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Restoring cerebral dopamine homeostasis by electrical forepaw stimulation: an fMRI study

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Abstract

Deviation of dopamine homeostasis is known to be associated with disorders like drug addiction and Parkinson's disease. As dopamine function is tightly regulated within the basal ganglia circuitry, cortical perturbation would lead to modulation of dopaminergic activity in the striatum. We proposed and tested if somatosensory activity such as forepaw stimulation could modulate dopaminergic function. Specifically, we tested in rats if electrical forepaw stimulation (EFS) could attenuate dopamine release in the brain if dopamine is excessive, and boost dopamine release if dopamine is deficient. We had previously demonstrated that EFS effectively attenuated excessive DA concentration in the striatum. We now show in this manuscript with fMRI that EFS boosted DA release on two DA deficient conditions: 1. with quinpirole challenge, and 2. partial Parkinsonism model (PD). Quinpirole alone decreased dopamine release and thus the cerebral blood volume (CBV) that was restored by EFS. EFS also succeeded in increasing CBV in the basal-ganglia circuitry of the PD rats, but not in the controls. Context-dependent connectivity analysis showed increased connectivity during the basal state in the PD rats, compared to the controls. This "enhanced" yet abnormal connectivity of PD rats was reduced post-EFS. Our results suggest that EFS resets the deficient DA system by partially increasing DA release, in the meanwhile lessening the need for recruiting extra functional network in the basal ganglia circuitry. This study shows not only the capacity of peripheral stimulation to perturb neurotransmitter function, but also the potential of peripheral stimulation to restore neurotransmitter homeostasis.

Introduction

This aim of this study was to investigate whether that peripheral electrical stimulation has the potential to restore or maintain cerebral dopamine (DA) homeostasis. To test this hypothesis, we designed a MRI study in rodents to show whether innocuous electrical forepaw stimulation (EFS) could attenuate DA release in the brain if DA is excessive, and boost DA release if DA is deficient.

We previously demonstrated that innocuous EFS (1mA at 2Hz or 100Hz) effectively attenuated excessive DA concentration in the striatum (Chen et al., 2008a; Chen et al., 2008b). The excessive DA condition was induced pharmacologically by D-amphetamine, a

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Author contributions:

YI Chen and KK Kwong conceived the study. JQ Ren conducted the 6-OHDA lesioning and TH stain. YI Chen designed and conducted the MRI studies and data analysis, with discussion and comments from KK Kwong at all stages of the research. TJK provided feedback throughout the experiment.

robust DA releaser. We then showed that EFS effectively reduced the excessive DA release, measured directly via microdialysis and indirectly via cerebral blood volume (CBV) measurement using functional MRI (fMRI) (Chen et al., 1997; Ren et al., 2009). These findings demonstrated that EFS is capable of restoring DA homeostasis from an excessive state.

In the current study, we examined the second aim of our hypothesis that EFS is capable of restoring DA homeostasis from a deficient state by boosting DA activity. To accomplish this goal, we induced DA deficient state with two approaches: 1) DA deficiency was induced pharmacologically by quinpirole; low dose of quinpirole would bind to DA autoreceptor and reduce DA release (Starke et al., 1989; Usiello et al., 2000). 2) DA deficiency was induced by infusing neurotoxin 6-hydroxy-dopamine (6-OHDA) into the striatum to partially eliminate DA neurons --- a well established rodent model for Parkinson's Disease (PD). We then tested the effect of EFS on these two DA deficient states.

Excess DA in the brain is associated with disorders like drug addiction, and too little DA is a known problem of Parkinsonism and attention deficit hyperactivity disorder (ADHD) (Curatolo et al., 2010; Durston, 2010). The successful demonstration of EFS to restore cerebral DA release to its normal state, even for a short period, would be a step forward in uncovering how the simple step of an external peripheral manipulation can effectively modulate the central system to regulate DA release and activity in response to the stress of excessive or deficient DA.

Methods

Pharmacological agents

Desipramine, 6-hydroxy-dopamine hydrochloride (6-OHDA), ascorbic acid, and (-)-quinpirole hydrochloride were obtained from Sigma-Aldrich (St. Louis, MO, USA). The MR-contrast agent MION (Shen et al., 1993) were synthesized locally by the biochemical laboratory at the Martino's Center, MGH.

