Introduction

Cigarette smoking represents a major health problem in the United States, and smoking cessation continues to be a difficult challenge. According to a recent survey, roughly 23% of Americans reported smoking cigarettes [1], and while most smokers express a desire to quit [2], only a small minority will be successful without treatment. Even after 6 months or more of effective treatment, only approximately 14 to 49% of smokers will achieve abstinence [3-8]. Due to the considerable health risks [9,10] and high societal costs [11,12] attributed to smoking, more efficacious smoking cessation treatments are urgently needed. In combination with other areas of research, functional brain imaging offers great potential for uncovering both molecular targets and brain circuits that mediate the acute effects of smoking and the chronic effects of nicotine dependence. Knowledge of the effects of smoking on brain function may lead to the creation of improved pharmacological and behavioral smoking cessation interventions.

Four main imaging modalities have been utilized in functional brain imaging studies of tobacco use and dependence: (1) functional magnetic resonance imaging (fMRI), (2) positron emission tomography (PET), (3) single photon emission computed tomography (SPECT), and (4) autoradiography. Researchers have used each of these modalities to determine the effects of acute and chronic cigarette smoking on brain function and smoking-related behaviors. For this review, the MEDLINE database was searched using keywords for each of the four modalities listed above cross-referenced with the words “tobacco,” “nicotine,” and “cigarette.” Only data driven functional imaging studies were included in this review, and relevant studies from reference lists within papers found on MEDLINE were also included. In order to examine brain activity responses to acute and chronic smoking in a cohesive fashion, this review will discuss studies of both nicotine and cigarettes, while acknowledging that cigarette smoke contains thousands of constituents other than nicotine [13,14]. Additionally, because blood flow and metabolism are closely linked under normal conditions [15], functional imaging techniques that measure each of these are combined under the general heading of brain activity.

The goal of this review is to provide a comprehensive summary of current findings from functional brain imaging studies of tobacco use and dependence. Topics covered in this review include: nicotine uptake into the brain; brain activity responses following acute nicotine/cigarette administration; effects of pharmacologic smoking cessation treatment on brain activity; functional imaging of nicotinic acetylcholine receptors; brain dopamine responses to nicotine and smoking; functional brain imaging of smoking-related cue-reactivity; functional brain imaging of cigarette withdrawal; relevance of other receptors, neurotransmitter systems, and genetics to nicotine dependence; and future directions.

Nicotine Uptake into the Brain

Numerous studies [16-18] have shown that the effects of many drugs, including nicotine, vary in relation to the rate of rise of the drug's concentration in the brain. Specifically, for drugs with addictive potential, it is thought that faster rates of rise are associated with more pronounced hedonic effects, thereby increasing the likelihood of eventual drug dependence [19,20]. Some recent studies [21,22] have utilized positron emission tomography (PET) and [(11)C] nicotine to help elucidate the cerebral kinetics of smoked nicotine.

In one recent study [21], PET scans of the brain were obtained in 11 healthy smokers following single puffs of [(11)C] nicotine-infused cigarettes. Investigators found that a single puff was associated with a "rise time", or the time in seconds for the [(11)C] nicotine concentration curve to rise from 20% to 80% of its maximal value, of 11-69 seconds, and was less than 15 seconds for most patients. Noting that this rise time is similar to that reported for smoked cocaine [23], the authors conclude that the cerebral kinetics of smoked nicotine contribute to dependence in smokers.
A similarly designed study performed by Rose and colleagues provides additional information [22]. In this study, 13 dependent smokers and 10 non-dependent smokers were imaged using PET while smoking [(11)C]nicotine-loaded cigarettes. The authors found that, because of slower nicotine washout from the lungs, dependent smokers had a slower rate of nicotine accumulation in the brain than observed in non-dependent smokers. This study, therefore, adds to the preceding study by indicating that rate of nicotine accumulation in the brain alone is not enough to explain dependence on smoked nicotine. It is clear that the link between nicotine’s kinetics in the brain and smoking behavior will be an important topic for future studies to clarify further.

**Brain Blood Flow and Metabolic Responses Following Acute Nicotine/Cigarette Administration**

A fair number of functional brain imaging studies have examined acute effects of nicotine administration or cigarette smoking compared with a placebo or control state (Table 1). Nicotine and cigarette smoking have been reported to alter the activity of a wide array of brain regions; however, several global and regional discoveries have been replicated, allowing for general conclusions about the acute effects of nicotine or smoking on brain activity [24].

One common finding is that cigarette smoking [25] and nicotine administration [26,27] result in decreased global brain activity. Furthermore, smokers who smoke cigarettes ad lib prior to SPECT scanning (including the morning of scanning) have reduced global brain activity compared to both former smokers and non-smokers [28]. These findings are generally supported by studies using transcranial Doppler ultrasound or the Xe 133 inhalation method to measure responses to smoking, with some [29-32], but not all [33,34], studies showing diminished cerebral blood flow.

