

Reduced Executive and Default Network Functional Connectivity in Cigarette Smokers

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Abstract: Altered functional connectivity has been associated with acute and chronic nicotine use. Connectivity alterations, specifically in the right and left executive control networks (RECN/LECN) and the default mode network (DMN), may contribute to the addiction cycle. The objective of this study was to determine if executive control network (ECN) and DMN connectivity is different between non-smokers and smokers and whether reductions in connectivity are related to chronic cigarette use. The RECN, LECN, and DMN were identified in resting state functional magnetic resonance imaging data in 650 subjects. Analyses tested for group differences in network connectivity strength, controlling for age and alcohol use. There was a significant group effect on LECN and DMN connectivity strength with smokers ($n = 452$) having lower network strengths than non-smokers ($n = 198$). Smokers had lower connectivity than non-smokers associated with key network hubs: the dorsolateral prefrontal cortex, and parietal nodes within ECNs. Further, ECN connectivity strength was negatively associated with pack years of cigarette use. Our data suggest that chronic nicotine use negatively impacts functional connectivity within control networks that may contribute to the difficulty smokers have in quitting. *Hum Brain Mapp* 36:872–882, 2015. © 2014 Wiley Periodicals, Inc.

Key words: cigarette; executive control network; default mode network; functional connectivity; nicotine; resting state; smoking

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INTRODUCTION

Despite accepted scientific evidence that tobacco use is the leading cause of preventable illness and death in the United States [National Cancer Institute, 2013], and the fact that most smokers understand the negative health consequences associated with continued use, relapse rates are higher than 85 percent in those who attempt to quit on their own [National Institute on Drug Abuse, July 2012]. The complex interaction of nicotine's addictive liability and long-term deleterious effects on cognitive processing in key brain networks likely contribute to this difficulty in breaking the addiction cycle [Jasinska et al., 2014].

Multiple large-scale connected networks have been shown to have distinct functional and behavioral domains [Fair et al., 2007; Laird et al., 2011] that can be examined with functional connectivity analyses, quantifying the connections between brain regions based on temporal correlation [Wig et al., 2011]. These networks, referred to as intrinsic connectivity networks (ICNs), have been mapped to canonical functions: auditory; primary and higher visual; language; sensorimotor; anterior and posterior salience; basal ganglia; dorsal, ventral, and precuneus default mode; and bilateral executive control systems [Beckmann et al., 2005; Chakravarthy et al., 2010; Damoiseaux et al., 2006; Greicius et al., 2003; Hampson et al., 2006; Kiviniemi et al., 2009; Seeley et al., 2007; Smith et al., 2009]. Recent research in substance users has primarily focused on networks putatively involved in addictions. In particular, the frontoparietal executive control networks [ECN; Goldstein and Volkow, 2011; Weiland et al., 2014]), as well as the internally focused default mode network (DMN), are neural circuits implicated in substance abuse [Sutherland et al., 2012].

To date only a limited number of studies have looked at these networks with specific focus on tobacco use [Andersen and Teicher, 2008]. Acute nicotine administration has been shown to depress activity in the DMN in non-smokers [Tanabe et al., 2011], and, in smokers, to enhance cingulate-cortical connectivity [Hong et al., 2009]. Nicotine replacement, in abstinent smokers, resulted in negative correlations between therapeutic effects and within-network functional connectivity in both the ECN and DMN [Cole et al., 2010]. The latter study also found that between-network functional connectivity changes, between the ECN and DMN, were positively correlated with the beneficial effects of nicotine therapy leading the authors to suggest that nicotine contributes to subjective improvements in cognition as well as reducing nicotine craving [Cole et al., 2010]. More recently, in evaluation of cessation aids in 24 smokers and 20 non-smokers, Sutherland et al. [2013] found a decrease in connectivity between the insula and DMN regions following varenicline and nicotine administration in abstinent smokers, but not in non-smokers, suggesting that cigarette use impacts connectivity which may contribute to withdrawal symptoms. In a follow-up study, Lerman et al. found that in smokers deprived of cigarettes for 24 h, connectivity between the left ECN (LECN) and the salience network

(SN) was reduced while that between DMN and SN increased [Lerman et al., 2014] demonstrating acute effects of nicotine withdrawal on functional brain systems.

Despite this work evaluating acute effects of nicotine and/or nicotine replacements, little work has been done to understand how chronic nicotine use may impact critical brain networks. It seems, in fact, crucial to first understand how repeated use of nicotine affects brain systems, which may then help clarify the mechanisms by which treatments may counteract the negative impacts of long-term use and/or withdrawal. In that vein, Janes and coworkers evaluated differences between nicotine dependent female smokers and age-matched controls finding enhanced left frontoparietal executive control network (LECN) connectivity with medial prefrontal cortical regions only in the smokers. This connectivity was correlated with striatal cue reactivity supporting a role for these networks in drug-cue responding [Janes et al., 2012], which might contribute to the addictive cycle of nicotine. However, this study was limited in generalizability as the subjects were all female and, despite significant findings, was relatively low powered including only 13 smokers and 16 controls.