Animals

Adult Spague-Dawley rats (260-350g) were used in this study. For the partial Parkinsonian model, rats were treated with desipramine (25 mg/kg, ip) before administering 6-OHDA into the right striatum (Debeir et al., 2005; Sanchez-Iglesias et al., 2007; Yuan et al., 2006). The stereotaxic coordinates for the 6-OHDA injection was [AP, ML, DV]: [1.0, -3.0, -5.0] from bregma. 6-OHDA (6 µg/5µl) was infused at a rate of 0.5 µl/min with a 50 µl Hamilton syringe (n=5, saline solution contained 0.2% ascorbic acid) (Sanchez-Iglesias et al., 2007). Control rats (n=5) received vehicle injections. Rats were allowed to recovery for at least 4weeks before used in the fMRI study. Degree of viable DA neurons were examined by tyrosine-hydroxylase (TH) stain in the post mortem brains at the end of experiment (Chen et al., 1999).

fMRI protocol

fMRI was carried out using a Bruker 9.4T scanner (Billerica, MA) with a house made surface coil. Animals were anesthetized with 1.2-1.5% isoflurane. For forepaw stimulation was carried out with two clinical graded acupuncture needles (0.18 × 8mm, Lhasa Medical, Inc, Weymouth, MA) connecting to a Han's Acupoint Nerve Stimulator (LH202H, Neuroscience Research Center, Peking University, Beijing, China; device safety was approved by MGH Biomedcial Engineering department). One of the needles was inserted to the dorsum of the hand, to the side of the midpoint of the second metacarpal bone, in the adductor pollicis muscle. This location is near the classic acupoint LI4 (He Gu). The other

needle was inserted the middle of the distal wrist crease, between the Palmaris longus and flexor carpi radialis tendons. This location is near the classic acupoint PC7 (Da Ling). EFS was set at electrical current intensity at 1mA or 3mA and oscillating at either 2Hz or 100Hz, a 2×2 design. The Hans acupoint nerve stimulator was programmed to deliver stimulating pulses repetitively (every 0.5 sec for 2Hz and 0.01sec for 100Hz; The waveform consisted of biphasic rectangular pulses: pulse width: 1 ms for 2 Hz, 0.2 ms for 100 Hz) for 10 minutes in our study. fMRI data was acquired using conventional gradient echo sequence FLASH (TR 235ms, TE 10ms, spatial resolution $0.195 \text{ mm} \times 0.195 \text{ mm} \times 1 \text{ mm}$, no gap between slices, temporal resolution 30 sec per image set). MR contrast agent MION was used to sensitize MR signal to cerebral blood volume (CBV) changes (Chen et al., 2001). The study protocols are listed below:

1. fMRI with quinpirole challenge: DA deficiency was induced pharmacologically via quinpirole challenge. 160 repetitive fMRI time points were acquired. MION was administered at time point #10, quinpirole (0.5mg/kg iv) was administered at time point #30, and forepaw stimulation was turned “on” between time points #56-#75 (for 10 minutes). Total of 6 rats were studied (quinpirole plus EFS stimulation). In order to determine that the observed effects were due to electrical stimulation and not some other confounding factor, the control animals had needles inserted at the same points, at the same depth and retained for the same amount of time as the EFS treated group, but without applying the electrical stimulation (quinpirole without EFS stimulation, n=8). This study aimed to compare the effect of EFS under quinpirole influence: experimental group with EFS whereas control group without EFS.
2. fMRI with partial Parkinsonian rats: DA deficiency was induced surgically in PD rats. 110 repetitive fMRI time points were acquired. MION was administered at time point #10, and forepaw stimulation was turned “on” for the period between time points #50 and #70 (for 10 minutes). The control study was the vehicle treated animals. This study aimed to investigate the effect of EFS between normal and DA deficient condition by comparing between vehicle-treated rats and PD rats. Note both the study group (the PD rats) and the control group (ie, the vehicle-treated rats) received EFS stimulation in this protocol.