In studies examining regional activity responses to nicotine or smoking, the most common findings are relative increases in activity in the prefrontal cortex (including the dorsolateral prefrontal cortex, and inferior frontal, medial frontal, and orbitofrontal gyri) [26,35,36], thalamus [26,36-40], and visual system [26,37-39]. Additionally, a Xe 133 inhalation study reported increases in frontal lobe and thalamic blood flow in smokers who smoked a cigarette [41]. Of these studies, those involving humans examined cigarette smokers, while those involving animals used non-dependent rats, with strong concordance of findings between each set of studies. While this group of studies demonstrate specific regional activation with cigarette smoking or nicotine administration, they also imply that activation of cortico-basal ganglia-thalamic brain circuits [42] mediates the subjective effects of smoking. Zubiena et al. [43] conducted a 15O-PET study using nicotine and denicotinized cigarettes in 19 smokers who were abstinent for 12 hours before PET. In this study, investigators discovered increases in the regional cerebral blood flow (rCBF) in the visual cortex and cerebellum, and reductions in rCBF in the anterior cingulate, right hippocampus, and ventral striatum. Cigarette craving in chronic smokers also correlated with rCBF in the right hippocampus, a region involved in associating environmental cues with drugs, and in left dorsal anterior cingulate, an area implicated in drug craving and relapse to drug-seeking behavior.

Since regional activity was normalized to whole brain activity in at least some of these studies, and whole brain activity has been found to decrease with nicotine or cigarette administration, the regional findings presented above may represent either increased regional activity or, possibly, less of a decrease in regional activity than in other brain areas. Regional decreases in activity are generally not seen with nicotine or cigarette administration, though at least two studies found relatively decreased activity in the amygdala (left [35] and right [40]). In summary, both reduced global activity as well as increased regional activity will be important topics for future studies to clarify further.

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**Table 1: Functional brain imaging studies of nicotine or cigarette administration.**

<table>
<thead>
<tr>
<th>Authors</th>
<th>Subjects</th>
<th>Method</th>
<th>Intervention</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>a) Animal Studies</td>
<td></td>
<td></td>
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<td></td>
</tr>
<tr>
<td>Mareno et al. [156]</td>
<td>Rats-chronically nic exposed vs. nic naive</td>
<td>2-deoxy-D-[1-14C] glucose autoradiography</td>
<td>SC nic (0.4 mg/kg) vs. saline</td>
<td>↑ thal, superior colliculus in chronically exposed; ↑ thal, superior colliculus, medial habenula and dorsal lateral geniculate in nic naive</td>
</tr>
<tr>
<td>London et al. [38, 39]</td>
<td>Rats</td>
<td>2-deoxy-D-[1-14C] glucose autoradiography</td>
<td>SC nic (0.1 to 1.75 mg/kg)</td>
<td>↑ nicotine rich regions, including thal, cereb, visual system, others</td>
</tr>
<tr>
<td>b) Human Studies</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Staley et al. [86]</td>
<td>16 smokers; 16 non-smokers</td>
<td>5IA-SPECT</td>
<td>Recent abstinence</td>
<td>↑ Striatum, Parietal cortex, frontal cortex, anterior cingulated, temporal cortex, occipital cortex, cerebellum</td>
</tr>
<tr>
<td>Zubiena et al. [43]</td>
<td>19 smokers</td>
<td>15O-PET</td>
<td>Nicotine containing versus denicotinized cigarettes</td>
<td>↓ global blood flow</td>
</tr>
<tr>
<td>Stapleton et al. [27]</td>
<td>4 smokers; 2 non-smokers</td>
<td>2 FDG-PETs (Fully quantified)</td>
<td>IV nic (1.5 mg) versus placebo</td>
<td>↓ global and most regions studied</td>
</tr>
<tr>
<td>Yamamoto et al. [25]</td>
<td>10 smokers</td>
<td>99mTc-ECD SPECT</td>
<td>Cigarette vs. abstinence</td>
<td>↓ global blood flow</td>
</tr>
<tr>
<td>Rose et al. [35]</td>
<td>34 smokers</td>
<td>18O-PET</td>
<td>Cigarette vs. no nic control conditions</td>
<td>↓ L frontal factor (incl prefrontal and ACC), ↓ L amygdala, ↑ rCBF</td>
</tr>
<tr>
<td>Zubieta et al. [40]</td>
<td>18 smokers</td>
<td>18O-PET</td>
<td>Nic nasal spray vs. pepper spray</td>
<td>↑ anterior thal; ↓ L ant temp and R amygdala</td>
</tr>
<tr>
<td>Domino et al. [26]</td>
<td>11 smokers</td>
<td>FDG-PET</td>
<td>Nic nasal spray vs. pepper spray</td>
<td>↑ visual cortex; ↓ normalized L ins and R inf occ ctb</td>
</tr>
<tr>
<td>Domino et al. [37]</td>
<td>18 smokers</td>
<td>18O-PET</td>
<td>Nic nasal spray vs. pepper spray</td>
<td>↑ visual cortex, ↓ rCBF</td>
</tr>
<tr>
<td>Stein et al. [36]</td>
<td>16 smokers</td>
<td>IMRI</td>
<td>IV nic (0.75-2.25 mg/70 kg wt) vs. placebo</td>
<td>↑ R NAc and bilateral amygd, cingulate, frontal lobes, thal, others</td>
</tr>
<tr>
<td>Rourke et al. [28]</td>
<td>8 smokers; 8 former smokers; 17 non-smokers</td>
<td>iodine-123 iodoamphetamine (IMP) SPECT</td>
<td>Smokers smoked the morning of the scan; other groups did not</td>
<td>↓ cortical uptake of IMP (a measure of blood flow) in current smokers compared to other groups</td>
</tr>
</tbody>
</table>

