

The effects of nicotine dependency on eye movements elicited by visual smoking cues

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# ABSTRACT

Previous research has shown that smokers have attentional biases towards smoking-related cues which are related to craving and the affective and motivational valence of stimuli (Mogg, Bradley, Field & Houwer, 2003). The current study used non-smokers (N=23), low dependent smokers (N=15) and heavily dependent smokers (N=15), to explore the effects of nicotine dependency on attentional biases, as indicated by eye movements (EM), towards smoking related cues. EMs were measured, using an eye tracking device, to smoking related and control cues contained within images when the images were presented simultaneously. The number of fixations, gaze duration and time to first fixation were analysed. The results revealed that smokers with high levels of nicotine dependence showed a significantly greater attentional bias, as indicated by observation length, and attended more, as indicated by fixation count, to smoking-related cues than nonsmokers. Moreover, smokers with low levels of nicotine dependence showed an enhanced attentional bias, and attended more to smokingrelated cues than non-smokers. This indicates that attentional biases for drug-related cues are functionally related to incentive processes, with severity of nicotine dependence being a positive predictor of such biases.

KEY WORDS:	SMOKING	ATTENTIONAL BIAS	EYE MOVEMENTS	DRUG CUES	NICOTINE DEPENDENCY
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#### Introduction

Drug dependence has profound psychological, physical and economic consequences. It is deeply complex and typically involves the uncontrollable behaviour pattern of drug seeking and drug taking behaviour which occurs at the expense of most other activities, even when the damaging health and social effects are known (West, 2006). Extensive research has shown that reactivity to drug-related cues is intrinsically linked to drug dependence (Field & Cox, 2008). This is thought to play an essential role in the maintenance of drug dependence as well as in drug relapse following attempts to quit. Theorists have more recently argued that biases in cognitive processing of drug-related cues are a crucial part of drug cue reactivity which has largely been overlooked. This is essential to understand when considering the effects of the exposure of drug cues on drug-seeking and craving (Field & Cox, 2008) and also of clinical importance when trying to prevent future relapse (Mogg, Field & Bradley, 2005).

Nicotine dependent individuals have reliably been shown to preferentially direct their attention to smoking-related cues at the expense of other cues (Mogg et al., 2005). For example, Sayette and Hufford (1994) found that when an auditory probe was presented to smokers, in the presence of smoking-related cues, they were slower to respond than when neutral cues were presented. Furthermore, Waters and Feyerband (2000) reported that in a modified stroop task, deprived smokers were slower at naming the colours of smoking-related words than control words. This suggests that smokers have problems ignoring drug-related stimuli. Similar findings have been reported in heroin dependence where heroin dependent participants showed considerable attentional biases for supraliminally presented heroin cues (Franken, Kroon, Wiers & Jansen, 2000).

However, these studies fail to reveal the precise nature of the cognitive processes which drug-related cues interfere with (Mogg et al., 2003). The effects demonstrated in the modified stroop were assumed to be the result of selective attention to salient information. However, they could have arisen from several other mechanisms, for example from competition for processing resources from task irrelevant processes triggered by the cues. Alternatively, Williams, Mathews and MacLeod (1996 as cited in Mogg et al., 2003) suggest from competition which occurs at later response output stages of processing. The attentional biases may therefore not reflect a selective attentional bias for drug-related cues (Mogg et al., 2003).

Attentional biases are of theoretical importance. Robinson and Berridge (2001) postulated that the incentive-salience mechanism, controlled by the level of dopamine in the mesolimbic dopamine system, mediates attentional biases for drug-related cues. The theory proposes that as addiction develops, the pleasure of the drug becomes less important while the incentive motivation to use the drug increases. This results from drug induced changes in the nucleus accumbens (NAcc) related circuitry which drives incentive salience. The incentive salience is a mental representation of the stimulus which can be characterised in terms of 'wanting'. The wanting system can operate outside of conscious awareness. Taking addictive drugs causes the system to become

sensitised resulting in a greater effect when further ingestion occurs. Drug association cues therefore cause 'wanting', 'grabbing' attention, as excessive incentive salience is attributed mainly to them. The neural systems responsible for the incentive salience are dissociable from the systems which cause the hedonic effects of drugs, for example the NAcc opioid neurotransmission. It is theorised that individuals are directed to incentive stimuli through the process of classical conditioning where stimulus-stimulus associations occur (West, 2006). There is evidence to suggest that this may be the Lazev, Herzog and Brandon (1999) trained smokers to associate a novel case. conditioned stimulus, such as a visual, olfactory or auditory stimulus, with cigarette smoking and found that greater subjective craving was elicited by the latter stimulus than one which had been explicitly unpaired with smoking. Furthermore, there is evidence to suggest, as proposed by Robinson and Berridge (2001), that these stimuli acquire incentive characteristics (Field & Cox, 2008). Mucha et al. (1998, as cited in Field & Cox, 2008) found that when smokers were trained to distinguish between an auditory stimulus that was paired with smoking and another stimulus that was unpaired with smoking, most chose to experience the stimulus paired with smoking rather than the unpaired stimulus.

Balfour (2004, as cited in West, 2006) has proposed a variation on the dopamine theory of reward, primarily concentrating on nicotine addiction. The nucleus accumbens consists of the shell and the core, and Balfour (2004, as cited in West, 2006) has predicted that the dopamine projections to each play complementary roles in the development of addiction. Nicotine increases extra-synaptic dopamine in the medial shell relating to the hedonic properties of smoking which in turn increases the probability that the behaviour will be repeated. However, nicotine also causes dopamine concentrations to increase in the core and this grants incentive salience to drug-related cues leading to the development of stimulus response behaviour when those cues are presented (Balfour, 2004 as cited in West, 2006). As theorised by Robinson and Berridge (2001), this effect of dopamine levels in the accumbens core is sensitised, underpinning the development of addiction.

