HOW HABITUAL EXERCISE CAN BENEFIT PARKINSON'S DISEASE

by

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AB, Dartmouth College, 2009

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Abstract

Exercise can improve symptoms of Parkinson’s disease (PD), including bradykinesia, balance, cognition and quality of life, but the therapeutic mechanisms of benefit are poorly understood. First, this thesis aimed to fill a gap in the literature through a systematic review on the effects of exercise on cognition in PD. This systematic review identified the benefits of exercise for cognition, but found studies seldom involved an intervention over 12 weeks, and few human studies investigated mechanisms of exercise in PD. Therefore, we next tested our novel research question: what are the effects of long-term regular (i.e., habitual) exercise on PD, and what may be the associated mechanisms of benefit for motor and non-motor symptoms? Our study compared 12 PD subjects allocated to one of two matched cohorts (n = 6 each), differing only in regular exercise levels. The primary outcome was dorsal and ventral striatal dopamine release in response to acute exercise (30 min cycling) measured using PET and displacement of \[^{11}\text{C}]\text{raclopride} (\text{RAC}) binding potential (BP). The secondary outcomes were response to reward in the ventral striatum measured with BOLD percent signal change (PSC) using fMRI, as well as clinical measures of motor function, cognition, mood and apathy. We found habitual exercisers did not release more striatal dopamine in response to acute exercise. In contrast, we found that habitual exercisers had increased RAC BP in their less affected anterior putamen post-exercise. During the fMRI card task habitual exercisers had greater BOLD PSC compared to baseline and both cohorts had greater activation during the reward phase compared to the anticipation phase. In terms of clinical outcomes, habitual exercisers had greater aerobic capacity (\text{VO}_2 \text{ peak}, confirming cohort allocation), as well as improved finger tapping, peg insertion, faster walking, less depression, more positive affect, and less apathy. In summary, habitual exercise does not
affect dopamine release in response to acute exercise, but may impact striatal RAC binding as well as response to reward in the ventral striatum. There may be dopaminergic contributions to the motor and mood benefits from habitual exercise in PD, but this topic requires further study.
Preface

Overview of my contribution to this thesis:

The work I describe in this thesis was possible through my collaborations with my primary research supervisor (Dr. Jon Stoessl), my supervisory committee (Dr. Silke Appel-Cresswell, Dr. Janice Eng, and Dr. Teresa Liu-Ambrose), as well as my many mentors and colleagues at the Pacific Parkinson’s Research Centre, the Movement Disorders Clinic, TRIUMF, the UBC PET Program, and the UBC MRI Research Centre. I have made every attempt to give credit to those who have contributed to my work through shared authorship in publications in the Acknowledgments section of this thesis. Generally, I wrote 100% of this thesis, including feedback that I incorporated from my supervisor and colleagues. I performed about three quarters of the work for the projects described in this thesis. Below is a more detailed description of the specific roles I held in these projects and the individuals who helped to make this work possible.

Contributions to Chapter 2:

A version of Chapter 2 has been published (Murray DK, Sacheli MA, Eng JJ, Stoessl AJ (2014) The effects of exercise on cognition in Parkinson's disease: a systematic review. Translational neurodegeneration 3:5). I conducted 100% of the systematic review of the literature for this paper. I also wrote the initial manuscript and all subsequent drafts, including incorporating feedback and comments from co-authors and reviewers. All authors critically revised drafts of this manuscript, and read and approved the final manuscript. All authors of this published paper declare no competing interests. This manuscript was not funded. AJS is supported by the Canada
Research Chairs, Canadian Institute of Health Research, Michael J. Fox Foundation, Pacific Alzheimer Research Foundation and the Pacific Parkinson’s Research Institute.

**Contributions to Chapter 3:**

The project described in Chapter 3 required UBC’s Research Ethics Board approval (certificate number: H13-00276). This project is a component of my group’s larger research initiative to investigate the effects of exercise in Parkinson’s disease. Dr. Stoessl is the Principal Investigator for this larger project, and the Co-Investigators include Matthew Sacheli (PhD student), Dr. Silke Cresswell, Dr. Lara Boyd, Dr. Kristin Campbell, Dr. Donald McKenzie, Dr. Martin McKeown, and Dr. Vesna Sossi.