All regional changes represent normalized activity, unless otherwise stated. SC = subcutaneous; nic = nicotine; thal = thalamus; cereb = cerebellum; SPECT = single photon emission computed tomography; IMRI = functional magnetic resonance imaging; IV = intravenous; R = right; L = left; NAc = nucleus accumbens; amyg = amygdala; FDG = 18F-fluorodeoxyglucose; PET = positron emission tomography; IFG = inferior frontal gyrus; PC = posterior cingulate; ins = insula; inf occ ctb = inferior occipital cortex; ant = anterior; temp = temporal lobe; ACC = anterior cingulate cortex.
activity in the prefrontal cortex, thalamus, and visual system are among some of the findings that have been replicated in functional brain imaging studies of nicotine and cigarette administration.

**Effects of Pharmacologic Smoking Cessation Treatment on Brain Blood Flow and Metabolic Activity**

Both bupropion hydrochloride, an atypical antidepressant, and varenicline, a partial agonist at α4β2 nicotinic acetylcholine receptors, have been shown to be efficacious additions to the pharmacologic smoking cessation armamentarium [3,8,44,45]. Although a relatively new area of research, some studies have begun to investigate how these smoking cessation treatments alter both resting cerebral glucose metabolism [46] and activity in brain regions mediating cue-induced craving [47,48].

A recent study [46] utilized (18F)-fluorodeoxyglucose positron emission tomography (FDG-PET) to examine the effects of 8 weeks of treatment with bupropion HCl, practical group counseling (PGC), or pill placebo on brain function. In this study, 54 tobacco-dependent cigarette smokers underwent resting FDG-PET scanning both before and after an 8-week intervention. Costello and colleagues found that smokers treated with either bupropion HCl or PGC (compared to placebo-treated smokers) showed a reduction in glucose metabolism in the posterior cingulate gyrus, with PGC having a greater effect than bupropion.

**Table 2: Up-regulation of alpha4beta2 nicotinic acetylcholine receptors in chronic smokers and following acute, but not prolonged, abstinence.**

<table>
<thead>
<tr>
<th>Author</th>
<th>Subjects</th>
<th>Method/Task</th>
<th>Intervention</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hartwell et al.</td>
<td>32 smokers</td>
<td>fMRI/smoking vs. neutral cues</td>
<td>“allow yourself to crave” vs. “resist craving”</td>
<td>↑ activation during “crave” condition in LACC, medial prefrontal cortex, left middle cingulate gyrus, bilateral posterior cingulate gyrus, and bilateral precuneus</td>
</tr>
<tr>
<td>Goudriaan et al.</td>
<td>17 non-smoking problem gamblers; 18 non-gambling smokers; 17 non-gambling non-smoking controls</td>
<td>fMRI/gambling, smoking-related, and neutral pictures</td>
<td>none</td>
<td>Smokers with higher FTND scores showed ↑ activation in VMPC, rostral ACC, insula, and middle/superior temporal gyrus while watching smoking-related pictures compared to patients with lower FTND scores</td>
</tr>
<tr>
<td>Janes et al.</td>
<td>13 smokers</td>
<td>fMRI/smoking-related vs. neutral images</td>
<td>Pre-quit vs. extended (52 +/- 11 days) abstinence aided by nicotine replacement therapy</td>
<td>Smoking cue induced ↑ activity during abstinence in subcortical caudate nucleus and prefrontal, primary somatosensory, temporal, parietal, occipital, and posterior cingulate cortices.</td>
</tr>
<tr>
<td>Janse Van Rensburg et al.</td>
<td>10 smokers</td>
<td>fMRI/smoking-related vs. neutral images</td>
<td>Exercise vs. sitting</td>
<td>Post-exercise ↑ activity in caudate nucleus, orbitofrontal cortex, parietal lobe, parahippocampal, and fusiform gyrus and ↑ activity in “default” regions compared to controls</td>
</tr>
<tr>
<td>McCleron et al.</td>
<td>30 smokers</td>
<td>fMRI/smoking-related vs. neutral images</td>
<td>none</td>
<td>↑ reactivity to cues in right anterior cingulate and OFC in patients with higher FTND scores; ↑ reactivity in left hippocampus and left OFC in males; ↑ reactivity in cuneus and left superior temporal gyrus in females</td>
</tr>
<tr>
<td>Brody et al.</td>
<td>42 smokers</td>
<td>fMRI/smoking-related vs. neutral images</td>
<td>Crave vs. resist craving</td>
<td>↑ activation during “resist” condition in left dorsal ACC, PCC, and precuneus; ↓ activation during “resist” condition in cuneus bilaterally, right postcentral gyrus, left lateral occipital gyrus</td>
</tr>
</tbody>
</table>