Alternative theories have emphasised the role of habit, instead of incentive processes, for mediating drug-taking behaviour. Tiffany (1990) has proposed that through repetition drug-taking falls under automatic cognitive control and that procedures for drug-taking behaviour are stored as action schemas. Drug-use is triggered by drugcues which activate these action schemas. Tiffany (1990) postulates that craving occurs as a result of action schemas which have been inhibited, for example through cigarette unavailability, or as a result of smoking abstinence. Thus, if drug-taking behaviour is obstructed the individual will have an increased attentional bias for drug-related cues as the individual is experiencing an increase in drug-craving (Field, Mogg, & Bradley, 2004a). Tiffany and Carter (1998, as cited in Catley, Shiffman & O'Connell, 2000) have highlighted that relapses to drug-taking often occur in the absence of craving. However, Catley et al. (2000) have shown that absent-minded relapses to smoking are rare. Recent research consistent with these theories involving visual probe tasks has shown an attentional bias for smoking-related cues in smokers but not in non-smokers (Bradley, Mogg, Field & Wright, 2003). In a visual probe task, two pictures are presented simultaneously. One picture shows a smoking-related scene, such as women inhaling on a cigarette, and the other shows a control scene, for example a woman applying lipstick. As soon as the pictures disappear, a probe appears in the position of one of the pictures. Participants are required to respond to the probe as fast as possible by pressing a key (Bradley et al., 2003). The regions of the visual display which are attended to, rather than unattended, are responded to faster (Posner, 1980).

Bradley and colleagues (2003) study indicated that smokers showed a greater awareness for smoking-related cues compared with non-smokers when pictures were shown for 2,000ms. Furthermore, when the images were presented for 500ms, smokers who had tried to stop smoking on several occasions, showed greater awareness for smoking-related scenes compared to non-smokers. This is consistent with a bias in the maintenance of drug-related cues. The longer duration of 2,000 ms allowed smokers to make numerous transfers of their gaze between the two pictures. Thus, this is more likely to reflect processes involved in the maintenance of attention. This suggests that smokers have difficulty in disengaging their attention from smokingrelated stimuli (Mogg et al., 2003). Mogg and colleagues (2003) have aptly phrased this as an 'attentional disengagement mechanism'. However, the results from the visual probe task suggest that only those smokers who had made repeated attempts to guit smoking showed a pronounced bias in the initial orientation for smoking-related cues. Those who had not tried to guit smoking did not show this bias. The shorter 500 ms condition is assumed to reflect the direction of initial orientation of attention (Mogg et al., 2003).

These results indicate that attentional biases in drug-dependence are an oversimplification. Two biases, the automatic capture of attention (Robinson & Berridge, 1993 as cited in Bradley et al., 2003) and the maintenance of attention (Mogg et al., 2003), may operate in different ways on attentional processes. Moreover, LaBerge (1995 as cited in Field et al., 2004a) has suggested that these two biases in attention have separate underlying neural subsystems, and motivational factors are more likely to influence maintenance of attention rather than initial orienting. The results from Bradley et al., (2003) suggest that addiction-related biases may operate in the maintenance of attention.

The visual probe task, however, only provides a view of attentional responses at the time of offset of the pictures; consequently providing a limited view of attentional responses (Mogg et al., 2003). Recent studies have assessed non-smokers and smoker's eye movements to smoking-related pictures and control pictures whilst they completed a visual probe task. Analysing eye-movements is an advantageous method of measuring attentional biases as they provide measures which are directly observable and are an accurate representation of visual orienting. Furthermore, eye movements are usually automatic and rapid, so individuals are inclined to look at stimuli which 'grab' their attention (Jonides, 1981; Kowler, 1995 as cited in Mogg et al., 2003).

Mogg and colleagues (2003) found that smokers looked for longer, as indicated by duration of initial fixation, at smoking-related pictures than control ones when the images were presented simultaneously in contrast to non-smokers who spent a similar time looking a both pictures. Furthermore, smokers were more likely to initially look at smoking-related pictures than control ones, as indicated by direction of initial fixation, than non-smokers. However, the eye movement direction results were suggestive as smokers and non-smokers did not differ significantly on this measure. In a similar study, Kwak, Na, Kim, Kim, and Lee (2007) found that smokers maintained their gaze for longer on and initially fixated on aversive cues compared to non-smokers.

The current findings of Mogg et al., (2003) and Kwak et al., (2007) are consistent with a bias in attention for drug-related cues in smokers compared with non-smokers. They indicate that these biases may operate in an attentional disengagement mechanism. Moreover, smokers will preferentially direct their gaze to smoking-related cues than control ones indicating that biases may also work in an initial shift mechanism. However, further research is needed to clarify this issue, as smokers and non-smokers did not differ significantly on this initial eye movement direction.

Recent research has also indicated that attentional biases for drug-related cues may be functionally related to frequency and quantity of substance use (Field & Cox, 2008). For example, in a visual probe task Townshend and Duka (2001) and Field, Mogg, Zetteler and Bradley (2004) found that biases in attention were stronger in heavier than light drinkers but only when pictures were shown for 500ms. Similarly, research has found a positive correlation between quantity and frequency of cannabis use and attentional biases for cannabis-related cues on visual probe and stroop tasks (Field, 2005; Field, Mogg & Bradley, 2004b). These results are consistent with the incentive motivational model postulated by Robinson and Berridge (2001). However, results from smokers are more inconsistent. Some research has indicated no relationship between frequency and quantity of smoking and nicotine dependence (Waters & Feyerabend, 2000; Munafò, Mogg, Roberts, Bradley, & Murphy, 2003). Other research has suggested a positive relationship (Zack, Belsito, Scher, Eissenberg, Corngall, 2001; Mogg & Bradley, Mogg, Field and Bradley (2005), in a visual probe task, found that low 2002). dependent smokers maintained their attention for longer and approached smokingrelated cues faster than high nicotine dependent smokers, as assessed by their eye movements. A negative relationship has also been reported in visual probe and stroop tasks (Hogarth, Mogg, Bradley, Duka & Dickinson, 2003; Waters, Shiffmann, Savette, Paty Gwaltney & Balabanis, 2003).

In order to account for these inconsistent results, Mogg et al.(2005) concluded that nicotine dependence is different from other dependence processes in that there is a switch to 'habit responding' from 'incentive responding', thereby diminishing the importance of the smoking-related cues. In high nicotine dependent smokers, smoking is therefore occurring out of habit as proposed by Tiffany (1990). This explanation is tentative and additional research is required to explain for these inconsistent results in smokers.