fMRI data was analyzed for amount of cerebral blood volume (CBV) changes, first on the individual cases, and then group averaged. fMRI data was analyzed using general linear model (GLM) on the individual study as described in (Chen et al., 2010), and then using random effect model on the group level (Worsley et al., 2002). GLM analysis was corrected for multiple comparisons using the Gaussian random fields approach (equation 3.1 using the probability density $p_3(t)$ from Table II under heading “B” in (Worsley et al., 1996)). A 2nd order correction for multiple comparisons was carried out using Monte Carlo simulation (Gold et al., 1998). A Monte Carlo simulation with 1000 iteration processed with AlphaSim program (Ward, 1997) determined the contiguous volume for a significance level of $\alpha < 0.001$, which met the overall corrected threshold of $p < 0.005$. We used AFNI function “3dclust” to carry out the 2nd order correction. Degree of activity induced by EFS task was presented by amount of CBV changes (only those brain areas with GLM statistical significance). The statistical CBV maps were masked to show only the pixels with statistical significant $P < 10^{-4}$.

For the functional connectivity analysis, time courses were divided into pre-EFS, EFS “on”, and post-EFS period for connectivity analysis. Basis function was extracted from a selected region of interest according to the rat brain atlas (Paxinos and Watson, 1997), detrended, and the context-dependent correlation coefficient was carried out using AFNI 3dDeconvolve for the whole brain map, according to method described in

http://afni.nimh.nih.gov/sscc/gangc/CD-CorrAna_old.html. The basis function was from the following brain areas: aCPu, S1FL, S2, mPFC, thalamus, hypothalamus, septum nucleus, and insular cortex --- the primary structures in the basal ganglia feedback loop.

All the experiments were carried out in accordance with the “Principles of laboratory animal care” (NIH publication No. 86-23, revised 1985) and were approved by the subcommittee on research animal care at Massachusetts General Hospital (IACUC Protocol #2008N000184).

Results

Effect of EFS on DA deficiency induced pharmacologically by quinpirole

Quinpirole alone (0.5mg/kg iv) led to a prolonged and significant CBV depression in the dopaminergic relevant brain areas, specifically in the brain areas with high density of dopamine terminals such as the caudate/putamen (CPu) and the medial prefrontal cortex (mPFC) (Figure 1A, half-live > 1hr). When EFS (either 100Hz/3mA or 2Hz/3mA) was applied, the depressed CBV was reversed toward the pre-quinpirole baseline level, with particularly eminent effect in the mPFC and the anterior CPu. The effect of EFS on the depressed CBV is shown in Figure 1B.

Effect of EFS on partial Parkinsonian brain

We then tested the effect of EFS in rats with partial Parkinsonism. Partial ablation of the dopaminergic neurons in the right striatum and substantia nigra was validated histologically with TH-stain (Figure 2). EFS at low stimulant strength (1mA, 2Hz or 100Hz) did not led to significant CBV changes in PD and vehicle-treated rats. EFS at higher yet innocuous stimulant strength (3mA, 2Hz or 100Hz) induced significant CBV increases (Figure 3) bilaterally in the PD rats. The innocuous pain perception to 3mA EFS stimulation was concluded by observing that awake rats were in general calm and not screeching for pain at 3mA EFS (data not included in this manuscript). The effect of EFS was specifically prominent in the DA-dominated areas such as CPu, nucleus accumbens (NAc), and medial prefrontal cortex (mPFC), and in the somatosensory areas such as the M1/S1 cortices and thalamus of the PD rats. For the control rats, EFS at 3mA induced significant CBV increases in fewer brain areas (part of mPFC, striatum, and thalamus) compared to the PD rats. For both PD and control rats, the degree of CBV increases was stronger for EFS at 100Hz than for EFS at 2Hz (Figure 3). Note that the lack of activity in the M1/S1 cortices in the control rats was due to the choice of anesthesia and statistical threshold ($p < 10^{-4}$). In general, isoflurane anesthesia preserves dopaminergic activity but reduces cortical response in the case of forepaw stimulation, compared to α -chloralose anesthesia that eliminates dopaminergic activity but preserves M1/S1 response to EFS (unpublished data by Dr. J. Mandeville, MGH). In order to preserve the dopaminergic activity in our study, we chose to use isoflurane anesthesia but trading off the sensitivity for cortical activity.