**Table 3: Functional brain imaging studies of smoking-related cue reactivity.**

<table>
<thead>
<tr>
<th>Author</th>
<th>Subjects</th>
<th>Method/Task</th>
<th>Intervention</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Froeliger et al.</td>
<td>17 smokers</td>
<td>fMRI/Stroop</td>
<td>24-h abstinence vs. ad libitum smoking</td>
<td>↑ activation in postcentral gyrus, insula, and fronto-parietal cortices in absent condition during choice selection; ↑ activation in frontal pole, insula, and paracingulate cortex in absent condition during reward anticipation</td>
</tr>
<tr>
<td>Addicott et al.</td>
<td>13 smokers</td>
<td>fMRI/wharf of fortune</td>
<td>24-h abstinence vs. ad libitum smoking</td>
<td>↑ activation in postcentral gyrus, insula, and fronto-parietal cortices in absent condition during choice selection; ↑ activation in frontal pole, insula, and paracingulate cortex in absent condition during reward anticipation</td>
</tr>
<tr>
<td>Sweet et al.</td>
<td>12 smokers</td>
<td>fMRI/2-Back</td>
<td>Abstinence plus nicotine patch vs. abstinence plus placebo patch</td>
<td>Withdrawal associated with ↓ activity in left and right temporal poles and left medial frontal gyrus</td>
</tr>
</tbody>
</table>

**Table 4: Functional brain imaging studies of nicotine withdrawal.**

<table>
<thead>
<tr>
<th>Author</th>
<th>Subjects</th>
<th>Method/Task</th>
<th>Intervention</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Addition et al.</td>
<td>6 smokers</td>
<td>fMRI/2-Back</td>
<td>24-h abstinence vs. ad libitum smoking</td>
<td>↑ activation in postcentral gyrus, insula, and fronto-parietal cortices in absent condition during choice selection; ↑ activation in frontal pole, insula, and paracingulate cortex in absent condition during reward anticipation</td>
</tr>
</tbody>
</table>

**Author Subjects Method/Task Abstinence Period Results**

<table>
<thead>
<tr>
<th>Author</th>
<th>Subjects</th>
<th>Method/Task</th>
<th>Intervention</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cosgrove et al.</td>
<td>18 smokers; 20 non-smokers</td>
<td>SPECT, 5-IA</td>
<td>1 day (n=7), 1 week (n=17), 2 weeks (n=7), 4 weeks (n=11), and 6-12 weeks (n=6)</td>
<td>Compared to non-smokers, beta(2) nAChR availability in smoker striatum, cortex, and cerebellum same after 1 day abstinence, ↑ after 1 week abstinence, same at 4 and 5-12 weeks abstinence</td>
</tr>
<tr>
<td>Wüllner et al.</td>
<td>7 male smokers; 7 male non-smokers</td>
<td>PET, 2-FA</td>
<td>none</td>
<td>↑ total brain distribution volume of 2-FA in smokers, most prominently in cerebellum and brainstem</td>
</tr>
<tr>
<td>Mamede et al.</td>
<td>10 male smokers; 6 male non-smokers</td>
<td>SPECT, 5-IA</td>
<td>4 hours (n=5), 10 days (n=5), and 21 days (n=5)</td>
<td>Binding potential of nAChRs in smokers ↓ 33.5% +/- 10.5% after 4 hours abstinence, ↑ 25.7% +/- 9.2% after 10 days abstinence, and ↓ to non-smoker level after 21 days abstinence</td>
</tr>
<tr>
<td>Staley et al.</td>
<td>16 smokers; 16 non-smokers</td>
<td>SPECT, 5-IA</td>
<td>7 days</td>
<td>Compared to non-smokers, recently abstinent smokers had ↑ 5-IA uptake in striatum, parietal cortex, frontal cortex, anterior cingulate, temporal cortex, occipital cortex and cerebellum</td>
</tr>
</tbody>
</table>

SPECT = single photon emission computed tomography; 5-IA = (S)-5-[(123)I]iodo-3-(2-azetidinylmethoxy) pyridine; nAChR = nicotinic acetylcholine receptor; PET = positron emission tomography; 2-FA = 2-[18F]fluoro-3-(2S)-azetidinylmethoxy) pyridine.