A possible reason for these conflicting findings is that the proportion of very high dependent smokers tends to be underrepresented in the smoker samples. In the research carried out by Mogg and colleagues (2005) the proportion of high dependent smokers was relatively small and their smoking histories short. On average, they recruited smokers to the heavily dependent group who had only been smoking for 6 years and smoked approximately 13 cigarettes per day. In comparison, their low nicotine dependency group had been smoking for approximately 3 years and smoked on average 8 cigarettes a day. The difference in smoking characteristics between the low and high nicotine dependence groups was small. Similarly, more than half the sample, in the study carried out by Mogg et al. (2003), smoked less than 20 cigarettes per day and the majority had smoked for less than 7 years. Thus, it would seem helpful, when investigating the relationship between attentional biases and nicotine dependence, to include a smoker sample where heavily dependent smokers are adequately represented. Accordingly, the current study recruited a wide range of smokers including those who were young with relatively short smoking histories, as little as one year, and those who were older with long smoking histories, as much as 60 years.

Furthermore, current studies have also presented dependence as an all or noting condition in which dependence is only presented as one given group. Dependence is assumed to arise when a smoker has been exposed to enough nicotine that they reach a threshold which is enough to trigger dependence processes. These processes are portrayed as different from those associated with 'normal' functioning (Tiffany, Conklin, Shiffman & Clayton, 2004). It has been suggested that as nicotine dependence develops, the process of dependence may change quantitatively (Robinson & Berridge, 2003 as cited in Smolka et al., 2006). Tarpet et al., (2003, as cited in Smolka et al., 2006) has shown that cue-induced brain activity increases continually with alcohol abuse.

To address these issues, the present study examined if attentional biases, as indexed by eye movements, could be extended to samples of heavier, long-term smokers. A major aim of the current study was to consider whether biases in visual orienting to smoking-related cues would be enhanced as a function of the nicotine dependence level. A further aim of the present study was to examine, not only attentional biases, but how quickly smokers and non-smokers approached the smoking-related cues (Mogg et al., 2005). Specifically, it was considered whether smokers with higher levels of nicotine dependence would be quicker to approach smoking-related cues, compared with less dependent smokers. This follows the incentive-salience mechanism postulated by Robinson and Berridge (2001) which suggests that attentional biases for drug-related stimuli are functionally related to incentive processes. Drug dependence is maintained and exacerbated through incentive sensitisation mechanisms. Therefore, in more dependent individuals where the incentive salience for smoking cues is stronger, they will show a greater attentional and approach biases for these cues (Mogg et al., 2005). Thus, biases in overt orienting in non-smokers, low dependent smokers, and highly dependent smokers were examined by measuring eye movements while participants completed a visual probe task with smoking-related and control pictures. Nicotine dependence was assessed by the Fagerström Test for Nicotine Dependence (FTND; Heatherton, Kozlowski, Frecker & Fragerström, 1991). The procedure was similar to the one used by Mogg et al., (2003). As noted earlier, measuring eye movements provides a direct and accurate representation of visual orientating (Mogg et al., 2003). Current studies have shown that eye movements are sensitive to changes in attention in smokers and non-smokers (Mogg et al., 2003; Kwak et al., 2007). Gaze duration may be an invaluable eye movement index as it shows the strength of the reinforcing effect of drug-related stimuli, which plays a fundamental role in maintaining drug-dependence. Furthermore, it may also highlight individual variation in vulnerability to drug addiction (Robinson & Berridge, 2003 as cited in Mogg et al., 2003). Importantly, measuring eye movements do not rely on the subjective experience of participants thus are more likely to reflect the automatic incentive salience process (Mogg et al., 2003).

In the present study the primary measures were the length of fixation within an area of interest, and the time to first fixation within an area of interest when smoking-related and control pictures were presented simultaneously. An area of interest was a useful tool for quantifying gaze data on a higher level in order to use area of interest (AOI) analysis. When using AOI analysis, an AOI within the stimuli can be defined. The current study defined an AOI as the area directly covering the smoking-related stimuli, such as cigarette or cigarette packet. This was then compared with the rest of the image including the control picture.

Based on Robinson and Berridge's (2001) incentive-salience mechanism, it was hypothesised that smokers, both low and high dependency would be more likely to hold their gaze for longer, as indicated by fixation and observation length, and attend more to, as indicated by fixation and observation count, to smoking-related AOIs than control pictures. Furthermore, it was expected that high and low dependent smokers would differ, in that highly dependent smokers would maintain their gaze for longer on an AOI. This would reflect a bias in maintaining attention on drug-related cues. It was also hypothesised that highly dependent smokers would be quicker to fixate, as indicated by time to first fixation, within an AOI compared with low dependent smokers and nonsmokers, reflecting a bias in the initial shift of attention.

## Method

## Design

This was a between-groups design with one independent variable; level of dependency which had three levels, non-smokers, low dependency and high dependency smokers. The dependent variables were determined using Tobii eye tracking equipment and included length of fixation within an AOI and time to first fixation within an AOI. It was hypothesised that smokers, both low and high dependency would be more likely to hold their gaze for longer, as indicated by fixation and observation length, and attend more, as indicated by the fixation and observation count, to smoking-related AOIs than control pictures. Furthermore, it was expected that high and low dependent smokers would differ, with highly dependent smokers maintaining their gaze for longer and attending more to an AOI. It was also hypothesised that highly dependent smokers would be quicker to fixate, as indicated by time to first fixation, within an AOI compared with low dependent smokers and non-smokers.

## Participants

Fifty-three participants in total were recruited for the study from among students and staff at Leeds Metropolitan University as well as family and friends of the researcher who volunteered. The control group consisted of 23 non-smokers (18 women and 5 men), with a mean age of 20.9 years (SD=2.49), who reported never having smoked regularly. The low dependency group (9 women and 6 men) had a mean age of 22.6 years (SD=3.48) and scored less than 5 on the FTND. On average, they had been smoking for 3.6 years (range: 1-8 years). The high dependency group (5 men and 10 women) had a mean age of 41.4 years (SD=19.59) and scored five or more on the FTND. On average, they had been smoking for 18.67 years (range 1-60 years). Participants all reported being healthy and those who were ex-smokers or who were currently attempting to quit smoking were exempt from the study.