Given the limited work evaluating functional connectivity effects of long term nicotine use, and the small samples in the nicotine connectivity studies published to date, this study evaluated the functional connectivity strength across and between 14 ICNs that encompass the majority of the cortical and sub-cortical gray matter [Shirer et al., 2012] to look for differences between smokers and nonsmokers. Our primary interest, in light of previous research, were the dorsal DMN and the right and left executive control networks (RECN and LECN), also referred to as the frontoparietal networks [Shirer et al., 2012], which are involved in neurocognitive and rewarding responses to nicotine. Understanding underlying functional connectivity differences between smokers and non-smokers may have important implications for the understanding and treatment of nicotine dependence. For example, the integrity of these networks, which we hypothesize will be negatively impacted by the quantity and chronicity of tobacco use, may underlie cognitive impairment more generally or contribute to cognitive impairments associated with nicotine withdrawal [Parrott et al., 1996; Xu, et al., 2005]. Therefore, using a large sample of 650 subjects, we investigated these networks in smokers compared to non-smokers. Due to the high comorbidity of smoking with alcohol use [Kalman et al., 2005], and the fact that its use [Weiland et al., 2014], as well as subject motion during scanning [Van Dijk et al., 2012] have been shown to have an impact on functional connectivity, we included these as covariates in our analyses.

MATERIALS AND METHODS

Participants

Six hundred and sixty-six individuals were recruited from the greater Albuquerque metropolitan region

through advertisements in local print, online media, and radio advertisements. Exclusionary criteria were previous brain injury or loss of consciousness for more than 5 min, a history of severe alcohol withdrawal, or a positive pregnancy test [Claus et al., 2011]. Subjects were instructed not to smoke for 2 h or drink for 24 h prior to the functional magnetic resonance imaging (fMRI) scan and had to pass a breathalyzer to participate. Individuals with excessive motion (>3 mm translational or 0.053 radians rotational movement, $n = 15$) or technical problems (wrong acquisition plane, $n = 1$) were excluded. Subjects were paid \$120 for participation in both the questionnaire and neuroimaging sessions. Written informed consent, approved by the University of New Mexico Human Research Committee, was obtained from all participants.

Substance Use

The 60-day time line follow back [TLFB; Sobell et al., 1979], an interviewer-administered assessment, was used to obtain estimates of daily cigarette, alcohol, and drug use. All participants were assessed with the alcohol use disorders identification test [AUDIT; Saunders et al., 1993] for use as a covariate. A subset of the smokers ($n = 316$) completed an additional smoking history questionnaire to collect information on frequency and duration of nicotine use. These data were used to calculate pack years (PY):

$$\text{PY} = (\text{packs of cigarettes/day from the TLFB}) \\ * (\text{years of regular smoking}).$$

Image Acquisition

Whole brain resting state fMRI was performed on a 3-T Siemens Trio scanner with a 12-channel radio frequency coil. In the scanner, tape was placed across the participants' forehead to serve as feedback for movement reduction. T_2^* -weighted functional images were acquired using a gradient-echo echo-planar imaging (EPI) sequence: TE = 29 ms, TR = 2 s, flip angle = 75°, slice thickness = 3.5 mm, slice gap = 1.05 mm, field of view = 240 mm, 64×64 matrix, voxel size = $3.75 \times 3.75 \times 3.5 \text{ mm}^3$. Resting-state scans were 5 min in duration. Subjects were instructed to keep their eyes open and fixate on a cross. High-resolution T_1 -weighted structural image were acquired with a five-echo multiecho MPRAGE sequence: TE = 1.64, 3.5, 5.36, 7.22, and 9.08 ms, TR = 2.53 s, TI = 1.2 s, flip angle = 7°, excitations = 1, slice thickness = 1 mm, field of view = 256 mm, resolution = $256 \times 256 \times 176$, voxel size $1 \times 1 \times 1 \text{ mm}$, and pixel bandwidth = 650 Hz.

Image Preprocessing

Functional images were preprocessed using an automated pipeline based around SPM 5 (<http://www.fil.ion.ucl.ac.uk/spm/software/spm5>) including realignment, slice-timing correction, spatial normalization to Montreal Neurological Insti-

tute space, reslicing, and smoothing with an 8 mm full-width half-max Gaussian kernel [Scott et al., 2011]. Time-series of cerebrospinal fluid and white-matter fluctuations, motion parameters and first derivative of motion parameters were progressively regressed from the time-series data followed by band-pass filtering (0.01–0.1 Hz) using in house scripts [Welsh et al., 2010]. The mean square derivative (MSD), which measures the existence and severity of motion, was calculated for the processed image data for each subject for use as a covariant metric in subsequent analyses [Christodoulou et al., 2013].