My contribution to my group’s larger project includes identifying this topic as an area of needed research. Together with my supervisor and colleagues, I helped to design the research protocol and have been involved in all decisions to amend the protocol as needed throughout the study. I helped to implement the logistics to carry out the protocol (i.e., identifying potential research subjects, as well as scheduling and facilitating study visits). My colleague, Matthew Sacheli, was equally involved in the design and implementation of this research project. Two Pacific Parkinson’s Research Coordinators, Nicole Heffernan and Jess McKenzie, were instrumental to implement this study and conducted all clinical motor assessments using the Unified Parkinson’s Disease Rating Scale (UPDRS). Based on this larger research project on the effects of exercise in Parkinson’s disease, I designed the specific research questions studied in this thesis.
For my specific projects, I helped to collect all data through subject testing during each of their three study visits. Matthew Sacheli is an exercise physiologist and led the VO\textsubscript{2} peak testing during study visit 1, as well as the exercise session between positron emission tomography (PET) scans. I was present for and helped to coordinate nearly all PET scans during study visit 2. PET data were acquired and realigned by Carolyn English and Siobhan McCormick. I processed and analyzed all scans. As part of our centre’s standard practice to confirm validity and reliability of PET data, Siobhan McCormick also analyzed all scans. All of the data presented in this thesis are from my own analysis. As for the magnetic resonance imaging (MRI) performed during study visit 3, I helped to design the card game task performed in the scanner by modifying a task that has already been used by our group and as well as a task that has been published by another group. I made these modifications in consultation with Matthew Sacheli, Katie Dinelle, Nasim Vafai, Elham Shahinfard, Dr. Stoessl and study co-investigators. Nasim Vafai wrote the program to run this card task. I was the primary coordinator for all MRI study visits and conducted all subject testing, except for UPDRS. MRI data were pre-processed by Nasim Vafai and Katie Dinelle, in consultation with Elham Shahinfard. Nasim Vafai wrote the Matlab scripts used to analyze the data and prepared all text files I used for my analysis. Dr. Martin McKeown was very helpful to determine the best method to analyze the fMRI data. I independently analyzed all MRI data with the guidance of Matthew Sacheli, Nasim Vafai, and Elham Shahinfard, and Dr. Jon Stoessl. I received guidance on my statistical analyses from Dr. Jon Stoessl, Dr. Michael Schulzer, Edwin Mak and Matthew Sacheli.
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<tr>
<td>6-OHDA</td>
<td>6-hydroxydopamine</td>
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<td>ANOVA</td>
<td>analysis of variance</td>
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<tr>
<td>ANCOVA</td>
<td>analysis of covariance</td>
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<td>BDNF</td>
<td>brain-derived neurotrophic factor</td>
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<td>BDI</td>
<td>Beck Depression Inventory</td>
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<td>BMI</td>
<td>body mass index</td>
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<td>BP</td>
<td>binding potential</td>
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<td>CI</td>
<td>confidence interval</td>
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<tr>
<td>CNS</td>
<td>central nervous system</td>
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<td>DA</td>
<td>dopamine</td>
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<td>DAT</td>
<td>dopamine transporter</td>
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<td>DBS</td>
<td>deep brain stimulation</td>
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<td>EF</td>
<td>executive function</td>
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<td>fMRI</td>
<td>functional MRI</td>
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<td>GABA</td>
<td>(\lambda)-aminobutyric acid</td>
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<tr>
<td>GDNF</td>
<td>glial cell-derived neurotrophic factor</td>
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<tr>
<td>HRRT</td>
<td>High Resolution Research Tomograph</td>
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<tr>
<td>H&amp;Y</td>
<td>Hoehn &amp; Yahr</td>
</tr>
<tr>
<td>L-dopa</td>
<td>levodopa</td>
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<tr>
<td>LTP</td>
<td>long-term potentiation</td>
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<tr>
<td>MCI</td>
<td>mild cognitive impairment</td>
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<td>Abbreviation</td>
<td>Full Form</td>
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<tr>
<td>MDS</td>
<td>Movement Disorders Society</td>
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<td>MET</td>
<td>metabolic equivalents</td>
</tr>
<tr>
<td>MNI</td>
<td>Montreal Neurological Institute</td>
</tr>
<tr>
<td>MSN</td>
<td>medium spiny neuron</td>
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<tr>
<td>MoCA</td>
<td>Montreal Cognitive Assessment</td>
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<tr>
<td>MPTP</td>
<td>1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine</td>
</tr>
<tr>
<td>MRI</td>
<td>magnetic resonance imaging</td>
</tr>
<tr>
<td>MSN</td>
<td>medium spiny neuron</td>
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<tr>
<td>ORP</td>
<td>overall Rotarod performance</td>
</tr>
<tr>
<td>PET</td>
<td>positron emission tomography</td>
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<tr>
<td>PD</td>
<td>Parkinson’s disease</td>
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<tr>
<td>PDQ-39</td>
<td>Parkinson’s Disease Questionnaire</td>
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<tr>
<td>PDQ-39-SI</td>
<td>Parkinson’s Disease Questionnaire-39-Single Index</td>
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<tr>
<td>PANAS</td>
<td>Positive and Negative Affect Schedule</td>
</tr>
<tr>
<td>p-CREB</td>
<td>phosphorylated cyclic AMP response element binding protein</td>
</tr>
<tr>
<td>PSC</td>
<td>percent signal change</td>
</tr>
<tr>
<td>RAC</td>
<td>$^{[11]}$C raclopride</td>
</tr>
<tr>
<td>ROIs</td>
<td>regions of interest</td>
</tr>
<tr>
<td>RPE</td>
<td>rate of perceived exertion</td>
</tr>
<tr>
<td>RPM</td>
<td>revolutions per minute</td>
</tr>
<tr>
<td>rs-fMRI</td>
<td>resting-state fMRI</td>
</tr>
<tr>
<td>RTM</td>
<td>Reference Tissue Model</td>
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<tr>
<td>rTMS</td>
<td>repetitive transcranial magnetic stimulation</td>
</tr>
<tr>
<td>Abbreviation</td>
<td>Full Form</td>
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<tr>
<td>SD</td>
<td>standard deviation</td>
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<tr>
<td>SE</td>
<td>standard error</td>
</tr>
<tr>
<td>SSRI</td>
<td>selective serotonin reuptake inhibitor</td>
</tr>
<tr>
<td>TH</td>
<td>tyrosine hydroxylase</td>
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<tr>
<td>TMT A &amp; B</td>
<td>Trail-Making Test A &amp; B</td>
</tr>
<tr>
<td>TR</td>
<td>repetition time</td>
</tr>
<tr>
<td>Trk-B</td>
<td>tyrosine kinase B</td>
</tr>
<tr>
<td>TUG</td>
<td>Timed Up and Go</td>
</tr>
<tr>
<td>UBC</td>
<td>University of British Columbia</td>
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<tr>
<td>UPDRS</td>
<td>Unified Parkinson’s Disease Rating Scale</td>
</tr>
<tr>
<td>MDS-UPDRS III</td>
<td>Movement Disorders Society Unified Parkinson’s Disease Rating Scale</td>
</tr>
<tr>
<td>VS</td>
<td>ventral striatum</td>
</tr>
<tr>
<td>W</td>
<td>watts</td>
</tr>
<tr>
<td>WCST</td>
<td>Wisconsin Card Sorting Task</td>
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</table>
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My first acknowledgment goes to my supervisor, Dr. Jon Stoessl. I will be forever grateful for the incredible opportunity you have given me to learn from you as your student. I am in awe of your ability to accomplish so much, help so many patients around the world, and still make me feel like I have your undivided attention when we meet. I greatly appreciate how carefully you always listen to me, revise my work, and instantly respond to me by email. Thank you for showing me first-hand what optimal patient care, success in research, and meticulous organization looks like; you have inspired me to similarly pursue a career as a clinician and scientist.

I am very grateful for the advice, support and encouragement of my supervisory committee members – Drs. Silke Appel-Cresswell, Janice Eng, and Teresa Liu-Ambrose – as well as my external examiner, Dr. Nazanin Virji-Babul.

My education has been enriched through the opportunity to work with the wonderful group at the Pacific Parkinson’s Research Centre; thank you for your support and friendship. In particular, Shamin Babul always brightened my day and quickly took care of all the details to make my work easier. I would like to especially thank Nicole Heffernan and Jess McKenzie who were phenomenally helpful to coordinate this project and recruit the participants; thank you both for your energy, effort and incredible expertise to make the data collection come together so efficiently. Thank you to Carolyn English for her brilliance to make each scan successful. It was a pleasure getting to work with you and know you. The members of TRIUMF, the MRI Research Centre, and UBC Sports Medicine Centre were invaluable to make this research project happen.