Changes in Functional Connectivity

As dopaminergic activity is an important modulator in the basal ganglia feedback loop, chronic changes in DA innervation would lead to alterations in neuronal activity along the feedback loop. The functional alternation may include changes in neurotransmitter activity in the downstream circuitry and recruiting compensation circuitries to accomplish the required tasks. We thus carried out functional connectivity analysis to study changes within the basal ganglia feedback loop in responding to the partial loss of DA terminals and to the EFS stimulation. The correlation analysis was carried out by correlating temporal profile among various functional structures using the “context-dependent” approach (Chen, 2009) that delineates the resting state and the “EFS-on” state (ie, with the contrast between the pre-EFS, during-EFS, and post-EFS states). While the control rats did not show significant

connectivity within the basal ganglia feedback loop, the PD rats showed increased (henceforth abnormal) connectivity during the pre-task resting state period, with enhanced connectivity between the right anterior CPU (the 6-OHDA injection site), thalamus, mPFC, and the S1_{FL}/S2 cortices. Figure 5 shows examples from the studies with 100Hz3mA EFS in PD and control rats. The enhanced pre-EFS functional connectivity were found in more than 75% of the cases including studies from all four of our imaging paradigms (ie, 1mA/2Hz, 3mA/2Hz, 1mA/100Hz, and 3mA/100Hz). Table 1A showed the cross-correlation of the “enhanced” pre-task (pre-EFS) resting-state connectivity in the PD rats (averaged from all 4 imaging paradigms). This “enhanced” functional connectivity of PD rats got further amplified when EFS was turned on as indicated in Table 1B. EFS at 3mA induced greater functional connectivity than EFS at 1mA did (Figure 6). However, the “enhanced” functional connectivity was greatly reduced post EFS task reaching the level of what was observed in the control group (Figure 5), suggesting that “enhanced” resting state connectivity was “normalized” by EFS. Group t-test analysis comparing the post-EFS connectivity using AFNI function “3dttest++” (Zhang et al., 2010) shows no significant differences between the PD and the control groups ($p>0.05$), supporting the “normalization” of the functional connectivity post EFS in the PD rats.

Discussion

Our study indicates that EFS is able to, at least temporarily, boost a depressed dopaminergic system in the basal ganglia feedback loop (Figures 1, 3, and 4). The effect of EFS on the depressed DA system is in line with the basal ganglia feedback theory that neuronal activity in the striatum maintains at a relatively stable status through cortical feedback. The basal ganglia feedback theory has been the foundation for treating dopamine related disorders. In the case of Parkinson’s disease, motor symptoms could be treated by dopamine replacement therapy (DRT) that directly compensates the diminished DA release in the striatum, or by deep brain stimulation (DBS) in various downstream structures to the striatum along the basal ganglia feedback loop. Thus, the oversimplified mechanism of using DRT and DBS to treat PD motor symptom is to facilitate functional activity along the basal ganglia feedback loop. Similar to DRT and DBS, our study shows that EFS is able to enhance the basal ganglia feedback loop. Since the approach of facilitating basal ganglia feedback loop through peripheral stimulation relies on the self-adjusting mechanism intrinsic to the basal ganglia loop, the adjustment theoretically can be applied to disorder conditions with either excessive DA (such as in addiction) or with deficient DA innervations (such as in PD). Indeed, such a phenomenon of bidirectional adjustment by EFS is supported by combining data present here for the DA deficient condition and the data published in (Chen et al., 2008a; Chen et al., 2008b) for the DA excessive condition. Interestingly, the effect of EFS on the depressed DA condition appeared to last for the duration of EFS in the quinpirole experiment, but was transient in the Parkinsonian experiment (Figure 4). The transient response may due to the fact that the Parkinsonian brain had less viable DA neurons in the striatum to release sufficient amount of DA. Thus, the therapeutic efficacy of applying EFS to treat PD symptoms remains to be studied, especially for early stage PD that still possesses a viable amount of DA neurons to bridge the feedback loop. Our data suggest, indirectly, that EFS helps in boosting dopaminergic function in the striatum in the PD brain. The CBV activity map showed the changes in activity is not restricted in the dopaminergic areas but extends to non-dopaminergic territories across the basal ganglia circuitry. Such finding suggests that the effect of EFS primarily enhances the performance the whole basal ganglia circuitry that may or may not go through dopaminergic innervation directly --- a future research with localized analysis for DA release near the partial lesioning site is needed.