**Note:** The results presented in Table 2 and Table 3 are illustrative and not exhaustive. Further research is needed to fully understand the complex interactions between smoking cessation treatments and brain function. The data presented should be interpreted within the context of the specific methodologies and patient populations studied.
bupropion HCl. This study supports the idea that bupropion HCl functions as a smoking cessation aid in part by reducing activity in the default mode network of the brain, similar to what is seen when performing a goal-oriented task.

Studies have also examined how treatments affect activity in brain regions mediating cue-induced craving. Culbertson and colleagues [47] performed a 8-week randomized, double-blind, before-after placebo-controlled trial on 30 nicotine-dependent smokers to assess alterations in regional brain activation in response to smoking-related cues from before to after treatment with bupropion HCl. Participants receiving bupropion HCl showed reduced activation in the left ventral striatum, right medial orbitofrontal cortex, and bilateral anterior cingulate cortex after treatment while actively resisting craving. A study using varenicline as the smoking cessation treatment yielded similar results. An arterial spin-labeled perfusion fMRI study scanned 22 non-abstinent, non-treatment-seeking smokers during rest and during exposure to smoking cues before and after 3 weeks of treatment with either varenicline or placebo [48]. Varenicline administration reduced smoking cue-elicited activity in the ventral striatum and medial orbitofrontal cortex. These results are remarkably similar to those observed in bupropion HCl studies, suggesting that similar mechanisms mediate the effects of both medications to improve patients’ ability to resist cue-induced craving.

Functional Imaging of Nicotinic Acetylcholine Receptors (Nicahrs)

Brain nicotinic acetylcholine receptors (nAChRs) are ligand-gated ion channels consisting of α and β subunits [49,50]. Because stimulation of nAChRs is closely associated with the effects of smoking, an important and evolving area of research is labeling these receptors using functional brain imaging. Of the many nAChR subtypes that have been discovered in the mammalian brain, the heteromeric α4β2 receptor is the most common subtype and the homomeric α7, the next most common [51]. Receptors containing the α7 subunit are reported to be central to the mediation of nicotine-induced reward, tolerance, and sensitization [52], while those containing the β2 subunit have been shown to be functionally significant in nicotine self-administration [53,54]. Animal studies have revealed that nicotinic acetylcholine receptors are located throughout the brain, with nAChR density greatest in the thalamus and descending in the following order: thalamus, basal ganglia, cerebral cortex, hippocampus, and cerebellum [55-65].

The radiotracers 2-[18F]F-A-85380, or simply 2-FA, and related compounds [66-68] are being used for PET imaging, while the radiotracer 5-[123I]iido- A85380, or 5-I-A, is being used for SPECT imaging [69-71] of α7, β2 nAChRs. Studies in both humans and non-human primates have researched nAChR distribution with these radiotracers and have demonstrated regional densities similar to those found in the animal work referenced above [66-72-76].

A recent 2-FA PET study [77] of human cigarette smokers found that smoking only one to two puffs of a cigarette led to 50% occupancy of brain α7β2 nAChRs with the occupancy lasting for at least 3.1 hours. Consistent with this finding, smoking a full cigarette led to 88% occupancy and was associated with reduced cigarette craving. A recent 5-I-A SPECT study utilizing nicotine inhalers produced similar results [78]. In this study, 13 tobacco smokers were imaged after undergoing either a nicotine inhaler (n = 9) or tobacco cigarette (n = 4) challenge. Participants receiving nicotine inhalers had an average 55.9 ± 6.4% occupancy of β2 nAChRs, compared to 67.6 ± 14.1% for those receiving a cigarette, and this occupancy lasted two to five hours post-challenge. Additionally, patients reported a significant decrease in withdrawal symptoms following inhaler use. While these findings are similar to those in the PET study previously outlined [77], the slightly different results may be accounted for by different radiotracers and imaging modalities used. Of note, smoking a low (0.6 mg nicotine) or de-nicotinized cigarette (0.05 mg nicotine) results in 79% or 26% α7β2 nAChR occupancy, respectively, demonstrating that nicotine inhalation during smoking appears to be solely responsible for receptor occupancy, with other factors having little or no effect [79]. Taken together, these studies demonstrate that smoking may alleviate withdrawal by maintaining nAChRs in a nicotine-bound state and that relatively small amounts of inhaled cigarette smoke lead to significant nAChR occupancy.

Exposure to secondhand smoke (SHS), too, can result in significant α7β2 nAChR occupancy in the brain [80]. In a recent study, tobacco-dependent cigarette smokers (n = 11) and non-smoking controls who were exposed to SHS on at least a monthly basis (n = 13) underwent two 2-FA PET scanning sessions. During these sessions, participants sat in the passenger’s seat of a car for 1 hour and were exposed to either moderate SHS (produced by a smoker who smoked 4 to 6 cigarettes in the driver’s seat of the car) or no SHS. Exposure to SHS led to an average 19% occupancy of brain α7β2 nAChRs, and smokers experienced an average 23% increase in craving following SHS exposure. These data may explain a contributing factor as to why children exposed to SHS are more likely to become teenage cigarette smokers, and adult smokers exposed to more sources of SHS are less likely to initiate or maintain abstinence.