Network Functional Connectivity Analysis

For the primary analysis, the LECN, RECN, and dorsal DMN networks were identified using a publicly available atlas of functionally defined regions of interest [ROIs; http://findlab.stanford.edu/functional_ROIs.html; Shirer et al., 2012]; see Figure 1. That study identified nodes for these networks by applying independent component analysis to resting state data from a sample of healthy control subjects. The nodes for the LECN include: dorsolateral prefrontal cortex (DLPFC), left middle/superior frontal gyrus (MFG), left superior parietal gyrus/angular gyrus (PAR), left inferior/middle temporal gyri (TL), right crus I/crus II/Lobule VI (CE), and left thalamus (TH). The RECN nodes include: right DLPFC, right MFG, right PAR, right medial superior frontal gyrus (mSFG), left CE, and right caudate (CU). The DMN nodes include: posterior cingulate cortex (pCC), medial prefrontal cortex/anterior cingulate cortex (mPFC), midcingulate cortex (mCC) left and right angular gyri (AG), thalamus (TH), left and right hippocampus (Hpc), and right superior frontal gyrus (SFG). The time-series for each node was extracted from each subject's resting state data. Functional connections between nodes, or each pairwise correlation, was defined as an edge, and used to create a correlation matrix between the time-series of all nodes within each network for each subject. A Fisher r -to- z transformation was applied to r -values to yield z -scores. Connectivity strength, as a global measure of connectivity for each network, was calculated as the mean of all pairwise correlations between nodes within each network [Lynall et al., 2010].

For exploratory analysis, we evaluated the connectivity strength within the other 11 networks as defined by Shirer et al. [2012]: auditory, basal ganglia, ventral default mode, language, precuneus, anterior and posterior salience, sensorimotor, primary and secondary visual, and visuospatial. Further, we calculated between network connectivity for all 14 ICNs to evaluate impact on nicotine on internetwork correlations. Connectivity strength between networks was calculated for each pair of networks as the average of all between-network, node-level pairwise correlations.

Data Analyses

SPSS Statistical Software version 21 (Chicago, IL) was used to perform univariate linear regressions to evaluate

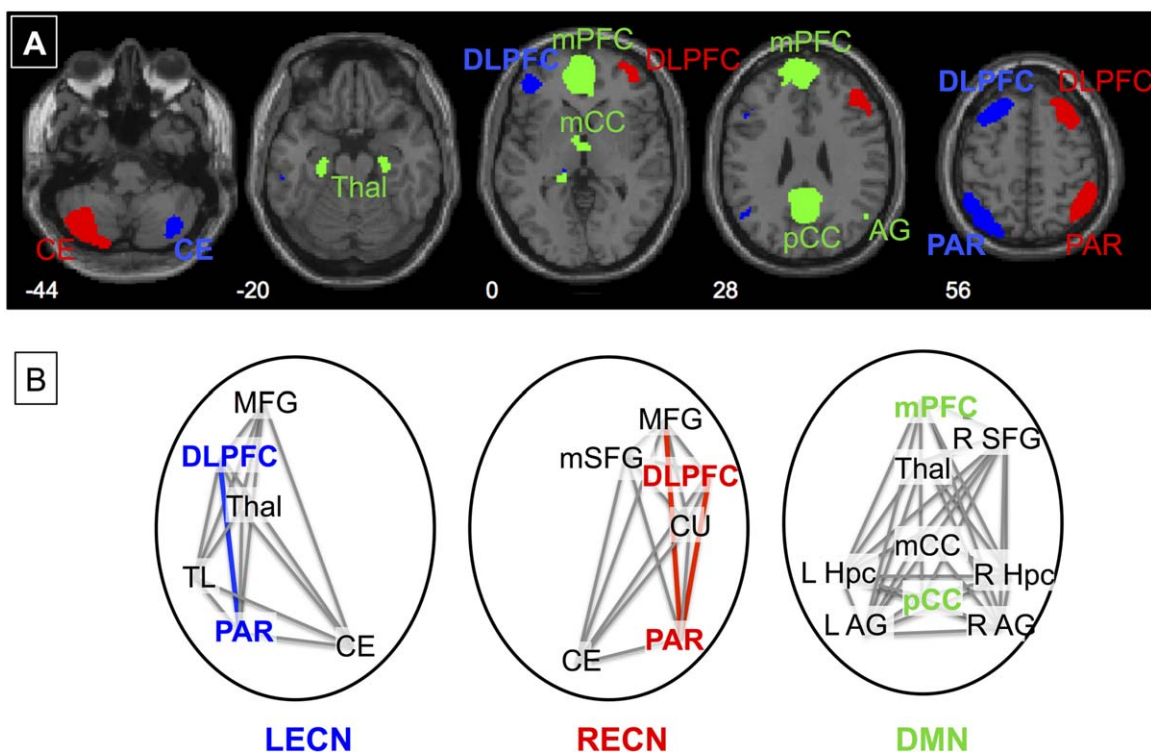


Figure 1.

A) Networks of Interest. Networks investigated include the left and right executive (LECN—blue and RECN—red) and default mode (DMN—green) networks. ROIs defining these networks were downloaded from Stanford’s FIND Lab [Shirer et al., 2012]. **B)** Key Nodes and Edges. Nodes and key hubs (bold) within the networks are shown with edges that were significantly less connected in smokers than non-smokers colored.