As proposed previously in (Chen et al., 2008a), the effect of EFS in the quinpirol pretreated rats and in the PD rats is likely the results of a dynamic interaction of several classes of

neurotransmitter. EFS, a peripheral sensorimotor stimulation, activates the cortico-striatal project and subsequently leads to glutamate release in the striatum. The striatal glutamate either will innervate DA terminal to enhance DA release (dubbed as “PUSH” effect for pushing synaptic DA concentration up), or innervate GABA interneuron to release GABA which subsequently inhibits DA release in the striatum (dubbed as “PULL” effect for pulling synaptic DA concentration back). EFS thus maintains DA homeostasis via balancing “PUSH” and “PULL”. Thus, when there is too much DA, EFS enhances “PULL” to suppress the extra DA release; whereas when there is too little DA, EFS enhances “PUSH” to increase DA release. The “PULLING” mechanism was observed in a group of animals with excessive DA release (pretreated with D-amphetamine) via microdialysis in the striatum showing that EFS led to increases in glutamate and GABA, and decreases in DA concentration (Chen et al., 2008a). Future microdialysis study is needed to confirm the “PUSHING” mechanism in associating with the phenomenon we observed in the current fMRI study.

The involvement of the basal ganglia feedback loop prompted us to investigate the functional connectivity throughout the whole system (figures 5 and 6). We found that during basal resting period, connectivity in the basal ganglia loop was increased in the majority of PD rats. Changes in resting state connectivity appears to be a common feature among many different neurological disorders (Kwak et al., 2010; Wu et al., 2008; Wu et al., 2009). For PD, the increased and more wide-spread pre-task connectivity may indicate the need of the surviving DA neurons and its downstream circuitry to recruit additional resource to maintain or accomplish neuronal tasks during the resting state. Our result is in line with other fMRI findings reporting that increased resting state connectivity in mild to moderate PD patients was reduced after L-dopa treatment (Kwak et al., 2010). A positive correlation was also reported (Wu et al., 2009) between the degree of functional connectivity in the motor circuitry and the Unified Parkinson’s Disease Rating Scale motor score (UPDRS) in the primary motor cortex (M1), premotor area (PMA), cerebellum, and parietal cortex, suggesting that as the PD progresses, the compensatory effect in some brain regions such as in M1 and PMA is more significant (Wu et al., 2009). Increases in cortico-cortical functional connectivity in PD patients were also found with electroencephalography (EEG) and magnetoencephalography (MEG) measurements. EEG study in advanced PD patients showed increased resting-state coherence in the 10-35 Hz range (Silberstein et al., 2005) and MEG study in early stages of PD patients also demonstrated similar pathological synchronization in the range of 8-10Hz (Stoffers et al., 2008). As PD progresses, the MEG study also showed that, the cortico-cortical functional connectivity was broadened to cover a wider frequency range (Stoffers et al., 2008). All of the literature reports along with our present animal study offers evidence to the brain’s ability to compensate functional deficit by recruiting non-typical circuitry to accomplish neuronal tasks. This kind of recruiting phenomenon can often be found in other neurological disorders such as stroke (Rehme et al., 2010; Ward et al., 2003) and carpal tunnel syndromes (Napadow et al., 2006).

Our study also found that the “enhanced” functional connectivity of PD rats at resting state was further amplified by EFS. Hypothetically, this “enhancing” phenomenon is due to enhanced DA release from the surviving DA neurons upon EFS and thus a even strengthened activity along the basal ganglia feedback loop (Robinson and Whishaw, 1988; Zhang et al., 1988). Similar to our finding, an fMRI study on manganese-exposed welders with degeneration in the globus pallidus showed increased neuronal activity in M1, PMA, and supplementary motor area (SMA), indicating the need to recruit additional neural resources for performing finger tapping task successfully (Chang et al., 2010). Remarkably, our data showed that the “enhanced” functional connectivity was reduced to a level similar to the resting state connectivity in normal control rats, suggesting that the EFS reset the basal ganglia feedback loop to a homeostatic state. Although the restoration phenomenon was

quite transient in our experiment, we hope that chronic repetitive EFS treatment will establish functional plasticity in the feedback loop and eventually help in alleviating symptoms in DA related disorders.