Thank you to Matt Sacheli for his collaboration, advice, support, and wisdom. I greatly appreciate the time and energy he dedicated to facilitate my work and his above-and-beyond help which enabled me finish this thesis. My numerous fellow graduate students were also a wonderful source of advice and inspiration throughout my degree. Dr. Vesna Sossi, Nasim Vafai, Elham Shahinfard, Katie Dinelle and Siobhan McCormick were particularly helpful as they kindly taught me about the more technical aspects of the neuroimaging in this project. I really appreciate how you were always willing to meet with me and answer my questions so quickly. A special thank you goes to Dr. Martin McKeown for sharing his time, energy, expertise, and his wonderful enthusiasm for teaching students. I would like to also express my sincere gratitude to Dr. Michael Schulzer and Edwin Mak for imparting their wisdom on statistics.

To each the research participants – thank you for kindly and enthusiastically participating in my research. You have made the last two years so rewarding and have further motivated me to dedicate my life to improving patient care. Let’s keep pedaling!

Thank you to my family and friends who have always encouraged my pursuits, without their support this thesis would not have been possible. Thank you to Andrew for making me many, many late-night second dinners when I would get home too late to have the first together, and for his incredible support which helped me to finish this project. Lastly, thank you to my other half, Jen, for being behind my every big dream and my every bounce.
Dedication

To my mother - for instilling my love of learning and challenges, and who gave me the opportunity to pursue my dreams.
Chapter 1: Overview of Parkinson’s disease and exercise as a therapy

1.1 General introduction and overview of thesis

James Parkinson became the first person to describe Parkinson’s disease (PD) as a medical condition when he wrote about the “shaking palsy” in the nineteenth century (Parkinson, 1817). PD is now characterized as more than just a movement disorder as it also involves non-motor complications, which often originate years before motor dysfunction (Gaig and Tolosa, 2009). The precise etiology of PD, how to slow disease progression and ultimately how to prevent this devastating disorder remain to be determined (Schapira and Jenner, 2011). Despite successful pharmacological and surgical treatment advancements, the debilitating motor and non-motor complications and associated symptoms of PD are poorly managed over time and many therapies have serious side effects (Fox et al., 2011a; Foltynie and Kahan, 2013). The symptoms of PD, in addition to the typical elderly age of onset, can make it difficult for the individual to remain physically active. However, recently exercise has been identified as a promising therapy for PD, so maintenance of physical activity should be maximized for those with PD (Fox et al., 2011a). Evidence suggests exercise may greatly improve motor and non-motor symptoms of PD and, unlike other available therapies, may additionally provide neuroprotection and neurorestoration (Zigmond et al., 2012a; Zigmond and Smeyne, 2014).

The overarching purpose of this thesis is to determine the basis for symptomatic and disease modifying benefits of exercise, and particularly habitual exercise, on non-motor and motor symptoms of PD. The following introductory chapter provides an overview of PD, which lays
the groundwork to study how habitual exercise may improve outcomes and impact the course of this disease.

1.2 Parkinson’s disease

Parkinson’s disease is the most common age-related neurodegenerative disorder, and is the second most frequent neurodegenerative disorder after Alzheimer’s disease (Goldman and Tanner, 1998). PD typically begins between ages 40 and 70 and is slightly more common in men (Goldman and Tanner, 1998). The prevalence and incidence of PD increases with age, affecting 1% of people over age 65 and 10% of those over age 80 (Goldman and Tanner, 1998; Tanner and Aston, 2000). Furthermore, age is the most important factor that affects the development, progression and course of PD (Hindle, 2010). With the aging population, the prevalence of this chronic condition is set to increase and the impact on society will worsen.

PD is characterized by a combination of the cardinal motor symptoms (bradykinesia, rest tremor, rigidity and postural stability) (Lees et al., 2009) as well as Lewy body inclusions (protein aggregation) in some remaining nerve cells (Gibb and Lees, 1988; Tolosa et al., 2006). PD is diagnosed with a clinical exam as there is not yet a validated biomarker for the disease, although genetic, imaging and cerebrospinal fluid/blood/tissue-based biomarkers are promising candidate biomarkers (Berardelli et al., 2013; Saracchi et al., 2014). The loss of dopaminergic neurons in the substantia nigra is the primary basis for the motor and non-motor function deficits seen in PD (Lees et al., 2009). The cardinal motor symptoms are necessary for clinical diagnosis, but there are also many secondary motor symptoms, such as micrographia, shuffling gait, freezing, dystonia, hypomimia, dysarthria, dysphagia, sialorrhea, festination and glabellar reflexes.
(Jankovic, 2008). Many of these motor complications can be improved with dopaminergic monotherapy and adjuncts (Fox et al., 2011a). Non-motor complications ultimately affect virtually all people with PD and may result in greater disability (Chaudhuri and Schapira, 2009). Non-motor symptoms include mood, behavioral and cognitive abnormalities, autonomic dysfunction, sleep disturbance, sensory abnormalities (i.e., anosmia), paresthesias and pain (Jankovic, 2008). In particular, cognitive decline is among the major contributors to decreased quality of life in PD (Schrag et al., 2000; Chaudhuri and Schapira, 2009). Non-motor symptoms are generally poorly managed, lack effective therapies and are thought to be non-dopaminergic in origin (Chaudhuri and Schapira, 2009). A recent review showed some of these symptoms, such as depression, restless leg syndrome and rapid eye movement sleep behavior disorder, may have contribution from dopaminergic systems and may benefit from targeted dopamine therapy (Chaudhuri and Schapira, 2009). The complex and variable presentation of the motor and non-motor symptoms of this neurodegenerative disorder makes it difficult to manage over time. Developing a greater understanding of the pathophysiology of PD can lead to more effective therapies.

1.2.1 Pathophysiology of Parkinson’s disease

The pathophysiology of PD is mostly related to deficits in the ascending dopaminergic projections of the substantia nigra pars compacta region of the basal ganglia in the cerebrum. The hallmark of PD pathology is Lewy body inclusions from altered protein handling in the remaining substantia nigra. It remains unclear whether Lewy bodies cause PD as there are cases of genetically-based or drug-induced parkinsonism without Lewy bodies (Langston et al., 1999; Gaig et al., 2007). Lewy bodies are also found postmortem in other neurodegenerative disorders,
such as Alzheimer’s disease, dementia with Lewy bodies, and multiple system atrophy (Nussbaum and Ellis, 2003). The primary contributor to Lewy body formation is protein aggregation of α-synuclein, which has been mostly localized to presynaptic dopamine nerve terminals (Schulz-Schaeffer, 2010; Schapira and Jenner, 2011). The exact cause of α-synuclein aggregation is not known, although recent findings suggest α-synuclein may be a prion-like protein as it can be misfolded under certain conditions and then propagate toxic α-synuclein misfolding to unaffected neurons, resulting in neurodegeneration (Olanow and Brundin, 2013).