Nodes include the dorsolateral prefrontal cortex (DLPFC), parietal lobe (PAR), middle frontal gyrus (MFG), temporal lobe (TL), thalamus (Thal), and cerebellum (CE) in the executive networks and the medial prefrontal cortex/anterior cingulate (mPFC), posterior cingulate cortex (pCC), midcingulate cortex (mCC), superior frontal gyrus (SFG), hippocampus (Hpc), angular gyri (AG), and thalamus (Thal) in the default network.

the effect of smoking group on network connectivity strength for each of the a priori networks (R/L ECN and DMN) as the dependent variable. AUDIT scores, MSD, age, and gender were included as covariates. Significance thresholds were false discovery rate (FDR) adjusted [Benjamini and Hochberg, 1995].

The exploratory analyses evaluated the connectivity strength within the other eleven ICNs (significance threshold: $P = 0.05/11 = 0.0045$), as well as between all networks (significance threshold: $P = 0.05/(14 \times 13/2) = 0.00055$), was performed with the same regression covariates above.

As PYs was non-normal (Kolmogorov-Smirnov Test = 0.197, $P < 0.001$) and could not be normalized by transformation, Spearman’s rho was used to test correlations with connectivity measures.

ROI/Hub Analyses

We hypothesized that within the networks of interest, specific nodes would be key hubs based on theories of

functional systems [Bullmore and Sporns, 2009]. In particular, within the ECNs, the DLPFC, and PAR nodes have been shown to be key hubs involved in cognitive control and adaptation and cognitive control [Cole et al., 2013], while within the DMN, the mPFC and pCC are considered key hubs [Buckner et al., 2009]. We first verified these nodes as key hubs within our sample by examining the edges, or pairwise correlations between all nodes within each network. We first calculated the average connectivity of each node, with all other nodes within each network: the key hubs were determined as those with average connectivity significantly higher than other hubs within the same network as tested with independent sample *t* tests. For these key hubs, we then tested for negative effects of smoking on the edges connected to these hubs using similar regressions as described above for network strengths, controlling for AUDIT, MSD, age, and gender. The significance thresholds for these analyses were based on the number of edges connected to these hubs (R/L ECN: $P = 0.05/9 = 0.0056$; DMN: $P = 0.05/15 = 0.0033$).

TABLE I. Subject characteristics for subjects by smoking group

	Non-smokers	Smokers	χ^2 or t	P
N	198	452		
Males:Females	117:81	276:176	0.224	0.636
Age	30.1 (9.3)	31.4 (9.4)	-1.649	0.100
TLFB Data				
Cigarettes/Day	0	12.2 (7.8)	-22.019	0.000
Drinks/Drinking Day	5.3 (2.9)	5.4 (4.2)	-0.191	0.849
AUDIT	14.3 (7.3)	11.8 (8.8)	3.600	0.000
Race/Ethnicity			2.075	0.354
Caucasian	88	226		
Non-Caucasian	95	191		
Unknown	15	35		
Image Motion Parameter				
MSD	134854 (81578)	140481 (97818)	-0.078	0.479
N —Smoking History Data	199	305		
PYs	0	10.8 (7.0)	-21.528	0.000

TLFB, timeline follow back; AUDIT, alcohol use disorder identification test; MSD, mean square derivative. Data presented as Mean (Standard Deviation) where applicable.

RESULTS

Demographic and Behavioral Measures

Table I shows sample characteristics for the two smoking groups which were determined from the TLFB data on cigarette use. The groups were well matched on age, gender, race, and drinks per drinking days. The non-smokers, however, scored higher on the AUDIT.

Group Network Connectivity Analyses

In the primary analyses, we found a significant effect of group on LECN and DMN connectivity, with non-smokers having significantly higher network connectivity strength than smokers, (Table II, Figs. 2 and 3). There was trend for an effect of group on the RECN connectivity ($P = 0.0610$). There were no significant effects of gender on the three primary networks of interest, but, in agreement with previous work [Weiland et al., 2014], an effect of AUDIT on the LECN. Motion significantly contributed to connectivity of all three networks while age had a significant effect on the ECNs.

For the 514 subjects with smoking history data available, PYs was significantly negatively correlated with all three a priori networks, strength (LECN: $\rho = -0.153$, $P = 0.001$; RECN: $\rho = -0.105$, $P = 0.029$; DMN: $\rho = -0.089$, $P = 0.047$). This effect remained significant for the LECN ($\rho = 0.128$, $P = -0.004$) controlling for age and AUDIT, but not for the other two networks (RECN: $\rho = -0.067$, $P = 0.133$; DMN: $\rho = -0.068$, $P = 0.130$). These correlations were not significant when excluding non-smokers (P 's > 0.255).

For the exploratory analyses of the other 11 ICNs, no group effect was found. Gender had a significant effect on

the secondary visual network. Age and motion contributed to the variance in the anterior salience, visuospatial, and ventral default networks with age also contributing to the variance in the language network (Table II).

The between network exploratory analysis found no effect of smoking group on any interconnectivity strengths (Supporting Information Table I and Fig. 2). Significant effects of AUDIT, motion, and age were found between some networks but did not appear to follow a consistent pattern. An effect of gender was seen between the anterior SN and three networks: RECN, precuneus, and DMN, as well as between the DMN and the language network. Posthoc tests found that the internetwork connectivity was lower in all four cases in the females compared to male subject independent of smoking status.