#### Materials

#### Dependency

Subjects completed the FTND, a six-item self report questionnaire (Heatherton et al., 1991), a copy of which can be found in appendix 1. The questions are designed to assess nicotine dependence through time to first cigarette of the day, as upon waking dependent smokers have decreased levels of plasma due to the short half-life of nicotine. Thus, unless these smokers have their first cigarette quickly they are likely to experience discomfort (Heatherton et al., 1991). The number of cigarettes smoked per day is derived as a face valid measure of nicotine dependence, as previous studies have reported dependence as a direct function of smoking rate (Brantmark, Ohlin & Westling, 1973 as cited in Heatherton et al., 1991). The remaining FTND questions are face valid measures of smokers difficulty refraining from smoking, for example in forbidden places or when ill, as well as increased smoking in the morning (Heatherton et al.

al., 1991). Participants responded by choosing one of a choice of answers which best reflected their smoking behaviour. Scores for each answer ranged from 0 to 3 with a total score range of 0 (non-dependent) to 10 (highly dependent). A score of five or more indicated a significant dependence, while a score of four or less showed a low to moderate dependence. Question seven was added by the investigator which required participants to give their age. The validity of the FTND has been confirmed by Payne, Smith, McCracken, McSherry and Antony (1994) as well as by Prokhorov et al. (2000) in a population of adolescent smokers. This was important since the FTND was used among a young smoking population. The FTND was also chosen because it is closely associated with biomedical measures of heaviness of smoking (Heatherton et al., 1991).

## **Pictorial Stimuli**

The pictorial stimuli consisted of 20 coloured photographs of smoking-related scenes, such as a woman holding cigarette to mouth or a cigarette packet sticking out of a dustbin. Each was paired with a photograph of another scene matched as closely as possible for content, but lacking any smoking-related cues, for example a woman applying lipstick or crisp packet sticking out of a dustbin. An additional 10 picture pairs, unrelated to smoking, were used as fillers. The majority of the selected pictures were taken by the researcher, and the rest were taken from Microsoft Word Clipart. A full copy of the pictures used can be found in appendix 2. The pictures were adjusted in size so they were approximately 800 pixels wide by 600 pixels high, and were all converted into jpegs. Pictures were presented side by side so there was no gap.

#### Visual Movements

The task was presented using Tobii StudioTM version 1.5.12 software, on a Pentium PC with 16 <sup>1</sup>/<sub>2</sub> inch VGA colour monitor and standard keyboard. Participants eve movements were recorded while they completed the visual probe task using a computerised eve tracking system (Tobii Eve Tracker, Model X120). The visual probe task was created in Tobii Studio TM. The stimuli were selected by adding media to the timeline. Each trial consisted of a central fixation, followed by a picture pair then a dot probe. The central fixation point preceding the image and the dot probe following the image were created using the instruction media type within Tobii Studio TM. Both the central fixation point and dot probe were disabled for visualisations as no eye tracking data were needed for these elements. A view time of 5 seconds was selected for the central fixation, which was then replaced by the image pair. The dot probe was shown until the participant either pressed the "1" key to indicate two vertical dots or the "2" key on the keyboard to indicate two horizontal dots. The picture pairs were imported from a catalogue of jpeg images into the Tobii Studio. Filler images were marked as Dummy Media as no visualisation data were needed for these images. A view time of 5 seconds was created for all images, which were then replaced by the dot probe. This procedure was repeated for all 30 picture pairs.

#### Procedure

Testing took place in the Vision and Movement Laboratory within Leeds Metropolitan University. First, all participants were given an information sheet, which can be found in appendix 3, and provided informed consent, a copy of which can be found in appendix 4. Smokers then completed the FTND (Heatherton et al., 1991) which determined which smoking group they should be assigned to. Each of the three groups including non-smokers, low dependent smokers and highly dependent smokers attended the laboratory on one occasion. The smoking groups were required to abstain from smoking participants had similar nicotine deprivation levels. Research by Field et al. (2004a) has indicated that when deprived, smokers gaze for longer at smoking-related stimuli than control stimuli, compared to when non-deprived.

Participants were seated 90 cm from the screen and positioned so that their gaze was central to the track status metre. The purpose of the track status metre is to verify that the participant is in the correct position in front of the eye tracker, and that the system is able to properly track the participant. The track status metre shows a black box with two white circles and a status bar at the bottom. The black box represents the field of view of the eye tracking camera and the white dots represent the eyes of the participant. The coloured status bar indicates the tracking ability. At the start of each recording, it was ensured that this bar was coloured green and not flickering, indicating that the system was tracking the participant well. Recordings were not started if the status bar was red or flickering between red, yellow and green as tracking would not be successful. The eye-tracking equipment was calibrated for each participant by having the participant track as small red round dot to five different locations on the screen. Calibration plots which showed error vectors and long lines were recalibrated. Also, the calibration dots which were not picked up by the eye tracker were recalibrated.

The attentional task consisted of 10 filler and 20 experimental trials, which were presented in the same order for each participant. Each trial started with a fixation cross, presented for 5 seconds in the centre of the screen. After an interstimulus interval of approximately 0.1 seconds, a picture pair was displayed for 5 seconds. Each picture was positioned either side of the central position. Two dots were presented immediately after the pictures until the participants made the correct response. Participants were instructed to press the "1" key for two vertical dots (:) and the "2" key for two horizontal dot (..) as quickly as possible.

On the experimental trials, each picture pair (20 smoking-control picture pairs and 10 filler picture pairs) were presented only once to reduce to the monotony on the task. The smoking related pictures appeared 10 times on the left and 10 times on the right. The probe of vertical and horizontal dots replaced the picture pairs in equal numbers. At the end of the session, participants were thanked for their time and given a debrief sheet which outlined the nature of the study, a copy can be found in appendix 5.

#### Preparation of eye-movement data (EM) and statistical analysis

Data was analysed using Tobii StudioTM version 1.5.12 software. An AOI was defined in each picture pair, apart from filler pairs, and was the area directly covering the smoking stimuli, for example, a cigarette packet.

The dependent measures of time to first fixation, fixation length, fixation count, observation length and observation count were analysed as follows:

*Time to first fixation* This was the time in seconds from when the stimulus was shown until the start of the fixation.

*Fixation length* This was calculated as the length of fixations in seconds within an AOI. *Fixation count* This included the number of fixations within an AOI.

*Observation length* This was defined as the total time in seconds for every time a participant had looked within an AOI, starting with a fixation within the AOI and ending with a fixation outside the AOI. There had to be a fixation within the AOI for it to be an observation

Observation count This included the total number of visits and re-visits to an AOI.

A mean for each of the dependent measures for each participant was calculated based only on the AOIs and not the fixation points or dot probes. Data from filler trials was not analysed. PASW statistics 17 for windows was used to conduct a one-way between analysis of variance which explored the impact of nicotine dependency on each of the above EM indices.