Genetics studies on familial parkinsonism and sporadic PD as well as research on animals with neurotoxic basal ganglia lesions have uncovered novel therapeutic targets from pathological events that increase susceptibility to PD. The key molecular processes identified that have been shown to cause deficits in PD are in mitochondrial maintenance, lysosome-mediated autophagy, endosomal trafficking, synaptic exocytosis and endocytosis (Venda et al., 2010; Lam et al., 2011; Piccoli et al., 2011; Vilarino-Guell et al., 2011; Matta et al., 2012; Saiki et al., 2012; Trinh and Farrer, 2013). These mechanisms are not mutually exclusive and are likely influenced by neuroinflammation, which has a role in inherited and sporadic PD (Deleidi and Gasser, 2013). Neuroinflammation results in microglial activation, demonstrated through inflammagen infusion (i.e., lipopolysaccharides) that produces selective nigral dopaminergic degeneration in the basal ganglia of rodents (Gao et al., 2002). Genes implicated in dominantly inherited late-onset PD include SNCA, which encodes α-synuclein; LRRK2, which has GTPase and kinase functions, as well as multiple roles in striatal neurotransmission, neuronal arborization, and endocytosis; and VPS35, which is a sorting protein that mediates endosome retrograde transport intracellularly (Trinh and Farrer, 2013). In addition to genetic factors, a recent meta-analysis showed that
environmental factors linked to PD include pesticides and solvents, although the specific chemical agents, how the level and length of exposure affect risk of PD has not been determined, and a cause-effect relationship has not been substantiated (Kamel, 2013; Pezzoli and Cereda, 2013).

Numerous brain functions are affected when dysfunctional molecular processes disrupt the dopaminergic circuitry of the basal ganglia. The basal ganglia are comprised of several subcortical nuclei that regulate motor planning, execution and sequencing, in addition to functions for habit formation, cognition, and emotion (Yin and Knowlton, 2006; Chakravarthy et al., 2010; Stocco et al., 2010). Other than the substantia nigra, the basal ganglia nuclei also include the striatum, subthalamic nucleus and globus pallidus. The striatum (caudate, putamen, and nucleus accumbens) and the subthalamic nucleus receive input from outside the basal ganglia, mostly from the cerebral cortex, but also from thalamic nuclei (Squire et al., 2008). Output from the basal ganglia is primarily inhibitory coming from the globus pallidus interna and the substantia nigra pars reticulata and going to the brain stem and thalamic nuclei (Squire et al., 2008). The striatum and its associated regions are further specified anatomically and functionally into the dorsal and ventral striatum. The dorsal striatum consists of the caudate nucleus and putamen and mostly coordinates voluntary motor function (Squire et al., 2008). The caudate and putamen are structurally divided by the white matter tracts of the anterior limb of the internal capsule. The ventral striatum includes the nucleus accumbens as well as the olfactory tubercle and ventromedial portions of the dorsal striatum (Selemon and Goldman-Rakic, 1985; Kunishio and Haber, 1994; Ubeda-Banon et al., 2007). The ventral striatum has been implicated in many brain functions, including anticipation and expectation of reward (de la Fuente-Fernandez et al.,
response to reward (Gottfried et al., 2002; Kirsch et al., 2003; Cox et al., 2005), appetitive learning (O'Doherty et al., 2004), evaluating learning strategies (Badre et al., 2014) and motivation (Ikemoto and Panksepp, 1999). While dopamine loss in the dorsal and ventral striatum occurs with age, striatal dopamine loss in idiopathic PD is much more severe and cannot be primarily attributed to age-related neurodegeneration (Kish et al., 1992). Unlike with aging, the pattern of dopamine loss in PD is uneven across the striatum, which has been demonstrated with an autopsy-based study that showed the most dopaminergic reduction in the caudal putamen as well as loss in the dorsal rostral portion of the caudate (Kish et al., 1988). How dopaminergic circuitry regulates the subcortical nuclei of the basal ganglia is complex, but critical to understand to develop new therapies that can mediate dopamine loss and its subsequent detriment in PD.

Dopaminergic circuits can be functionally divided into three pathways: mesolimbic, mesocortical and nigrostriatal (FitzGerald and Folan-Curran, 2002). The mesolimbic and mesocortical pathways originate in the ventral tegmental area and regulate non-motor function (Salamone et al., 2007). The mesolimbic pathway projects to the limbic system via the nucleus accumbens and is mostly involved in reward processing, which is important for learning and memory (Haber and Knutson, 2010). The mesocortical pathway projects to the frontal cortex and is generally involved in cognitive executive functions (Floresco and Magyar, 2006). The third dopaminergic circuit is the nigrostriatal pathway, which projects from the substantia nigra to the striatum and thereby modulates activity in cortico-striato-pallido-thalamo-cortical circuitry (Alexander et al., 1986). The nigrostriatal pathway is thought to coordinate the direct and indirect outputs of the basal ganglia through opposing balanced control of motor, cognitive and motivational processes.
(Mink, 1996; Smith et al., 1998; Mink, 2003; Maia and Frank, 2011; Tai et al., 2012; Cui et al., 2013). The most prevalent type of neuron in the striatum is the medium spiny neuron (MSN). MSNs use the inhibitory neurotransmitter \( \lambda \)-aminobutyric acid (GABA) and their dendritic spines contain dopamine receptors (Squire et al., 2008). There are five types of G protein-coupled dopamine receptors (D\(_1\)-D\(_5\)) that have been identified with D\(_1\) and D\(_2\) representing the primary receptors and different families (Squire et al., 2008). The classic model for how dopamine modulates the basal ganglia is that MSNs with D\(_1\) receptors increase the effect of cortical input onto striatal neurons (and of thalamic output back to cortex) through the striatonigral ‘direct’ pathway, whereas the stimulation of D\(_2\) receptor expressing MSNs of the striatopallidal ‘indirect’ pathway results in reduced thalamo-cortical output (Mink, 2003; Squire et al., 2008; Gerfen and Surmeier, 2011). The substantia nigra pars compacta is part of the direct excitatory pathway that supplies the striatum with dopamine and disinhibits basal ganglia output, resulting in facilitated movement (Wichmann et al., 2011). The indirect pathway opposes the effects of the direct pathway by inhibiting output from the thalamus, resulting in suppressed movement. In a healthy individual these parallel opposing output pathways modulate the thalamic drive to cortical targets. Neurodegeneration in the substantia nigra pars compacta in PD leads to decreased activity in the direct pathway, thus more influence from the indirect pathway and resulting decreased motor function for the individual. This model of basal ganglia circuitry is oversimplified as it only accounts for some symptoms of PD, such as bradykinesia, but does not account for other symptoms, such as tremor and rigidity (Hutchison et al., 2004). The view that these pathways are functionally opposing and anatomically distinct has been challenged by studies that have traced single cells in rats and monkeys and demonstrated impure projections in both pathways, as well as bridging by the external segment of the globus pallidus (Kawaguchi et
Further research to elucidate the pathophysiological mechanisms that cause the wide-spread motor and non-motor deficits in PD can lead to a greater understanding of the complex neural pathways of the basal ganglia and how dopamine and other neurotransmitters modulate this circuitry.