Hub Analyses

Within the LECN, the PAR hub had the highest connectivity with other nodes, (0.52 ± 0.17) which was significantly higher than the next hub, the DLPFC (0.49 ± 0.18 ; $t = 9.903$, $P < 0.001$). Both of these hubs had significantly higher connectivity compared with all other LECN nodes (P 's < 0.001). Within the LECN, there was a significant effect of group on the connections between the key hubs, PAR—DLPFC ($P = 0.0054$), with smokers having lower connectivity; no other edges met the significance threshold corrected for multiple comparisons but all edges had lower marginal means for smokers compared to non-smokers. PAR—DLPFC edge strength negatively correlated with PYs ($\rho = -0.122$, $P = 0.006$). This correlation remained at trend-level ($\rho = -0.080$, $P = 0.063$) when controlling for age and AUDIT, but was not significant when excluding non-smokers ($\rho = 0.012$, $P = 0.838$).

TABLE II. Statistics for GLMs evaluating effect of smoking group on between network connectivity controlling for AUDIT score, motion, age, and gender

Network	Est'd Marginal Mean	Std Error	SMOKERS	Variance accounted for by Group			Variance accounted for by AUDIT			Variance accounted for by MSD			Variance accounted for by Age			Variance accounted for by Gender			
				F	P	Partial η^2	F	P	Partial η^2	F	P	Partial η^2	F	P	Partial η^2	F	P	Partial η^2	
Primary Analyses	LECN	0.0093	0.3960	0.0061	8.9862	0.0028	0.0138	5.2980	0.0217	0.0082	70.3515	<0.0001	0.0988	23.9559	<0.0001	0.0360	2.5364	0.1117	0.0039
	RECEN	0.0090	0.4060	0.0060	3.5230	0.0610	0.0055	2.1716	0.1411	0.0034	44.9622	<0.0001	0.0655	30.9747	<0.0001	0.0460	2.1217	0.1457	0.0033
	DMN	0.0085	0.3930	0.0056	5.3941	0.0205	0.0083	0.6354	0.4257	0.0010	43.1017	<0.0001	0.0629	2.2844	0.1312	0.0035	2.7558	0.0974	0.0043
Exploratory AUD Analyses	HV	0.4540	0.0112	0.0074	0.0269	0.8698	0.0000	0.0002	0.9887	0.0000	1.1239	0.2895	0.0017	0.8124	0.3677	0.0013	0.1699	0.6804	0.0003
	LNG	1.2660	0.0186	0.0123	0.0999	0.7520	0.0002	2.6936	0.1012	0.0042	1.3348	0.2484	0.0021	4.5564	0.0332	0.0070	21.0038	<0.0001	0.0317
	SM	0.3430	0.0077	0.0051	1.7615	0.1849	0.0027	4.4791	0.0347	0.0069	5.1992	0.0229	0.0080	14.0442	0.0002	0.0214	4.4834	0.0346	0.0069
	PS	0.2870	0.0084	0.0055	3.0792	0.0798	0.0048	3.4102	0.0653	0.0053	3.2059	0.0738	0.0050	1.7064	0.1919	0.0027	0.3600	0.5487	0.0006
	P	0.2130	0.0051	0.0034	4.8365	0.0282	0.0075	0.0173	0.8953	0.0000	1.7103	0.1914	0.0027	4.2144	0.0405	0.0065	0.6433	0.4228	0.0010
	AS	0.4910	0.0103	0.0068	0.0004	0.9838	0.0000	1.7730	0.1835	0.0028	0.9614	0.3272	0.0015	5.7021	0.0172	0.0088	0.6388	0.4244	0.0010
	VS	0.4000	0.0085	0.0056	0.7808	0.3772	0.0012	1.1337	0.7148	0.0002	11.8521	0.0006	0.0181	18.4579	<0.0001	0.0279	1.1523	0.2835	0.0018
	VDMN	0.3900	0.0080	0.0053	0.7649	0.3821	0.0012	1.8386	0.1756	0.0029	11.5736	0.0007	0.0177	23.7975	<0.0001	0.0357	1.2212	0.2695	0.0019
	BG	0.3860	0.0078	0.0051	0.0009	0.9758	0.0000	0.2410	0.6237	0.0004	11.2140	0.0009	0.0172	15.3078	0.0001	0.0233	4.0973	0.0434	0.0063
	PV	0.1760	0.0064	0.0042	0.0460	0.8302	0.0001	1.1218	0.2899	0.0017	0.6124	0.4342	0.0010	0.3813	0.5371	0.0006	0.7111	0.3994	0.0011

Networks in **bold** had significant effect of smoking group. Significance values in **bold** met thresholds for effects of other covariates, see text for details.

LECN, left executive control; RECEN, right executive control; AS, anterior salience; PS, posterior salience; DMN, dorsal default mode; vDM, ventral default mode; P, precuneus; BG, basal ganglia; SM, sensorimotor; AUD, auditory; LGN, language; V1, primary visual; V2, high visual; VS, visuospatial; AUDIT, Alcohol Use Disorder Identification Test; MSD, mean square derivative.