## Results

The analysis was conducted as follows: a one-way between groups analysis of variance was conducted to explore the impact of nicotine dependency on each of the EM indices. This was performed to test the hypotheses and enable the comparison of results across studies. Subjects were divided into three groups according to their dependency (Group 1: non-smokers; Group2: low dependency; Group 3: highly dependent smokers).

## **Group Characteristics**

To test the hypotheses, participants were allocated to the high dependency group depending on whether their Fagerström score was five or greater. If their score was four or less they were assigned to the low dependency group. An independent-samples t-test was conducted to compare the FTND scores for the low and high dependency groups. There was a statistically significant increase in FTND scores for the high dependency group compared with the low dependency group (t(28) = -11.45, p>.0005). Table 1 shows a summary of group characteristics.

	Low n deper	icotine Idence	High nicotine dependence		
	М	SD	M	SD	
Age (years)	22.6	3.48	41.4	19.59	
Nicotine dependence (mFTQ)	1.47	1.25	7.13	1.46	
Years of smoking	3.60	2.90	18.67	18.84	

# Table 1Mean characteristics of low and high nicotine dependence groups

#### **Eye Movement Data**

The mean fixation count, fixation length, time to first fixation, observation length and observation count within an AOI as well as standard deviations for non-smokers, low and highly dependent smokers are presented in table 1.

# Table 2 Mean response of each dependency group (Standard Deviation in Parentheses)

Dependency	Fixa cou	ntion unt	on Fixation t length (secs)		Time to first fixation (secs)		Observation length (secs)		Observation count	
	М	SD	М	SD	М	SD	М	SD	М	SD
Non (N=23)	1.27	0.36	0.28	0.10	1.23	0.43	0.34	0.11	1.11	0.34
Low (N=15)	1.79	0.70	0.34	0.11	0.96	0.42	0.45	0.45	1.39	0.51
High (N=15)	2.22	1.70	0.35	0.17	1.01	0.41	0.54	0.37	1.53	0.94

Table 2 indicates that non-smokers took longer to fixate within an AOI than either the low or highly dependent groups. Furthermore, the low dependency group appear to be faster at fixating within an AOI than the highly dependent group. Table 2 also suggests that both low and highly dependent smokers fixated within an AOI longer than non-smokers. Moreover, highly dependent smokers appear to fixate for longer than low dependent smokers, although this difference appears to be small. It also appears to be the case that highly dependent smokers fixated more within an AOI than either non-smokers or low dependent smokers. Table 2 indicates that both the low and high dependency groups observed for longer within an AOI than non-smokers. Moreover, the highly dependent group appeared to observe for longer than the low dependency group. Table 2 also indicates that the observation count was greater for highly dependent smokers than either low dependent or non-smokers. Additionally, low dependent smokers were higher than non-smokers.

*Gaze Length* A one-way between groups analysis of variance revealed no significant difference between level of dependency and fixation length within an AOI (F(2,50) = 1.53, p=0.23). However, the one-way between groups analysis of variance did

reveal a significant difference in the observation length within an AOI for the three dependency groups (F(2,50) = 3.72, p<.05). The effect size, calculated using eta squared was .13 which indicated a large difference in mean observation length between the groups. Post-hoc comparisons using Tukey HSD test indicated than the mean observation length for non-smokers was significantly different from highly dependent smokers (p<.05). The low dependency group did not differ significantly from either the non-smokers (p=0.29) or high dependency group (p=0.55). Refer to table 2 for the mean observation length for each group.

*Time to initial fixation* A one-way between groups analysis of variance revealed no significant difference between time to first fixation within an AOI and level of dependency (F(2,50) = 2.31, p=0.11).

Attention rate A one-way between groups analysis of variance also showed that there was a statistically significant difference in the number of fixations within an AOI between the three dependency groups (F(2,50) = 4.15, p<.05). The effect size, calculated using eta squared, was .14 which indicated a large difference in the mean number of fixations between the groups. Post hoc comparisons using Tukey HSD test indicated that the mean number of fixations for Group 1 was significantly different from Group 3 (p<.05). Group 2 did not differ significantly from either Group 1 (p=0.27) and Group 3 (p=0.48). Refer to table 2 for the mean fixation count for each group. However, there was no significant difference in the mean observation count within an AOI between the three dependency groups (F(2,50) = 2.29, p=0.11).

#### Discussion

A major aim of the current study was to investigate whether biases in visual orienting to smoking-related cues would be enhanced as a consequence of nicotine dependence level. Furthermore, this study aimed to examine not only attentional biases to smoking-related cues but also how quickly they orientated their attention to these cues. Specifically, it was considered whether smokers with high nicotine dependence levels would approach smoking-related cues faster compared with smokers with lower nicotine dependence levels.

A comparison of non-smokers, low and high nicotine dependence groups showed that smokers with high levels of nicotine dependence levels attended significantly more than non-smokers to the smoking-related cues, as indicated by fixation count. Furthermore, the high dependency group showed an enhanced attentional bias for smoking-related cues than non-smokers, as reflected by their longer observation length. Despite showing an attenuated response than non-smokers and a dampened response in contrast to the high dependency group, the low dependence nicotine group did not differ significantly from either the high dependency group or the non-smokers on the latter two variables. The bias to approach smoking cues, as indicated by time to first fixation, did not differ significantly between the three groups.

As hypothesised, in comparison with non-smokers, those with higher levels of nicotine dependence spent significantly longer looking at and attended more to smoking-related cues than control ones, which is consistent with an enhanced attentional bias for smoking cues in the high dependence group. These results are compatible with those of Mogg et al., (2003) and Kwak et al., (2007) who found that smokers hold their gaze for longer on smoking-related pictures than control pictures. The present results also indicated that biases for smoking-related cues are enhanced as a result of nicotine dependence. The high dependence group was more likely to look for longer, and attend more to smoking-related cues than the low dependency group who in turn attended more and looked for longer than non-smokers. This is consistent with Mogg and Bradley (2002) and Zack et al. (2001) who found a positive correlation between the strength of attentional bias and severity of nicotine dependence.

