarding withdrawal symptoms: there was a main effect of Sex, $F(1,54) = 4.98$, $p = .030$, $\eta^2_p = .084$, and a significant Cues by Sex interaction, $F(1,54) = 6.92$, $p = .011$, $\eta^2_p = .114$. Overall, at this stage of the study, females ($M = 35.37$, $SE = 2.63$) reported significantly higher withdrawal symptoms than males ($M = 27.61$, $SE = 2.27$). However, the males' withdrawal symptoms following the caffeine auditory/olfactory cues ($M = 26.16$, $SE = 2.24$) were significantly lower than their withdrawal symptoms following the neutral cues ($M = 29.06$, $SE = 2.38$), $p = .002$; for females, there was no difference in withdrawal symptoms between cues (see Figure A.2). There were no other key findings regarding sex differences during the auditory/olfactory cues.

In our main results section, we found no main effects or interactions for units of coffee consumed. However, after conducting the analysis again, this time including Sex (female, male) as a factor, we found a three-way interaction between Gum Told, Gum Received, and Sex, $F(1,56) = 8.475$, $p = .005$. For females, average coffee self-administration was less than one unit for each condition except the told placebo/received placebo condition; participants in the told placebo/received had an average of two units of coffee during the self-administration period. For males, having matched Gum Told and Gum Received conditions resulted in fewer units of coffee consumed compared to unmatched conditions (see Table A.2).

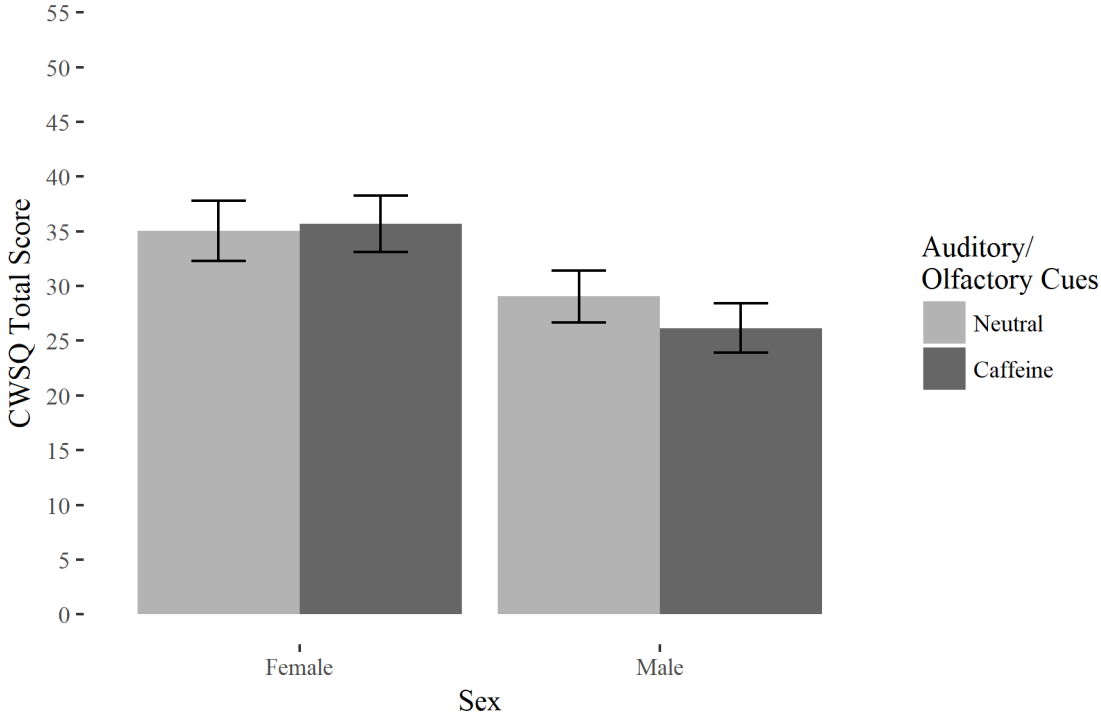

Figure A.2 Estimated marginal means (±standard error (SE)) for Caffeine Withdrawal Symptom Questionnaire (CWSQ) following the auditory/olfactory cues. Males reported fewer caffeine withdrawal symptoms following caffeine cues as compared to neutral cues, $p = .002$. Females reported no significant difference in withdrawal symptoms between caffeine and neutral cues.

Table A.2 Estimated marginal means (SE) for units of coffee consumed (up to 3) in each condition when including sex as a factor.

	Females		Males	
Condition	Told Caffeine	Told Placebo	Told Caffeine	Told Placebo
Received Caffeine	0.86 (.42)	0.63 (.45)	0.56 (.37)	1.78 (.37)
Received Placebo	0.88 (.39)	2.00 (.45)	1.88 (.39)	1.22 (.37)