Chronic cigarette smoking leads to up-regulation of nAChRs in brain (Table 2). Human postmortem tissue studies show that chronic smokers have an increased number of nAChRs compared to non-smokers [81,82], while former smokers have nAChR densities that are similar to non-smokers [81]. Furthermore, many laboratory animal studies demonstrate up-regulation of nAChRs in response to chronic nicotine administration [83,84]. In addition, recent studies [85,86] using single photon emission computed tomography (SPECT) or positron emission tomography (PET) demonstrate that the up-regulation of β2-containing nAChRs found in smokers normalizes to levels of non-smokers in roughly three [85,87] to four weeks or longer [88]. Calculations in one of these studies [87] demonstrate that levels of β2-containing nAChRs are within a few percent of non-smoker levels by four weeks of abstinence. In another of these studies [86], β2-containing nAChR levels did not correlate with the severity of Nicotine Dependence, severity of nicotine withdrawal, or the desire to smoke, indicating perhaps that the presence or absence of nicotine may primarily account for the up-regulation of nAChRs found in smokers.

Brain Dopamine Responses to Nicotine and Smoking

The brain dopamine (DA) reward pathway serves as a common conduit for the positive reinforcement associated with addictive drugs [89,90]. Many laboratory animal studies have shown that DA release in the ventral striatum (VST)/nucleus accumbens (NAc) accounts for the reinforcing properties of nicotine [89,90]. Using nicotine dosages that generally simulated human cigarette smoking, both lesion [91] and microdialysis [92-95] studies of rats indicate that nicotine-induced DA release is most pronounced in the VST/NAc. In fact, it is stronger in this area than the DA release observed in other related structures receiving dopaminergic input, including the dorsal striatum [95]. Chronic cigarette exposure has been found to up-regulate D1 and D2 receptor mRNA in the VST [96], and acute cigarette and nicotine exposure has
been observed to up-regulate DA transporter mRNA in the substantia nigra and ventral tegmental area (VTA) [97]. Furthermore, in vitro studies have reported increased DA release in the VST following nicotine administration [98-102].

Functional brain imaging studies have provided an exciting opportunity to substantiate and further develop these laboratory findings related to the DA system. Studies in both non-human primates and humans have repeatedly demonstrated striatal DA release following a cigarette or nicotine challenge [103-108], with most of these studies utilizing PET and 11C-raclopride (a radiotracer that specifically binds the D2/D3 DA receptor) to illustrate DA release through 11C-raclopride displacement. These, along with other studies have described a wide range of DA concentration change. For example, two studies [106,107] found that nicotine produced less 11C-raclopride displacement than amphetamine, while other studies have reported that nicotine-induced DA release is of similar magnitude to that caused by other drugs of addictive potential [93]. Additionally, associations between 11C-raclopride displacement and the subjective hedonic effects of smoking have been demonstrated [109,110], though these studies did not find an overall difference between the smoking and non-smoking conditions. A more recent study demonstrated that tobacco-dependent smokers receiving regular cigarettes exhibited a significantly greater reduction in ventral striatal 11C-raclopride binding potential and greater improvement in mood when compared to those receiving a denicotinized cigarette [103]. Interestingly, this decrease in binding potential following nicotine administration does not seem to be seen in non-smokers, perhaps reflecting enhanced DA release in smokers [111]. Disparities between these studies may be explained in part by different methodologies (e.g. cigarette smoking versus nicotine administration) and technical complexities in performing such research.

Other functional imaging studies of the DA system have reported increased 18F-DOPA uptake (a marker for increased DA turnover) [112], decreased D1 receptor density [113], both decreased 18F-DOPA uptake and D1 receptor density [114], and no alterations [115] in dopamine transporter binding in smokers. More recent studies have also demonstrated reduced availability of D2/D3 DA receptors in nicotine-dependent smokers as compared to non-smokers [116].

In summary, there is a great body of evidence that cigarette smoking and nicotine administration result in activation of the brain DA mesolimbic pathway, resulting in increased DA turnover and release in the VST/NAC. Since dopaminergic input to the NAc modulates neurotransmission through cortico-basal ganglia-thalamic circuitry [117], increases in DA concentration following smoking may explain some of the clinical effects of cigarette use.

**Functional Brain Imaging of Smoking-Related Cue Reactivity**

Dependent cigarette smokers experience craving for cigarettes minutes after their last cigarette, and this craving intensifies over the next 3 to 6 hours of abstinence [118,119]. Compared to neutral cues, smoking-related cues reliably enhance craving during this period [120], and functional brain imaging of this craving reveals important information (Table 3).