Consequently, the present results seem compatible with the incentive-sensitisation theory (Robinson & Berridge, 2001). Robinson and Berridge (2001) suggested that an incentive-salience mechanism mediates attentional biases for drug-related cues. Consequently, drug-related cues are perceived as highly salient and difficult to ignore. These drug-related stimuli become 'wanted' thus capturing attention and eliciting approach behaviours. Thus, attentional biases for drug-realted stimuli are an index of incentive processes. Accordingly, it would follow that in more nicotine dependent individuals the attentional biases are attenuated as the incentive salience is greater for them (Mogg et al., 2005). The present results support the latter: high levels of nicotine dependence were associated with an enhanced attentional bias for smoking cues (in observation length) and with increased salience of smoking cues (fixation count). Balfour (2004, as cited in West, 2006) proposed that drug-related cues are granted incentive salience due to increased concentrations of dopamine in the nucleus accumbens core which leads to the development of stimulus response behaviour when those cues are presented. The present results indicate that

dopamine levels in the core of the nucleus accumbens may operate on a continuum; that as nicotine dependence develops, the effects of dopamine levels is the accumbens core is increasingly sensitised. However, the latter needs to be treated with caution as the low dependency nicotine group did not differ significantly on either the observation length or fixation count index from the high dependency group or non-smokers. Further research is needed to clarify if biases for drug-related cues are enhanced as a result of increasing nicotine dependence.

Tiffany (1990) postulated that drug-taking behaviour is mediated by habit. When drug-taking behaviour is inhibited, non-automatic effortful processing resources are assigned to obtaining the drug and the person will experience an increase in drug urges. Consequently, when the drug is unavailable the individual will have increased attentional bias for drug-related cues as the individual is experiencing an increase in drug-craving (Field et al. 2004a). The current findings showed that smokers tended to fixate for longer on smoking-related cues supporting Tiffany's (1990) model. However, since participants urge to smoke was not assessed in the present study, this is not conclusive. Further research showing an association between longer fixation length and increased urge to smoke would highlight that when the drug is not available, drug urges play an imperative role in mobilising drug-seeking behaviours and eliciting attentional biases for drug cues (Mogg et al., 2003). Everitt, Dickinson and Robbins (2001, as cited in Smolka, 2006) have suggested that both the incentive and habit-based mechanisms are not mutually exclusive. Instead they are perhaps complementary and both contribute to the maintenance of drug dependence.

The categorical model of dependence assumes that dependence arises when smokers reach a threshold of nicotine dependence which is high enough to activate dependence processes. These are portrayed as different from normal functioning (Smolka et al., 2006). However, this neglects that basic elements of the dependence processes are present in the early stages of cigarette use, and may change qualitatively during the development of nicotine dependence (Tiffany, Conklin, Shiffman & Clayton, 2004). Smolka et al., (2006) have already shown that severity of nicotine dependence is associated with cue-induced brain activity in separate neuronal networks including neural circuits linked to visuospatial attention and in the motor system. FTND scores were positively associated with BOLD responses in the dorsal anterior cingulate cortex, inferior parietal cortex, secondary visual areas and the parahippocampal and fusiform gyri. Moreover, the strongest association between nicotine dependence level and cue-induced brain activity was found in the premotor cortex and supplementary motor area as well as in the left primary motor cortex. The current findings are consistent with the idea that the influence of these mechanisms may increase continuously with nicotine use. Highly dependent smokers tended to show an enhanced attentional bias for drug-related cues on all EM indexes apart from time to first fixation. For example, highly dependent smokers fixated more times on smoking-related cues than low dependent smokers who in turn fixated more times than non-smokers.

Subsequently, the present results are inconsistent with some of the visual probe studies which found that daily smoking rate correlated negatively with attentional biases, indicating that lighter smokers have greater attentional biases (Bradley et al., 2003, Experiment 1; Hogarth et al., 2003). Furthermore, the results are incompatible

with that of Mogg et al. (2005) which found that, during a visual probe task, smokers with lower levels of nicotine dependence maintained their attention for longer, as indicated by gaze dwell time, and approached smoking-related cues faster than those with high levels of nicotine dependence. The authors concluded this supported an incentive-habit theory where the effects of the incentive motivational properties of the drug-related cue on behaviour diminishes as there is a switching in response from 'incentive responding' to 'habit responding'. However, as noted earlier, these studies tended to recruit smokers with relatively short smoking histories and low levels of nicotine dependence. In the present study, the aim was to sample a wide range of smokers and ensure that those with high nicotine dependence were well represented. Thus, the current research could be considered to be a truer reflection of the effects of level of nicotine dependence on attentional biases as this research contained smokers at both extreme ends of the spectrum of nicotine dependent smokers.

Interestingly, in the current study, the relationship between higher nicotine dependence and enhanced attentional bias was most apparent in the observation length measure than in the other attentional bias measure, fixation length. Observation length required there to be a fixation within the AOI to start and a fixation outside the AOI to end in contrast to fixation length which only required there to be a fixation within the AOI to start and ended as soon as the individual's gazed The data from the observation length index indicated that the high moved. dependency group took longer to fixate on something else outside of the smokingrelated AOI. This is consistent with research which has suggested that attentional biases for drug-related cues operate in an attentional disengagement mechanism (Mogg et al., 2003). Once the high dependency group fixated within the smokingrelated AOI they had difficulty fixating on something else outside the AOI. The low dependency group also tended to look for longer within a smoking-related AOI than non-smokers, but not as long as the high dependency group indicating that attentional disengagement mechanisms may be functionally related to the severity of nicotine dependence. Thus, with increasing nicotine dependence smokers have increasing difficulty in disengaging their attention from smoking-related stimuli.

The present study also indicated that smokers were quicker to direct their attention to smoking-related cues, while non-smokers took longer. This is consistent with Mogg et al., (2003) who suggested that biases may also operate in an attentional shift mechanism. However, this evidence is not conclusive as the groups did not differ significantly on this EM index. Moreover, there was no indication that this was functionally related to dependence as the low dependent group were faster to fixate on the smoking-related cues than highly dependent smokers.

There are several methodological limitations which need to be addressed and could account for the lack of significant differences of some of the EM indexes. All smoking participants were instructed to take a 12 hour abstinence from smoking prior to the experiment. However, there is some doubt over whether a few participants undertook this abstinence. Consequently, some participants may have differed markedly in the levels of nicotine withdrawal resulting in varying degrees of craving for smoking-related cues. Field et al., (2004a) have shown that when deprived, smokers maintained their gaze for longer on smoking-related cues than

control cues, relative to when non-deprived. Future research should consider getting participants to provide a sample of expired carbon monoxide (CO) sample on a smokerlyzer (Bedfont Scientific, Ltd) at the start of the experiment.