In conclusion, our study presents an important finding that shows not only the capacity of peripheral stimulation to perturb neurotransmitter function, but also the potential to restore neurotransmitter homeostasis with peripheral stimulation. Although much is still need to be investigated, our result may shed light on some of the alternative therapies such as tender point treatment and acupuncture practice. Interestingly, the EFS sites we used are the traditional acupoints LI4 and PC7, the most used acupuncture points for treating PD, drug addiction, and many other central nerve system disorders (Napadow and Kaptchuk, 2004; Napadow et al., 2004).

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References

- Chang Y, Song HJ, Lee JJ, Seo JH, Kim JH, Lee HJ, Kim HJ, Kim Y, Ahn JH, Park SJ, Kwon JH, Jeong KS, Jung DK. Neuroplastic changes within the brains of manganese-exposed welders: recruiting additional neural resources for successful motor performance. *Occup Environ Med.* 2010; 67(12):809–815. [PubMed: 20833761]
- Chen, G. Electronic Version ed. AFNI/NIfTI Server; 2009. Context-Dependent Correlation Analysis.
- Chen YI, Brownell AL, Galpern W, Isacson O, Bogdanov M, Beal MF, Livni E, Rosen BR, Jenkins BG. Detection of dopaminergic cell loss and neural transplantation using pharmacological MRI, PET and behavioral assessment. *Neuroreport.* 1999; 10(14):2881–2886. [PubMed: 10549790]
- Chen YI, Choi JK, Xu H, Ren J, Andersen SL, Jenkins BG. Pharmacologic neuroimaging of the ontogeny of dopamine receptor function. *Dev Neurosci.* 2010; 32(2):125–138. [PubMed: 20523024]
- Chen YI, Galpern WR, Brownell AL, Matthews RT, Bogdanov M, Isacson O, Keltner JR, Beal MF, Rosen BR, Jenkins BG. Detection of dopaminergic neurotransmitter activity using pharmacologic MRI: correlation with PET, microdialysis, and behavioral data. *Magn Reson Med.* 1997; 38(3):389–398. [PubMed: 9339439]
- Chen YI, Mandeville JB, Nguyen TV, Talele A, Cavagna F, Jenkins BG. Improved mapping of pharmacologically induced neuronal activation using the IRON technique with superparamagnetic blood pool agents. *J Magn Reson Imaging.* 2001; 14(5):517–524. [PubMed: 11747003]
- Chen YI, Ren J, Wang FN, Xu H, Mandeville JB, Kim Y, Rosen BR, Jenkins BG, Hui KK, Kwong KK. Inhibition of stimulated dopamine release and hemodynamic response in the brain through electrical stimulation of rat forepaw. *Neurosci Lett.* 2008a; 431(3):231–235. [PubMed: 18178315]
- Chen YI, Wang FN, Nelson AJ, Xu H, Kim Y, Rosen BR, Kwong KK. Electrical stimulation modulates the amphetamine-induced hemodynamic changes: an fMRI study to compare the effect of stimulating locations and frequencies on rats. *Neurosci Lett.* 2008b; 444(2):117–121. [PubMed: 18722508]
- Curatolo P, D'Agati E, Moavero R. The neurobiological basis of ADHD. *Ital J Pediatr.* 2010; 36(1):79. [PubMed: 21176172]
- Debeir T, Ginestet L, Francois C, Laurens S, Martel JC, Chopin P, Marien M, Colpaert F, Raisman-Vozari R. Effect of intrastriatal 6-OHDA lesion on dopaminergic innervation of the rat cortex and globus pallidus. *Exp Neurol.* 2005; 193(2):444–454. [PubMed: 15869947]
- Durston S. Imaging genetics in ADHD. *Neuroimage.* 2010; 53(3):832–838. [PubMed: 20206707]
- Gold S, Christian B, Arndt S, Zeien G, Cizadlo T, Johnson DL, Flaum M, Andreasen NC. Functional MRI statistical software packages: a comparative analysis. *Hum Brain Mapp.* 1998; 6(2):73–84. [PubMed: 9673664]