Hartwell et al. [121] examined the neural correlates of craving and resisting craving in nicotine dependent smokers. In this study, 32 nicotine-dependent smokers underwent fMRI scanning while viewing smoking-related and neutral cues. While viewing these cues, participants were instructed to "allow yourself to crave" or "resist craving." During the crave condition, increased activation was observed in the left anterior cingulate cortex (LACC), left middle cingulate gyrus, medial prefrontal cortex, bilateral precuneus, and bilateral posterior cingulate gyrus. These areas are associated with decision-making, episodic memory, and attention. During the resist condition, increased activation was observed in the LACC and areas of the prefrontal cortex. Investigators could only draw clear distinctions between the crave and resist groups after factoring in the specific strategies participants used to resist the urge to smoke, suggesting that craving and resisting the urge to smoke are intimately linked. In a similar fMRI study of forty-two tobacco-dependent smokers, Brody et al. [122] exposed participants to one of three conditions: smoking cues while being allowed to crave, smoking cues while resisting craving, and matched neutral cues. Subjects in the smoking cue resist condition demonstrated increased activation in the LACC, posterior cingulate cortex, and precuneus when compared to those in the smoking cue crave condition, providing further evidence that resisting the urge to smoke involves activation of limbic brain regions. Additionally, participants in the smoking cue resist condition had decreased activation in the left lateral occipital gyrus, right postcentral gyrus, and cuneus bilaterally, suggesting that resisting the urge to smoke is also accompanied by a relative deactivation of primary sensory and motor cortices.

Recent studies have also explored the idea that individual factors including differences in nicotine dependence, withdrawal symptoms, and gender influence brain cue reactivity [123,124]. Specifically, studies demonstrate that patients with higher Fagerström Test of Nicotine Dependence (FTND) scores show increased reactivity to smoking-related cues in the right anterior cingulate and orbitofrontal cortices [124] as well as the ventromedial prefrontal cortex, insula, and middle/upper superior temporal gyrus [123]. Male smokers exposed to smoking-related cues show increased reactivity in the left hippocampus and left orbitofrontal cortex, while females show increased reactivity in the cuneus and left superior temporal gyrus [124]. These studies suggest that individual factors play a role in cue reactivity, and that smoking cessation therapies attempting to reduce cue reactivity should consider these variables.

Smoking-related cue reactivity may change depending on smoking status. A 2009 fMRI study exposed 13 tobacco-dependent smokers to smoking-related versus neutral cues prior to a smoking cessation attempt, and again after 52 ± 11 days of abstinence aided by nicotine replacement therapy [125]. Smoking-related cues induced increased activity during abstinence in the subcortical caudate nucleus and prefrontal, primary somatosensory, temporal, parietal, occipital, and posterior cingulate cortices. Therefore, fMRI smoking cue reactivity is increased in some brain regions during extended abstinence, perhaps contributing to continued relapse vulnerability. Interestingly, exercise has been shown to reduce cigarette craving and cue-related activity in the caudate nucleus, orbitofrontal cortex, parietal lobe, parahippocampal gyrus, and fusiform gyrus, shifting activation towards structures associated within the brain default mode network [126].

**Functional Brain Imaging of Cigarette Withdrawal**

Three recent studies have examined abstinence-induced changes in dependent smokers using fMRI (Table 4) [127-129]. In Froeliger and colleagues' 2011 fMRI study, 17 smokers underwent imaging while performing an affective Stroop task, once following 24-h abstinence and once following ad libitum smoking [127]. They found that smoking abstinence increased Stroop blood-oxygenation-level-dependent response in both the rostral anterior cingulate and right middle frontal gyr. Additionally, withdrawal-induced negative affect
correlated with decreased activation in frontoparietal regions during the processing of negative emotional information, while, during negative emotional Stroop trials, negative affect predicted increased activation in frontal regions. The authors conclude that the increased activity in the frontal executive network observed during abstinence may signify a requirement to recruit additional executive functions to meet task demands.

Another 2011 fMRI study also examined effects of acute nicotine abstinence on executive function [128]. In this study, 13 smokers were scanned following 24-h of abstinence and ad libitum smoking. During scanning, participants performed a “wheel of fortune” decision-making task with probabilistic monetary outcomes. During choice selection, participants in the abstinent condition exhibited slower reaction times and increased activation in the insula, postcentral gyrus, and frontoparietal cortices than those in the ad libitum condition. During reward anticipation, participants in the abstinent condition showed increased neural activation in the insula, frontal pole, and paracingulate cortex compared to those in the ad libitum condition. Taken together, these results demonstrate that smoking withdrawal may lead to increased recruitment of frontal, parietal, and insular cortical areas during tasks requiring executive function.

Sweet et al. [129] examined the effects of nicotine withdrawal on verbal working memory (VWM) and associated changes in brain activity. Twelve smokers underwent a 2-Back VWM challenge during two fMRI sessions. Participants did not smoke prior to each session but did apply a nicotine patch before one session and a placebo patch before the other in a counterbalanced fashion. Withdrawal was associated with decreased activity in the left medial frontal gyrus and left and right temporal poles, regions associated with the default network. These results point to reduced default activity during withdrawal, perhaps compensating for disrupted neural processing.