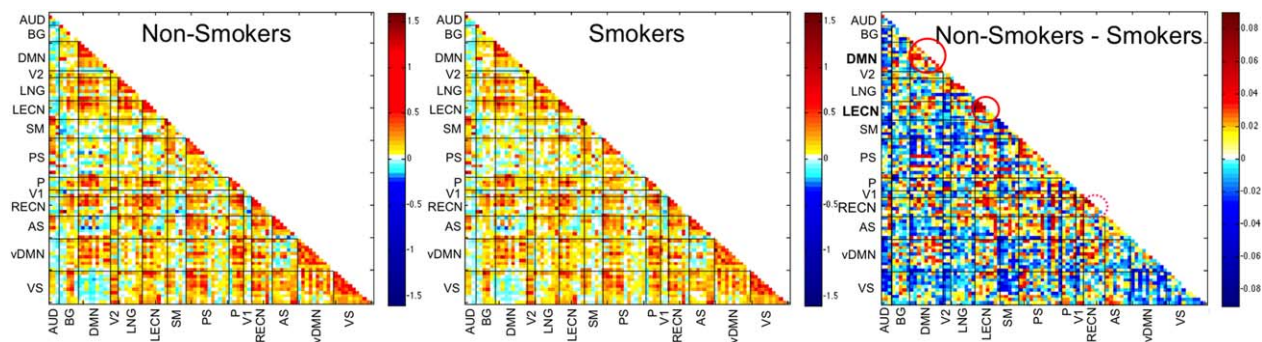


Figure 2.

Average correlation matrices of functional connectivity across all 14 ICNs by group: non-smokers ($n = 198$); smokers ($n = 452$). The difference matrix, non-smokers-smokers, identifies the networks that differed between groups with smokers having lower network functional connectivity strength: LECN and DMN. The RECN showed a trend for lower connectivity in smokers. No

differences were found in internetwork connectivity between groups. LECN, left executive control; RECN, right executive control; AS, anterior salience; PS, posterior salience; DMN, dorsal default mode; vDM, ventral default mode; P, precuneus; BG, basal ganglia; SM, sensorimotor; AUD, auditory; LGN, language; V1, primary visual; V2, secondary visual; VS, visuospatial.

Within the RECN, the DLPFC hub had the highest connectivity with other nodes, (0.60 ± 0.16) followed by, and significantly higher than, the PAR hub (0.54 ± 0.16 ; $t = 20.721$, $P < 0.001$). Both of these hubs had significantly higher connectivity compared with all other RECN nodes (P 's < 0.001). There was a significant group effect on the PAR—DLPFC and PAR—MFG (P 's < 0.001), with lower connectivity in smokers compared to non-smokers. These edges both had negative correlations with PYs (PAR—DLPFC: $\rho = -0.155$, $P = 0.001$; PAR—MFG: $\rho = -0.096$, $P = 0.033$). These correlations remained significant for PAR—DLPFC when controlling for age and AUDIT ($\rho = -0.112$, $P = 0.012$) but not for PAR—MFG ($\rho = -0.051$, $P = 0.256$). Neither edge correlation was significant when excluding non-smokers (P 's > 0.479).

For the DMN, the pCC hub had the highest connectivity with other nodes, (0.51 ± 0.15) followed by, and significantly higher than, the mPFC hub (0.49 ± 0.15 ; $t = 6.595$, $P < 0.001$); both having significantly higher connectivity than all other DMN nodes (P 's < 0.001). There were no edges that met the significance threshold corrected for multiple comparisons. Results of GLMs for edges are presented in Supporting Information Table 2; Figure 1 displays the edges different by smoking group and Figure 3 shows relationships between these edges and PYs.

DISCUSSION

Using a large sample of non-smokers and regular smokers, this study investigated the relationship between nicotine use and resting state functional connectivity of brain networks critically involved in regulation and control: the LECN, RECN, and the DMN. We found a significant effect

of group on network connectivity strength in both the LECN and the DMN with reduced connectivity strength within these networks in the smokers. Further, as recent neuroimaging research supports evidence of key hubs within networks [Buckner, et al., 2009; Bullmore and Sporns, 2009; Cole, et al., 2013], we identified the key hubs and evaluated edges connecting to these hubs within each of the networks of interest. Smokers, compared to non-smokers, had significantly lower connectivity between the two key hubs, PAR and DLPFC within both ECNs. These results suggest that smoking may contribute to the degradation of key pathways within the executive systems and this may be a critical neurobiological mechanism contributing to the etiology of nicotine addiction. In addition, we report that increasing PYs of cigarette use are associated with decreased connectivity strength of the ECNs, suggesting a negative impact of chronic smoking on functional connectivity.