- Kwak Y, Peltier S, Bohnen NI, Muller ML, Dayalu P, Seidler RD. Altered resting state cortico-striatal connectivity in mild to moderate stage Parkinson's disease. *Front Syst Neurosci*. 2010; 4:143. [PubMed: 21206528]
- Napadow V, Kaptchuk TJ. Patient characteristics for outpatient acupuncture in Beijing, China. *J Altern Complement Med*. 2004; 10(3):565–572. [PubMed: 15253864]
- Napadow V, Kettner N, Ryan A, Kwong KK, Audette J, Hui KK. Somatosensory cortical plasticity in carpal tunnel syndrome—a cross-sectional fMRI evaluation. *Neuroimage*. 2006; 31(2):520–530. [PubMed: 16460960]
- Napadow V, Liu J, Kaptchuk TJ. A systematic study of acupuncture practice: acupoint usage in an outpatient setting in Beijing, China. *Complement Ther Med*. 2004; 12(4):209–216. [PubMed: 15649834]
- Paxinos, G.; Watson, C. The rat brain in stereotaxic coordinates. Academic Press; San Diego: 1997.
- Rehme AK, Fink GR, von Cramon DY, Grefkes C. The Role of the Contralateral Motor Cortex for Motor Recovery in the Early Days after Stroke Assessed with Longitudinal fMRI. *Cereb Cortex*. 2010
- Ren J, Xu H, Choi JK, Jenkins BG, Chen YI. Dopaminergic response to graded dopamine concentration elicited by four amphetamine doses. *Synapse*. 2009; 63(9):764–772. [PubMed: 19484725]
- Robinson TE, Whishaw IQ. Normalization of extracellular dopamine in striatum following recovery from a partial unilateral 6-OHDA lesion of the substantia nigra: a microdialysis study in freely moving rats. *Brain Res*. 1988; 450(1-2):209–224. [PubMed: 3135914]
- Sanchez-Iglesias S, Rey P, Mendez-Alvarez E, Labandeira-Garcia JL, Soto-Otero R. Time-course of brain oxidative damage caused by intrastriatal administration of 6-hydroxydopamine in a rat model of Parkinson's disease. *Neurochem Res*. 2007; 32(1):99–105. [PubMed: 17160721]
- Shen T, Weissleder R, Papisov M, Bogdanov A Jr, Brady TJ. Monocrystalline iron oxide nanocompounds (MION): physicochemical properties. *Magn Reson Med*. 1993; 29(5):599–604. [PubMed: 8505895]
- Silberstein P, Pogosyan A, Kuhn AA, Hotton G, Tisch S, Kupsch A, Dowsey-Limousin P, Hariz MI, Brown P. Cortico-cortical coupling in Parkinson's disease and its modulation by therapy. *Brain*. 2005; 128(Pt 6):1277–1291. [PubMed: 15774503]
- Starke K, Gothert M, Kilbinger H. Modulation of neurotransmitter release by presynaptic autoreceptors. *Physiol Rev*. 1989; 69(3):864–989. [PubMed: 2568648]
- Stoffers D, Bosboom JL, Deijen JB, Wolters E, Stam CJ, Berendse HW. Increased cortico-cortical functional connectivity in early-stage Parkinson's disease: an MEG study. *Neuroimage*. 2008; 41(2):212–222. [PubMed: 18395468]
- Usiello A, Baik JH, Rouge-Pont F, Picetti R, Dierich A, LeMeur M, Piazza PV, Borrelli E. Distinct functions of the two isoforms of dopamine D2 receptors. *Nature*. 2000; 408(6809):199–203. [PubMed: 11089973]
- Ward, B. Simultaneous inference for FMRI data. Biophysics Research Institute, Medical College of Wisconsin; Milwaukee, WI: 1997.
- Ward NS, Brown MM, Thompson AJ, Frackowiak RS. Neural correlates of outcome after stroke: a cross-sectional fMRI study. *Brain*. 2003; 126(Pt 6):1430–1448. [PubMed: 12764063]
- Worsley KJ, Liao CH, Aston J, Petre V, Duncan GH, Morales F, Evans AC. A general statistical analysis for fMRI data. *Neuroimage*. 2002; 15(1):1–15. [PubMed: 11771969]
- Worsley KJ, Marrett S, Neelin P, Vandal AC, Friston KJ, Evans AC. A unified statistical approach for determining significant signals in images of cerebral activation. *Hum Brain Mapp*. 1996; 4(1):58–73. [PubMed: 20408186]
- Wu T, Long X, Zang Y, Wang L, Hallett M, Li K, Chan P. Regional homogeneity changes in patients with Parkinson's disease. *Hum Brain Mapp*. 2008
- Wu T, Wang L, Chen Y, Zhao C, Li K, Chan P. Changes of functional connectivity of the motor network in the resting state in Parkinson's disease. *Neurosci Lett*. 2009; 460(1):6–10. [PubMed: 19463891]