1.2.2 Therapy for Parkinson’s disease

Despite numerous approaches based on evidence from neurotoxic basal ganglia lesions in animals, there is no pharmacological therapy that has shown any convincing impact on neuroprotection or neurorestoration in humans with PD (Olanow and Schapira, 2013). Particularly in the early stages, symptomatic therapy for PD based on dopamine replacement strategies may be highly effective. Levodopa is a biochemical precursor to dopamine and, as the gold standard dopamine replacement drug, has remained the mainstay of therapy for over 40 years (Poewe et al., 2010). However, over time levodopa use may be associated with fluctuations in motor response in which patients may be severely incapacitated during “off” times, and patients may develop involuntary movements known as dyskinesias, which may be dose-limiting (Poewe et al., 2010). The long-term effects of levodopa use on PD remain uncertain (Fahn et al., 2004). Dopamine agonists and adjuncts, such as monoamine oxidase inhibitors, help to prolong the effectiveness of levodopa and reduce motor complications, but may be associated with sedation and severe impulse control disorders, and all therapies may result in confusion and hallucinations (Antonini et al., 2009; Sprenger and Poewe, 2013). There are potential non-dopaminergic drugs that could alleviate the need for levodopa, although recent randomized controlled trials on non-dopaminergic targets (e.g., serotonergic and glutamatergic systems)
suggest that no drug will be able to replace levodopa in terms of tolerability and efficacy (Kalia et al., 2013). New therapies are greatly needed to improve the lives of those with PD.

Other than immediate- and controlled-release levodopa, pharmacological therapies available for the motor symptoms of PD include efficacious monotherapies, dopamine agonist adjuncts to levodopa, therapies to prevent or delay motor complications and treatment of motor fluctuations (Fox et al., 2011b). There are more therapies available to alleviate motor symptoms than non-motor symptoms of PD, but both types of symptoms are important contributors to the health status of an individual with PD. A recent study of the clinical variables that contribute to health status in PD showed that depression has more than double the impact of motor symptoms and anxiety, separate from depression, and is also an important factor that determines health status (Hinnell et al., 2012). Non-motor symptoms are often more difficult to treat as there have not been enough controlled trials in PD to develop therapies. Efficacious and clinically useful treatments are pramipexole for depression, clozapine for psychosis, rivastigamine for dementia, and botulinum toxin A and B for sialorrhea (Seppi et al., 2011). High quality controlled studies are needed to develop therapies for other non-motor symptoms, including apathy, anxiety, sweating, urinary dysfunction, rapid eye movement sleep behavior disorder, and medication-related impulse control disorders (other than pathological gambling) (Seppi et al., 2011).

The best surgical advance for motor complications of PD is deep brain stimulation (DBS), a technique that has mostly replaced procedures that ablate brain structures, such as the globus pallidus (Okun, 2013). DBS involves implanting leads into specific regions of the brain and then applying continual electrical current to those areas. The main targeted regions include the
subthalamic nucleus and the globus pallidus interna, but consensus has not been reached on the best surgical technique or targets (Wagle Shukla and Okun, 2014). DBS is the preferred surgical treatment for carefully screened candidates because it can be programmed based on the individual’s needs, it is reversible, and can be implanted bilaterally. Over 100,000 devices have been implanted since DBS was approved for use in PD and essential tremor by the US Food and Drug Administration in 2002. However, there are significant adverse effects associated with this powerful therapy, including decline in cognition, mood and speech (Bronstein et al., 2011; Castrioto et al., 2014). DBS may improve non-motor symptoms and quality of life in some patients, but the evidence has not been well established (Ashkan et al., 2013). Overall, DBS may be an effective treatment option for PD, but only after other interventions have been considered as it is highly invasive and only works in those who undergo rigorous selection for the procedure. DBS has offered hope for greater well-being to many in the PD community, but a better treatment option available to all patients without risk of serious complications is still needed.

There are some potentially effective non-pharmacological therapies for the management of PD, but more trials are needed to further understand their effects. For motor symptoms, the Movement Disorders Society (MDS) Task Force on Evidence-Based Medicine has indicated that physical therapy is likely efficacious and a possible useful symptomatic adjunct therapy (Fox et al., 2011b). The MDS Task Force also found that trials on exercise therapy were of limited quality, but that exercise is an important area for further study (Fox et al., 2011b). Indeed, many trials since the Task Force’s publication in 2011 have identified exercise as promising treatment
for PD, and understanding how exercise can benefit PD could even lead to developing preventative therapies.

1.3 Exercise as a therapy for Parkinson’s disease

1.3.1 Effects of exercise on older adults

In order to understand how exercise can affect symptoms and may modify PD, it is important to first review the general effects of exercise in older adults without PD. Age-related cognitive decline in those without PD has been attributed to typical structural and functional physiological changes that take place as the brain ages (Reuter-Lorenz and Park, 2010). Structural changes that increase with age include atrophy, reduced gray matter volume and decreasing white matter integrity primarily in the frontal cortex and in the hippocampus of the medial temporal lobe (an area important for memory) (Drag and Bieliauskas, 2010). Functional physiological changes that occur with age include preserved neural mechanisms, but overall declining activity is observed in regions such as the medial temporal lobe (Grady, 2008). With declining activity in some regions, other brain areas then overcompensate, including from regions such as the ventral and dorsal prefrontal cortex, which are areas important for coordinating thoughts, actions and executive functions (Grady, 2008). These changes and overcompensation by some brain areas can be detrimental, but exercise may help to mitigate the effects of age on the brain.