While acute nicotine administration may have enhancing effects on cognition [reviewed in Swan and Lessov-Schlaggar, 2007], nicotine withdrawal is associated with negative symptoms ranging from craving, anxiety, and depression as well as cognitive deficits [Hughes, 2007]. Long term use of nicotine has also been associated with cognitive decline in several domains [Swan and Lessov-Schlaggar, 2007] but may be age dependent [Reitz et al., 2005; Starr, et al., 2007] and mediated by genetic variation [Jacobsen et al., 2006; Reitz et al., 2005]. Understanding how continued nicotine use may impact brain networks involved in cognition and control is important as failure of executive control is thought to contribute to the addiction cycle [Goldstein and Volkow, 2002; Volkow et al., 2011]. Within both ECNs, the edge strength between the key hubs, PAR and DLPFC, was reduced in smokers compared

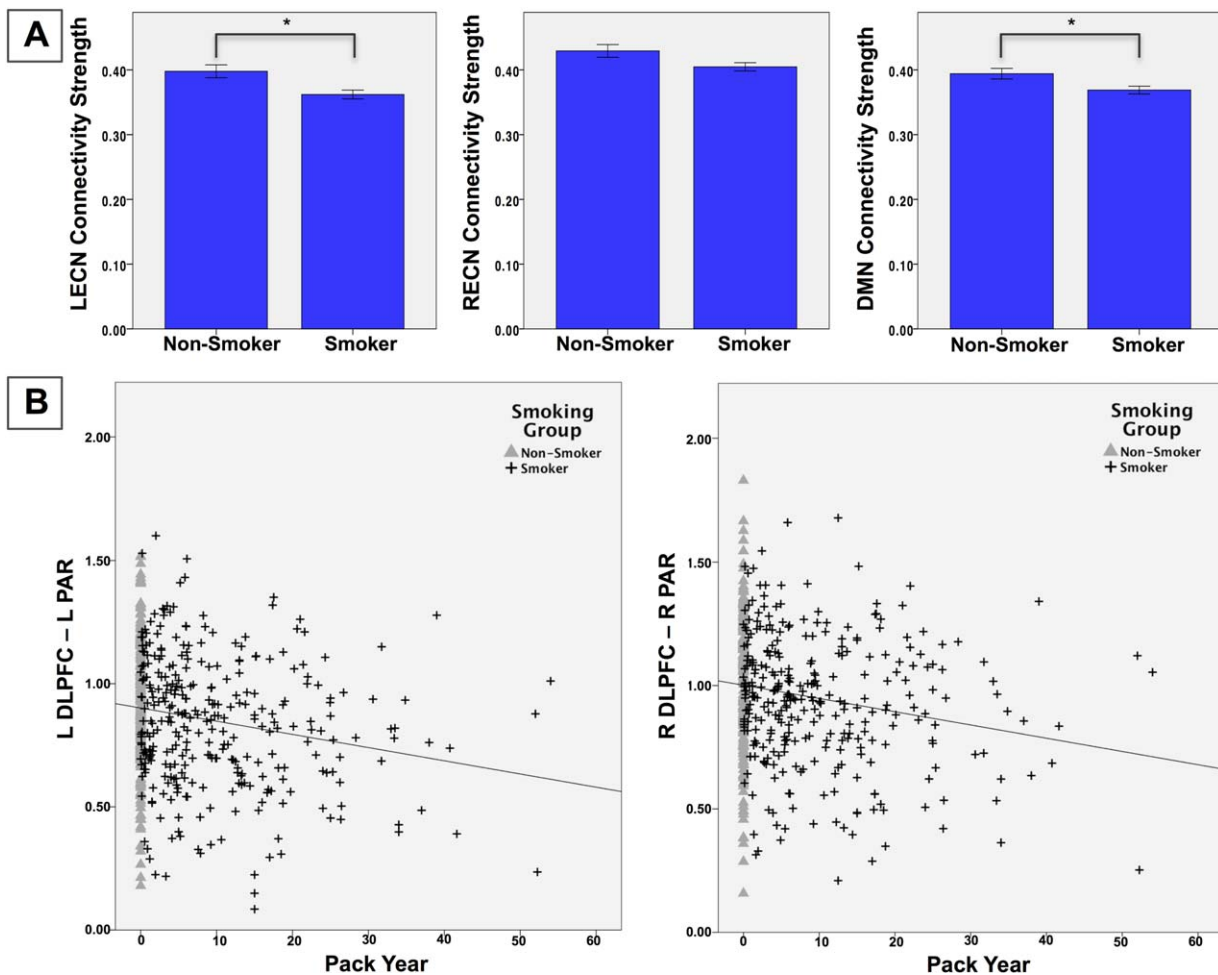


Figure 3.

A) Network Connectivity Strength by Smoking Group. Network connectivity strength for non-smokers ($n = 198$) and smokers ($n = 452$). *significant group difference, FDR corrected. **B)** Edges Associated with Chronic Cigarette Use. Scatter plots for the L/ R ECN edges between the DLPFC and PAR nodes, which corre-

lated with PYs of cigarette smoking. LECN, left executive control network; RECN, right executive control network; DMN, default mode network; DLPFC, dorsolateral prefrontal cortex; PAR, parietal lobe.

to non-smokers. This reduction in connectivity between these important nodes may represent degradation within the control systems that could contribute to the cognitive declines associated both with chronic smoking and withdrawal.