- Yuan H, Liang LW, Chen ZJ, Ji HR, Wang MK, Zhang HY, Li C, Xu JY. R-apomorphine protects against 6-hydroxydopamine-induced nigrostriatal damage in rat. *Neurosci Bull.* 2006; 22(6):331–338. [PubMed: 17690718]
- Zhang D, Snyder AZ, Shimony JS, Fox MD, Raichle ME. Noninvasive functional and structural connectivity mapping of the human thalamocortical system. *Cereb Cortex.* 2010; 20(5):1187–1194. [PubMed: 19729393]
- Zhang WQ, Tilson HA, Nanry KP, Hudson PM, Hong JS, Stachowiak MK. Increased dopamine release from striata of rats after unilateral nigrostriatal bundle damage. *Brain Res.* 1988; 461(2): 335–342. [PubMed: 3141002]

Figure 1.

DA deficiency was induced pharmacologically by quinpirole. (A) Quinpirole alone led to significant CBV decreases in striatum and medial prefrontal cortex. (B) EFS (100Hz3mA) was turned on 10-20 minutes after quinpirole challenge. Maps of CBV increases by EFS from the depressed CBV levels induced by quinpirole. Significant CBV increases are seen in the anterior CPU, mPFC, and thalamus.

Figure 2. TH-stain for DA neurons in the partial Parkinsonian brain. Arrows indicate the ablation of DA neurons.

Figure 3. Effect EFS at 3mA on DA deficiency in partial Parkinsonian rats. EFS had minimal CBV changes in the control rats (A), but led to significant CBV increases in the PD rats (B).

Figure 4.
CBV responses to EFS at DA deficient states. Time courses were taken from CPu.

Figure 5.

Context-dependent functional connectivity for EFS at 100Hz/3mA, with basis function from the anterior CPu (aCPu, blue circles) in lesioned hemisphere. Control rats (n=5) showed focal functional connectivity in aCPu only while PD rats (n=5) showed enhanced correlation between aCPu and 1. mPFC, 2. S1FL, 3. S2 cortex, 4. Thalamus during the pre-task resting state. PD rat also showed increased connectivity while EFS task was “on”. The post-task resting state connectivity was reduced.

Figure 6. Functional connectivity during “EFS-on” task was stronger with 3mA (n=5) stimulation than with 1mA stimulation in PD rats (n=5). Basis function was from the ipsilateral anterior CPu (aCPu) where the 6-OHDA lesioning was.

Table 1

Cross-correlation between structures in the PD group

(A) pre-task resting state							
	aCPu (cntr.)	aCPu (ipsi.)	S1 (cntr.)	S1 (ipsi.)	S2	mPFC	NAc
aCPu(ipsi)							
S1(cntr)							
S1(ipsi)							
S2							
mPFC							
NAc							
Thal.							
Ins.							

(B) EFS “on”-task							
	aCPu (cntr.)	aCPu (ipsi.)	S1 (cntr.)	S1 (ipsi.)	S2	mPFC	NAc
aCPu(ipsi)							
S1(cntr)							
S1(ipsi)							
S2							
mPFC							
NAc							
Thal.							
Ins.							

Cross-correlation between structures in the PD group, with average from all EFS paradigm. Shaded grids indicate significant correlation between the two structure ($R>0.10$, equivalent $p<0.01$). In addition, striped grids indicate the connectivity during EFS “on” period was significantly stronger than during the “pre-task” period. Control group did not show significant cross-correlations between structures in the pre-task, “on”, and most of the post-task states.