Exercise is traditionally one of two types: 1) aerobic, such as cycling, running, and jumping rope, or 2) resistance training, such as weight lifting. Other types of exercise are generally milder, and include stretching- and balance-based activities. Interventional, longitudinal and cross-sectional studies show that healthy older adults who are physically active have better cognitive function.
(Dustman et al., 1984; Hillman et al., 2002; Barnes et al., 2003; Renaud et al., 2010). The type of exercise that leads to the most gains is not yet known, but the best evidence for the potential for physical activity to improve cognition is from longitudinal and interventional studies showing benefits specifically from vigorous, aerobic, and long-term (i.e., habitual) participation in physical activity (Dustman et al., 1984; Rikli and Edwards, 1991; Hawkins et al., 1992; Kramer et al., 1999; Aichberger et al., 2010). Aerobic exercise has been shown to modulate the brain through increased neurotrophic factors, neural plasticity, neuroprotection, and development of synapses (Barde, 1994; Neeper et al., 1995; Lu and Chow, 1999; Cotman and Engesser-Cesar, 2002). Imaging studies have also shown aerobic exercise enhances frontal, prefrontal and parietal cortical areas in older adults, which are the areas most associated with age-related cognitive decline (Colcombe et al., 2003; Colcombe et al., 2004). In particular, positron emission tomography (PET) neuroimaging has shown cycling increases brain activity in the primary sensory and motor cortices and anterior cerebellum (Christensen et al., 2000). Aerobic exercise may also increase hippocampal volume and related spatial memory improvement, as well as serum brain-derived neurotrophic factor (BDNF) (Erickson et al., 2011). A recent study specifically on older women with probable mild cognitive impairment (MCI) also found aerobic exercise can improve hippocampal volume (Ten Brinke et al., 2014). Other types of exercise and combinations of exercise varieties have also shown benefits in older adults. A randomized-controlled trial in cognitively normal and sedentary older women looked at the effects of a multimodal exercise program and showed improved cognitive function (i.e., memory and executive function), physical performance and increased plasma BDNF (Vaughan et al., 2014). The 16-week multimodal program entailed cardiovascular and strength training as well as motor fitness training (e.g., agility, balance, coordination) for 60 minutes twice per week. The control
group was put on a waiting list, so this study did not control for the potential benefit of social interaction associated with the intervention. The increased levels of plasma BDNF suggest neurogenesis may contribute to the cognitive improvements from exercise in older individuals.

In addition to studies on aerobic exercise, there are some controlled reports of resistance-based exercise benefitting brain function in older adults. A randomized-controlled trial on resistance training improved executive function, conflict resolution and selective attention in older females (Liu-Ambrose et al., 2010). This group also showed resistance exercise can increase functional plasticity in cortical brain regions associated with response inhibition, including the anterior left middle temporal gyrus and the left anterior insula into the lateral orbitofrontal cortex (Liu-Ambrose et al., 2012). Functional plasticity was measured by changes in hemodynamic activity in 12 regions of interest during functional magnetic resonance imaging (fMRI). Women who participated in this resistance training program twice per week for 12 months had improved behavioral measures of executive function and functional plasticity, but these effects were not present in those who trained only once per week. Given there is not a specific prescription of exercise that has been identified as most beneficial, it is likely that exercise affects a variety of neurological mechanisms and the effects of exercise are also individualized.

Another way that exercise may improve the brain health of older adults is through dopaminergic systems. Brain mechanisms linked to impaired motor neuron activation during prolonged exercise includes decreased striatal dopamine release (Nybo & Seecher, 2004). Exercise may be able to combat the decreased dopamine release that is seen with decline in physical activity in various species (Ingram, 2000). Aerobic exercise has been shown to increase dopamine release in normal rats (Hattori et al., 1994), but human studies using PET so far have not demonstrated
an increase in dopamine release after vigorous aerobic exercise in healthy adults (Wang et al., 2000). However, these adults were not dopamine-depleted, so it could be that exercise only increases dopamine availability, and to an extent that is measurable with PET, in cases of deficiency. The benefits of exercise in adults may result from combined activation of the motor and non-motor dopaminergic pathways, including the nigrostriatal, mesolimbic and mesocortical circuits, as well as non-dopaminergic mechanisms.

1.3.2 Effects of exercise in Parkinson’s disease

Exercise not only improves brain health in normal ageing, but also benefits neurological function for those with various central nervous system (CNS) disorders, and may additionally promote neuroprotection in PD (Zigmond et al., 2012a). Exercise is relatively easy and inexpensive to implement, is not associated with negative side effects when practiced safely, and can mitigate general age-related health decline. The literature strongly supports exercise as a promising therapy for PD, although the best frequency, intensity, type and timing of exercise as a therapy for PD are not currently known. Vigorous physical exercise has also been weakly associated to a lower risk of PD (Campdelacreu, 2012). However, regular exercise alone cannot prevent PD as regular exercisers are not immune to developing PD. Once an individual is diagnosed with PD they should be encouraged to continuing exercising, or start exercising if they have been sedentary, as the benefits of exercise have now been purported in the PD community for decades. It is only recently have there been controlled studies demonstrating symptomatic benefits in bradykinesia, postural balance and quality of life (Ridgel et al., 2009b; Allen et al., 2011a; Ridgel et al., 2012a; Lauhoff et al., 2013). There have been unsubstantiated suggestions that exercise may improve cognition and mood in PD (Tanaka et al., 2009a; Cruise et al., 2011b;
It is not yet known if exercise can modify disease and what neuroprotection exercise may provide.

### 1.3.3 Possible mechanisms of exercise in Parkinson’s disease

Aerobic exercise has demonstrated benefits in motor and non-motor function in animal models of neuroprotection in PD (Tillerson et al., 2003; Ahlskog et al., 2011; Petzinger et al., 2013). Animal models of neurological diseases effectively improve our understanding of neural anatomy and circuitry; however, the extent of the similarities and differences between rodent and human molecular mechanisms of exercise are still unknown (Kramer & Erickson, 2007). Exercise likely has an impact on circuitry within and outside the basal ganglia in PD. Indeed, a study on cerebral blood flow in rodents with a basal ganglia lesion showed that exercise may lead to functional reorganization of basal ganglia-thalamocortical circuits, with increased dorsal striatal and secondary motor area activation (Wang et al., 2013). Most of the research on the potential for exercise to be neuroprotective in PD is from studies of basal ganglia lesions in animals demonstrating increased neurotrophic factors, such as BDNF and glial cell-derived neurotrophic factor (GDNF) levels following exercise (Cohen et al., 2003; Lau et al., 2011; Wu et al., 2011; Gerecke et al., 2012; Petzinger et al., 2013; Real et al., 2013). Three studies to date have investigated BDNF levels from serum samples in PD patients. Two studies investigated levels of BDNF in PD not related to exercise; one study showed BDNF in PD is lower on average in PD compared to healthy controls (Scalzo et al., 2010) and the other study showed BDNF levels may decrease proportionally to striatal dopamine transporter binding (DAT) (Ziebell et al., 2012). The third study found intensive rehabilitation including aerobic exercise for 28 days increased BDNF in PD compared to PD control subjects (Frazzitta et al., 2014).
However, the mechanisms underlying the motor and non-motor benefits of exercise in human PD remain poorly understood.