Additionally, it should also be considered that non-smokers may have found many of the drug-related cues highly salient dues to the emotional valence of the smokingrelated stimuli. This means the small difference between the non-smokers and smoking groups could have been to non-smokers perceiving the drug-related stimuli as highly unattractive. Future research should consider including a questionnaire which identifies how non-smokers feel about the images presented. Moreover, Rösler et al. (2005) have found that in comparison to older adults, young adults show a stronger bias towards emotionally negative material. The mean age of the nonsmokers and low dependent smokers in the current study was approximately 20 years less than that of the high dependency group. Subsequently, due to their younger age, the non-smokers and the low nicotine dependency group may have automatically oriented towards the smoking-related cues because of the negative emotional valence of the cues, resulting in minimal differences between the groups. This suggests that future research should include three types of stimuli; substance related, emotionally valenced but unrelated to substance use, and emotionally neutral (Field & Cox, 2008).

Smoking participants were assigned to the low and high dependency category based on their scores to the FTND. However, there are a couple of major implications related to the FTND. The FTND was originally designed to assess nicotine dependence in adult smokers. Consequently, this instrument is employed with the assumption that the characteristics associated with adult-end-stage dependence are wholly applicable to young adults in the early stages of smoking (Tiffany et al., 2004). There are various problems with this assumption. Firstly, the experiences of nicotine dependence in the young, low dependent smokers in the present study may be qualitatively different from the older, high dependent smokers (Colby et al. 2000a as cited in Tiffany et al., 2004). Young smokers may therefore give higher or lower salience to certain aspects of their smoking behaviour. Secondly, young smokers may still experience the same arrangement of dependence effects but may use different qualifiers to express those effects. Lastly, the set of behaviours that constitute markers of behaviour may change over the course of smoking. Thus, what is relevant for assessing dependence in highly experienced smokers is not necessarily appropriate for assessing dependence in less experienced smokers (Tiffany et al., 2004). This has implications for the current study resulting in younger smokers being placed in the wrong dependency category as consequence of them characterising their smoking behaviour differently in contrast to older, high dependent smokers. Instruments which are specially designed to the capture the early stages of dependence will substantially enhance our understanding of dependence (Tiffany et al., 2004).

In addition, many participants who would have described themselves as heavily dependent, smoking more than 20 cigarettes a day were classed as low dependency as a result of the answers they gave to some questions on FTND. For example, question one asks participants how soon after waking would they have their first cigarette. Since many of the younger participants lived in rented accommodation or with parents and had to go outside to smoke this tended to mean they would answer

after 60 minutes. Consequently many smokers were placed in the low dependency group. This could have resulted in the finding that there was no significance difference between the low and high dependency groups on any EM index. As well as including the FTND, future research should also include respective questions from the Structured Clinical Interview for DSM-IV, the criteria for assessing nicotine dependence (First et al., as cited in Smolka et 2006).

Despite these methodological limitations, understanding biases in cognitive processing of drug-related cues which underlie drug-taking behaviour are not only important theoretically, but are also of clinical importance when identifying factors which cause drug relapse (Mogg et al., 2005). De Jong et al. (2006, as cited in Field & Cox, 2008) has suggested that by reducing an individual's attentional bias for drug-related cues may be one way of eliminating craving. This may be possible through cognitive and psychological interventions such as the Alcohol Attention-Control Training Program (AACTP) suggested by Fadardi and Cox (2009). Fadardi and Cox (2009) showed that following AACTP, harmful drinkers had a decrease in their attentional biases for alcohol related-cues, and furthermore showed reductions in alcohol consumption which were maintained three months later. Future research should consider developing a similar program for smokers as by reducing a smoker's attentional bias for smoking-related cues may be one way of alleviating nicotine craving, thus preventing relapse after attempts to quit. This may be more effective than current forms of nicotine replacement therapy such as gum and skin patches where only between 1 in 20 and 1 in 10 smokers stop smoking who would not otherwise would have done (West, 2006).

In addition, it may also be fruitful to consider the effects of manipulating deprivation level in smokers. As noted by Mogg et al., (2005) under extreme deprivation conditions, strong incentive effects may be elicited making the relationship between nicotine dependence and attentional biases less clear. Attentional biases for smoking-related cues may consequently rise to high levels in all smokers. Given that cognitive and behavioural indices of incentive salience mechanisms, such as attentional biases, are thought to play a crucial role in maintaining drug-taking behaviour and increasing relapse rates this may help to indentify the exact nature between nicotine dependence and these indices (Mogg et ., 2005).

The EM indexes have provided an invaluable objective of strength of activation of incentive motivation processes, and suggested that this varies on a continuum. Its main advantage is that it does not rely on introspection or subjective self-report thus is invaluable in reflecting non-conscious incentive salience processes (Mogg et al., 2003). Further research, should therefore investigate the extent to which these EM indexes are predictive of smoking behaviour. For example, relapse rates after attempts of quitting (Mogg et al., 2003). Furthermore, relapse rates after attempts of quitting should be examined to see if they are functionally related to nicotine dependence.

In summary, this study replicates earlier findings that showed evidence of attentional biases in smokers towards smoking related cues. Smokers with higher levels of nicotine dependence spent significantly longer looking at, as indicated by observation length, and attended more, as indicated by fixation count, to smoking-related cues than non-smokers. It was concluded that this was consistent with the

incentive-sensitisation theory of drug dependence (Robinson & Berridge, 2001). The results also indicated that biases for drug-related cues are enhanced as result of increasing nicotine dependence. This indicates that attentional biases for drug-related cues are functionally related to incentive processes, with severity of nicotine dependence being a positive predictor of such biases. Modelling nicotine dependence as a parametric phenotype shows how the processing of smoking-related cues is functionally related to severity of dependence and allows the detection of differences between occasional, non-dependent smokers and heavy dependent smokers (Smolka et al., 2006). Future research should consider how predictive the EM indexes are of smoking behaviour (Mogg et al., 2003).

## References

Bradley, B.P., Mogg, K., Field, M., & Wright, T. (2003). Attentional bias in drug dependence: vigilance for cigarette-related cues in smokers. *Psychology of Addictive Behaviours*, 17, 66-72.