Of interest, other studies have found differences within the ECNs between smokers and non-smokers. Smokers have been shown to have greater response to smoking cues in the right DLPFC [Zhang et al., 2011] and greater bilateral PAR activation during an inhibitory control task [Luijten et al., 2013] than non-smokers. These results suggest that control networks in nicotine users may be compromised. Similarly, we suggest that the reduced connectivity of the DLPFC—PAR edge, may represent inefficient within network communication that could con-

tribute to the difficulty smokers have in quitting, particularly when dealing with cue-elicited craving [Hyman et al., 2006]. Our finding, however, is in disagreement with that of Janes et al. who reported increased within-network connectivity in the LECN in their study of female smokers [Janes et al., 2012]. These differences may be attributable to differences in methodology, or network definitions, or to the fact their study was limited to women and had a significantly smaller sample size.

The other primary network of interest, the DMN, is hypothesized to contribute to internal mentation [Buckner et al., 2008] associated with self-referential mental activity and emotional processing tasks [Gusnard et al., 2001]. Our analysis verified the mPFC and pCC as key hubs of this network in our subjects. Of particular relevance,

compromised prefrontal cortical function has been linked with substance abuse disorders in both human [Bonson et al., 2002] and animal [McFarland and Kalivas, 2001] studies involving craving [Koob, 2006]. In contrast to our findings within the ECNs, however, we did not find particular edges driving the reduced connectivity within the DMN, but rather a global reduction in within network strength. We also failed to find significant group effects in any of the other eleven functional networks we explored though there was a trend for lower connectivity in the posterior salience network (SN) in smokers than non-smokers. Neither were there significant differences in between-network connectivity, which may not be unexpected given the exploratory nature of these analyses and the conservative significance threshold targets. As females showed less connectedness between the anterior SN and several other networks, including the DMN, gender effects need to be considered in future studies concerning these networks, particularly in studies that may utilize nodes within one of these networks as a seed.

Nicotine is known to bind to nicotinic acetylcholine receptors, found in many of the regions included in the ECNs and DMN particularly the frontal and cingulate cortices [Mamede et al., 2004]. Of interest, neuroanatomical studies have found structural abnormalities associated with chronic cigarette use in these same regions. Compared to non-smokers, smokers had smaller grey matter volume and grey matter densities in bilateral PFC [Brody et al., 2004; Gallinat et al., 2006; Zhang et al., 2011] as well as the mPFC/ACC [Gallinat et al., 2006] with these volumes negatively correlated with lifetime PYs. Further, in smokers, nicotine has been shown to have detrimental effects on working memory [Park et al., 2000], a task associated with the DLPFC [Goldman-Rakic, 1987]. Mechanistically, these negative effects of nicotine, on both structure and function, are posited to be mediated by the high density of nicotine binding sites in these areas [Nordberg et al., 1989]. This is supported by a study showing that smokers, treated with varenicline, a nicotinic receptor partial agonist, had increased DLPFC activation during a working memory task [Loughead et al., 2010]. While this study reports a small effect size of smoking on network connectivity strength, the edge analyses found stronger effects associated with prefrontal node connectivity within all three networks of interest, further supporting a potential negative impact on these important regions.

Several limitations of this study should be noted including the connectivity analysis technique used. The method used here does not evaluate causal or directional relationships, but only evaluates interregional temporal correlations. Further the current analyses made use of a publicly available set of ROIs to define networks implicated in addiction [Shirer et al., 2012]. The definition of brain networks is an ongoing research question and our future work may utilize other network definitions or data driven techniques. As smokers were asked to not smoke for 2 h prior to the scan, it is possible that acute withdrawal

effects may have impacted the resting state data; future studies should more carefully control acute nicotine use. Further, as the associations between PYs and connectivity did not remain significant when non-smokers were excluded, our data does not unequivocally support a cumulative cigarette exposure effect. One interpretation may be that there is some threshold (rather than a linear dose-effect relationship) that determines the effect that all smokers in this study have reached with an end result that the correlation with the non-smokers included may indeed simply mirror the group effects. Conversely, the relatively young age of our smokers (30.1 ± 9.3 years; 10.8 ± 7.0 PYs;) may be a factor in not finding a dose effect. It may be possible that with an older sample, with corresponding greater cumulative exposure, such an association might be detectable. Finally, as this study was cross sectional, it is also unclear whether differences in functional connectivity were preexisting in the smokers; however, the associations found with smoking history, when including non-smokers, support negative impacts of chronic nicotine use on functional connectivity.

CONCLUSION

This study of 650 subjects indicates that smokers have overall reduced connectivity strength in the LECN network compared to non-smokers. In addition, in the ECNs, smokers have lower connectivity between important nodes suggesting reduced within-network communication and function associated with nicotine use. Reduced correlation of edges connected to the DLPFC node was associated with increased lifetime cigarette consumption suggesting an adverse impact on control networks with chronic smoking. Our data suggest that chronic cigarette use is negatively related to functional connectivity within control networks that may play a role in the addiction cycle and merits further study.

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