While neurotrophic factors may provide exercise-induced neuroprotection in PD, the major factor contributing to the motor deficits of PD is decreased dopamine release from nigrostriatal terminals (Smith and Villalba, 2008). Understanding how exercise affects dopaminergic mechanisms may shed more light on how exercise could be therapeutic in PD. While exercise has not been shown to release dopamine in healthy individuals without PD (Wang et al., 2000), stimulating the human prefrontal or motor cortex with repetitive transcranial magnetic stimulation (rTMS) causes dopamine release in the striatum, although the response is lower in PD (Strafella et al., 2001; Strafella et al., 2003; Strafella et al., 2005b). Increased dopamine release was further demonstrated in the contralateral dorsal putamen of healthy individuals and comparable increase in the ipsilateral dorsal putamen of individuals with PD in a study that used a simple motor activity (foot extension/flexion) and $[^{11}\text{C}]$raclopride (RAC) binding during PET (Ouchi et al., 2002). The ipsilateral activation may indicate the lack of coordinated movement resulting from hypokinesia in the PD subjects. A recent study showed the effect of subthalamic nucleus deep brain stimulation on the release of dopamine during exercise in PD (Nozaki et al., 2013). Dopaminergic activation was increased in the ventromedial striatum and not in the putamen, which indicates possible compensation from the non-motor reward-based dopaminergic pathway. The effects of exercise in PD may be from increased dopamine release, thereby contributing to improved motor function in the dorsal striatum, and to enhanced mood and reduced apathy in the ventral striatum. Exercise may also enhance dopamine transmission via upregulation of dopamine D$_2$ receptors (Petzinger et al., 2007; Vuckovic et al., 2010a). If an
individual with PD regularly exercised and exercise released dopamine consistently, that could lead to sustained coordination of corticostriatal pathways through upregulation of D2 receptors, which could strengthen dwindling dopaminergic connections. However, if exercise changes the expression of D2 receptors it could also change the expression of D2 receptors on the pre-synaptic terminal that serve as auto-receptors and regulate the amount of dopamine in the synaptic cleft. Thus, upregulation of D2 on the pre-synaptic terminal from participation in regular exercise could plausibly decrease dopamine transmission. Recent research on mice with a basal ganglia lesion showed that 30 days of treadmill exercise increased density of spines and arborization of both MSNs containing D2 and D1 receptors (Toy et al., 2014). As the mice were euthanized and stained immediately following the exercise program, it remains to be explored whether these changes could persist over time and if this finding can be translated to humans. However, these results on how exercise can change the brain are promising as they are consistent with other animal-based research, although not on PD, that has shown exercise can increase spine density in the hippocampus and the cerebellum (Pysh and Weiss, 1979; Stranahan et al., 2007). How exercise could affect plasticity in the basal ganglia in human PD is an important topic that merits further study.

Determining the mechanism of exercise in PD may lead to insight into how physical activity interacts with, and ideally contributes to, other therapies for PD, such as medications and surgical stimulation of the basal ganglia (i.e., DBS). There may also be individuals with PD who are more susceptible to the benefits from exercise based on a genetic basis of their disease or genetic factors that may mediate response to exercise. These topics are important to consider, but beyond the scope of this thesis. Most of all, determining the mechanisms of exercise in PD could
highlight the mechanisms of the disease, which may identify new therapeutic targets and ultimately move us closer to understanding the cause, prevention and a cure for PD.

1.4 General purpose and design of thesis

To evaluate the basis for symptomatic and disease modifying benefits of exercise on motor and non-motor symptoms of PD, I examined how regular participation in exercise affects PD. In Chapter 2, I present a systematic review of the literature to specifically understand the effects of exercise on an important non-motor symptom of PD, cognition, which is a topic that has received little research attention. In order to study the mechanism of exercise in PD, in Chapter 3 I present the findings of our research comparing two cohorts of PD subjects: 1) those who participate in regular exercise (i.e., habitual exercisers), and 2) non-regular exercisers (i.e., normally sedentary individuals). Twelve subjects with PD (n = 6 per cohort) were assessed in terms of PET and fMRI neuroimaging outcomes, as well as clinical measurements. Overall, this thesis combines an extensive literature review with original investigations to provide novel insight into how regular exercise may affect the motor and non-motor manifestations of PD.
Chapter 2: The effects of exercise on cognition in Parkinson’s disease: a systematic review

2.1 Summary

Cognitive impairments are highly prevalent in Parkinson’s disease (PD) and can substantially affect a patient’s quality of life. These impairments remain difficult to manage with current clinical therapies, but exercise has been identified as a possible treatment. The objective of this systematic review was to accumulate and analyze evidence for the effects of exercise on cognition in both animal models of PD and human disease. This systematic review was conducted according to the Preferred Reporting Items for Systematic reviews and Meta-Analyses (PRISMA) statement. Fourteen original reports were identified, including six pre-clinical animal studies and eight human clinical studies. These studies used various exercise interventions and evaluated many different outcome measures; therefore, only a qualitative synthesis was performed. The evidence from animal studies supports the role of exercise to improve cognition in humans through the promotion of neuronal proliferation, neuroprotection and neurogenesis. These findings warrant more research to determine what roles these neural mechanisms play in clinical populations. The reports on cognitive changes in clinical studies demonstrate that a range of exercise programs can improve cognition in humans. While each clinical study demonstrated improvements in a marker of cognition, there were limitations in each study, including non-randomized designs and risk of bias. The Grading of Recommendations Assessment, Development and Evaluation (GRADE) system was used and the quality of the evidence for human studies were rated from “low” to “moderate” and the strength of the recommendations

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were rated from “weak” to “strong.” Studies that assessed executive function, compared to
general cognitive abilities, received a higher GRADE rating. Overall, this systematic review
found that in animal models exercise results in behavioral and corresponding neurobiological
changes in the basal ganglia related to cognition. The clinical studies showed that various types
of exercise, including aerobic, resistance and dance can improve cognitive function, although the
optimal type, amount, mechanisms, and duration of exercise are unclear. With growing support
for exercise to improve not only motor symptoms, but also cognitive impairments in PD, health
care providers and policy makers should recommend exercise as part of routine management and
neurorehabilitation for this disorder.