Catley, D., Shiffman, S., & O'Connell, K.A. (2000). Absent minded lapses during smoking cessation. *Psychology of addictive behaviours*, 14, 73-76.

Cox, L.S., Tiffany, S.T., & Christen, A.G. (2001). Evaluation of the brief questionnaire of smoking urges (QSU-brief) in laboratory and clinical settings. *Nicotine and Tobacco Research*, 3, 7-16.

Fadardi, J.S., & Cox, W. M. (2009). Reversing the sequence: reducing alcohol consumption by overcoming alcohol attentional bias. *Drug and Alcohol Dependence*, 101, 137-145.

Field, M., (2005). Cannabis 'dependence' and attentional bias for cannabis-related words. *Behav. Pharmacol.* 16, 473–476.

Field, M., & Cox, W.M., (2008). Attentional bias in addictive behaviors: A review of its development, causes, and consequences. *Drug and Alcohol Depend*, 97, 1-20.

Field, M., Mogg, K., & Bradley, B.P. (2004a). Eye movements to smoking-related cues: effects of nicotine deprivation. *Psychopharmacology*, 173, 116-123.

Field, M., Mogg, K., & Bradley, B.P., (2004b). Cognitive bias and drug craving in recreational cannabis users. *Drug Alcohol Depend*. 74, 105–111.

Field, M., Mogg, K., Zetteler, J., & Bradley, B.P., (2004). Attentional biases for alcohol cues in heavy and light social drinkers: the roles of initial orienting and maintained attention. *Psychopharmacology (Berl.)* 176, 88–93.

Franken, I.H.A., Kroon, L.Y., Wiers, R.W., & Jansen, A. (2000). Selective cognitive processing of drug cues in heroin dependence. *Psychopharmacology*, 14, 395-400.

Heatherton, T.F., Kozlowski, L.T., Frecker, R.C., & Fagerström, K. (1991). The Fagerström for nicotine dependence: a revision of the Fagerström tolerance questionnaire. *British Journal of Addiction*, 86, 1119-1127.

Hogarth, L.C., Mogg, K., Bradley, B.P., Duka, T., & Dickinson, A., (2003). Attentional orienting towards smoking-related stimuli. *Behav. Pharmacol.* 14, 153–160.

Kwak, S.M., Na, D.L., Kim, G., Kim, G.S., & Lee, J.H. (2007). Use of eye movement to measure smokers' attentional bias to smoking-related cues. *CyberPsychology & Behaviour*, 10, 299-304.

Lazev, A.B., Herzog, T.A., & Brandon, T.H., (1999). Classical conditioning of environmental cues to cigarette smoking. *Exp. Clin. Psychopharmacol*, 7, 56-63.

Mogg, K., & Bradley, B.P., (2002). Selective processing of smoking-related cues in smokers: manipulation of deprivation level and comparison of three measures of processing bias. *J. Psychopharmacol.* 16, 385–392.

Mogg, K., Bradley, B.P., Field, M., & Houwer, J. (2003). Eye movements to smokingrelated pictures between attentional biases and implicit and explicit measures of stimulus valence. *Addiction*, 98, 825-836.

Mogg, K., Field, M., & Bradley, B.P., (2005). Attentional and approach biases for smoking cues in smokers: an investigation of competing theoretical views of addiction. *Psychopharmacology (Berl.)* 180, 333–341.

Munafo, M., Mogg, K., Roberts, S., Bradley, B.P., & Murphy, M. (2003). Selective processing of smoking-related cues in current smokers, ex-smokers and never-smokers on the modified Stroop task. J. *Psychopharmacol.* 17, 310–316.

Payne, T.J., Smith, P.O., McCracken, L.M., McSherry, W.C., & Antony, M.M. (1994). Assessing nicotine dependence: A comparison of the Fagerstrom Tolerance Questionnaire (FTQ) with the Fagertsrom Test for Nicotine Dependence (FTND) in a clinical sample. *Addictive Behaviors*, 19, 301–317.

Posner, M. (1980). Orienting of attention. *The quarterly journal of experimental psychology*, 32, 3-24.

Prokhorov, A.V., De Moor, C., Pallonen, U.E., Hudmon, K.S., Koehly, L., & Hu, S. (2000). Validation of the Fagerstrom Questionnaire with salivary cotinine among adolescence. *Addictive Behaviors*, 25, 429–433.

Robinson, T.E., & Berridge, K.C. (2001). Mechanisms of action of addictive stimuli: incentive-sensitization and addiction. *Addiction*, 96, 103-114.

Rösler, A., et al. (2005). Effects of arousing emotional scenes on the distribution of visuospatial attention: changes with aging and early subcortical vascular dementia. *Journal of the Neurological Sciences*, 229–230, 109–116.

Sayette, M.A., & Hufford, M.R. (1994). Effects of cue exposure and deprivation on cognitive resources in smokers. *Abnormal Psychology*, 103, 812-818.

Smolka, M,N., Bühler, M., Klein, S., Zimmermann, U., Mann, K., Heinz, A., et al. (2006). Severity of nicotine dependence modulates cue-induced brain activity in regions involved in motor preparation and imagery. *Psychopharmacology*, 184, 577-588.

Tiffany, S.T., (1990). A cognitive model of drug urges and drug-use behavior: role of automatic and nonautomatic processes. *Psychol. Rev.* 97, 147–168.

Tiffany. S. T., Conklin, C.A., Shiffman, S., & Clayton, R. R. (2004). What can dependence theories tell us about assessing the emergence of tobacco dependence? *Addiction*, 99, 78-86.

Townshend, J.M., & Duka, T., (2001). Attentional bias associated with alcohol cues: differences between heavy and occasional social drinkers. *Psychopharmacology* (*Berl.*) 157, 67–74.

Waters, A.J., & Feyerabend, C. (2000). Determinants and effects of attentional bias in smokers. *Psychology of addictive behaviours*, 14, 111-120.

Waters, A.J., Shiffman, S., Sayette, M.A., Paty, J.A., Gwaltney, C.J., & Balabanis, M.H., (2003). Attentional bias predicts outcome in smoking cessation. *Health Psychol.* 22, 378–387.

West, R. (2006). Theory of addiction. Oxford: Blackwell Publishing Ltd.

Zack, M., Belsito, L., Scher, R., Eissenberg, T., & Corrigall,W.A., (2001). Effects of abstinence and smoking on information processing in adolescent smokers. *Psychopharmacology (Berl.)* 153, 249–257.