Relevance of Other Receptors, Neurotransmitter Systems, and Genetics to Nicotine Dependence

Interestingly, some studies have implicated other receptors, neurotransmitter systems, and genetics as playing important roles in nicotine’s effect on the brain. Specifically, mu-opioid receptor availability [130], inhibition of cerebral aromatase [131], inhibition of cerebral monoamine oxidase [132], and variations in the chromosomal region encoding nAChRs [133,134] may contribute to tobacco addiction.

Monoamine oxidases (MAOs) catalyze serotonin, dopamine, and norepinephrine [135,136], and some have suggested that the increase in monoamine availability following MAO inhibition may contribute to some of the clinical effects of smoking [136]. Fowler and colleagues have demonstrated that, when compared to non-smokers and former smokers, current smokers have average reductions of 30 and 40% for MAO A and B [135]. These findings were subsequently replicated in more recent PET studies [132]. Importantly, these reductions are the result of chronic smoking behavior as opposed to smoking a single cigarette [137-139], and are not as pronounced as the reductions seen with antidepressant MAO inhibitors [140]. Peripheral MAO A and B levels were also shown to be reduced in smokers when compared to non-smokers [141]. Monoamine oxidase inhibition may enhance the rewarding aspects of smoking by both increasing dopamine availability and improving mood and anxiety [135,136]. A very recent study demonstrated increases in prefrontal and anterior cingulate cortex MAO A binding during tobacco withdrawal, leading the researcher group to hypothesize that this phenomenon may contribute to the depressed mood experienced by heavy smokers during withdrawal [142].

Other recent studies have begun to link genetic variability at the chromosomal region encoding nicotinic acetylcholine receptors (CHRNA4) with differential susceptibilities to nicotine dependence [133,134]. Remarkably, these studies demonstrate that subjects possessing rare CHRNA4 variants are less likely to respond to smoking cues or recall smoking-related memories and may be protected against nicotine dependence. Additional studies of this type are sure to add important insight into tobacco addiction and may reveal new approaches to smoking cessation therapy.

Relevance of Tobacco Smoke Constituents Other Than Nicotine in Tobacco Smoke

Throughout this review, nicotine and tobacco studies have been discussed together; however, it is important to consider that the effects of smoked tobacco may include those of the thousands of other constituents of tobacco smoke, as well as behavioral factors (such as the feel, taste, and smell of a cigarette) [13,14]. For example, Bacher and colleagues have investigated the role of harman, a substance in cigarette smoke known to inhibit MAO-A [143-145]. In their PET study, 24 healthy non-smokers and 24 otherwise healthy smokers were scanned following administration of harman labeled with carbon-11. Healthy controls underwent scanning once, while smokers were imaged after acute withdrawal and after active cigarette smoking. In participants who were heavy smokers (defined as ≥25 cigarettes per day), MAO-A density was increased in the prefrontal and anterior cingulate cortices during withdrawal when compared to both active smoking and healthy non-smoking controls. Interestingly, the difference in MAO-A density in the prefrontal and anterior cingulate cortices between withdrawal and heavy active smoking states correlated with a reduction in plasma harman concentrations in these regions. This decrease in plasma harman level, and associated increase in MAO-A binding, may contribute to withdrawal-induced depressed mood.

While acknowledging studies such as these, this review included nicotine and tobacco studies for completeness, and because nicotine is the component of cigarette smoke most intimately linked to tobacco dependence [146]. Furthermore, nicotine inhalation during smoking appears to be solely responsible for nAChR occupancy [79], which is presumed to mediate feelings of reward [89], improved attentional performance [147], reduced anxiety and irritability [148], and improved reaction times [149] following cigarette smoking. Therefore, while nicotine is the most well-studied constituent of tobacco smoke, emerging brain imaging technologies will likely focus on the influence of both nicotine and other factors on the brain mediation of tobacco dependence.

Future Directions

New radioligands targeting nAChRs are currently in development. The radiotracers 2-FA, 6-FA, and 5-I-A, which have affinity to bind to the α4β2 nAChR subtype, are available, and other radiotracers are in development for this receptor subtype [150,151]. There is a need, however, for radioligands that have affinity for other subtypes of nicotinic receptors, including the α7 subtype, which is the second most abundant in humans. Some groups have recently begun to develop and characterize such radiotracers [152-154]. Future research will likely continue to focus on developing radiotracers for imaging α2β4 nAChR in the thalamus with faster kinetics than 2-FA, 6-FA, and 5-I-A. Additionally, radiolabeled α2β4 nAChR antagonists may prove...
beneficial for developing a greater understanding of receptor binding and, ultimately, for creating pharmacological agents for smoking cessation [154,155]. Continued imaging studies with pharmacologic smoking cessation agents like varenicline may provide more information about the role of the α4β2 nicotinic acetylcholine receptor in nicotine dependence.

References


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