2.2 Introduction
Aside from well-documented motor symptoms, most Parkinson’s disease (PD) patients suffer
from associated non-motor complications, including cognitive impairment, mood disorders,
olfactory dysfunction, sleep disturbance, fatigue and anxiety (Aarsland et al., 1999; Macht et al.,
2005; Chaudhuri et al., 2006). Of the non-motor symptoms, cognitive impairments are
particularly prevalent in PD with up to 83% of patients developing dementia after 20 years (Hely
et al., 2008). The non-motor symptoms of PD can be at least as detrimental as motor
manifestations for a patient’s health and overall quality of life, but unfortunately remain difficult
to manage with current clinical therapies (Aarsland et al., 1999; Macht et al., 2005; Chaudhuri et
al., 2006).

The current gold-standard for testing global cognitive capacity in clinical practice includes
objective verbal and written tests. Of the quick screening cognitive tests available, the Montreal
Cognitive Assessment (MoCA) (Nasreddine et al., 2005) has been widely accepted for use in PD populations (Zadikoff et al., 2008b) by assessing multiple domains of cognitive function including memory, language, complex visuospatial processing, and executive function. This validated tool has been helpful to measure the impact of treatments on cognition. Animal models of PD provide a more readily controlled means to assess cellular dysfunction, neurochemical alterations and other neural mechanisms that may contribute to disease pathogenesis in humans.

Exercise is thought to improve overall well-being in older adults and benefit cognitive functions of those with neurodegenerative diseases (Muller and Kuhn, 2009). It has been suggested that exercise may improve the motor manifestations of PD and that restricted use in rats may potentiate neurodegeneration (Tillerson et al., 2003). Specifically, evidence has shown that exercise is beneficial for bradykinesia, postural balance and quality of life in patients with PD (Ridgel et al., 2009a; Allen et al., 2011b; Ridgel et al., 2012b; Lauhoff et al., 2013). The extent to which exercise specifically impacts cognition in PD, and how, is unclear. A non-systematic review from 2011 suggested that vigorous exercise may have a neuroprotective effect in PD (Ahlskog, 2011). A more recent systematic review similarly showed that non-pharmacological interventions improve cognition in PD. However, this review included only those studies published before December 2011 and used limited search terms related to cognition, resulting in the review of only four clinical studies (Hindle et al., 2013). A subsequent analysis of the literature was needed to include recent clinical studies and to incorporate animal-based research that might help identify potential mechanisms in humans. Therefore, this systematic review was conducted to evaluate all original research reports that assessed exercise interventions in human PD or in animal models of PD, with a primary or secondary outcome to examine cognitive
function. To provide the most comprehensive overview of the literature, non-randomized, pre-post and cohort trials were included in addition to randomized controlled trials. The combination of these findings should be used to further guide clinical practice and neurorehabilitation exercise programs toward treating cognitive deficits in PD.

2.3 Methods

2.3.1 Systematic review protocol

This systematic review was conducted according to the Preferred Reporting Items for Systematic reviews and Meta-Analyses (PRISMA) statement guidelines (Moher et al., 2009). The PRISMA statement includes a 27-item checklist and standardized instructions for conducting a systematic review (Appendix A). The complete search methodology, information sources and results for this review are described within this report and Appendix B.

2.3.2 Eligibility criteria for study characteristics

The participants included healthy human subjects, subjects with PD and animals with experimental PD. Studies were included if their primary intervention was exercise and their primary or secondary outcomes were to assess either behavioral or neurobiological markers of cognitive function. All articles were included where the authors treated the outcome measure as a test of cognition. In some cases, the measure was a surrogate (e.g., biomarker associated with cognitive function), was a sub-score of cognition from a larger scale, or was influenced by motor capacity. An exercise intervention was defined as any purposeful increase in the subject’s physical activity through a single bout of exercise or prolonged exercise over the course of a structured or unstructured program. Cohort and experimental study designs were included,
whereas case series, case–control, cross-sectional and descriptive studies were excluded. Original research articles were included from 1966 through October 2013. Studies were considered if they were written in the English language and either published or “in press.”

2.4 Results

2.4.1 Study selection and synthesis

There were 14 records included in this analysis (Figure 2.1). Thirteen records were found through searching databases and one record (Fisher et al., 2004) was found through searching the references of articles identified for inclusion in the analysis. The records comprised six pre-clinical animal studies (all on rodents) and eight clinical studies in humans. Of the six pre-clinical studies, all were randomized controlled studies. Two studies examined the effects of exercise on unspecified aspects of cognition (Gorton et al., 2010; Tajiri et al., 2010), and four studies examined the effects of exercise specifically on learning and memory (Fisher et al., 2004; Aguiar et al., 2009; Pothakos et al., 2009; Goes et al., 2014). Of the eight clinical studies, four studies examined the effects of exercise on unspecified aspects of cognition (Baatile et al., 2000; Muller and Muhlack, 2010; dos Santos Mendes et al., 2012; Pompeu et al., 2012), and four studies examined the effects of exercise specifically on tasks of executive function (Tanaka et al., 2009b; Cruise et al., 2011a; Ridgel et al., 2011; McKee and Hackney, 2013). The clinical studies included five randomized controlled trials, one controlled trial and two pre-post trials. A quantitative comparison or meta-analysis could not be performed for either the pre-clinical or clinical studies because there were only a small number of reports identified, and when compiled together they had heterogeneous patient populations, exercise interventions and outcome measures.
2.4.2 Study characteristics and results for pre-clinical studies

All six pre-clinical studies were randomized controlled trials. Three different toxins were used to generate basal ganglia lesions and develop models of PD in rodents. Three studies used 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) in mice (Fisher et al., 2004; Pothakos et al., 2009; Gorton et al., 2010), two studies used 6-hydroxydopamine (6-OHDA) in rats (Tajiri et al., 2009;
2010; Goes et al., 2013), and one study used reserpine in rats (Aguiar et al., 2009), resulting in reversible monoamine depletion. The timing of toxin administration varied relative to the onset of the exercise program. Four studies tested the effect of exercise that started following the lesion (Fisher et al., 2004; Gorton et al., 2010; Tajiri et al., 2010; Goes et al., 2013). The interventions were five days per week for 30 days starting within a week after administration of either MPTP or 6-OHDA. One study tested the effects of exercise for one week before, five weeks during, and for 8–12 weeks following chronic MPTP administration (Pothakos et al., 2009). The study that used reserpine tested exercise five days per week for 30 days prior to administration of the toxin in order to assess how exercise may prevent cognitive impairment in PD (Aguiar et al., 2009).

Four studies looked at forced exercise on a treadmill compared to no exercise (Fisher et al., 2004; Pothakos et al., 2009; Tajiri et al., 2010; Goes et al., 2013). Three studies showed potential neurobiological correlates of observed behavioral changes that the authors related to cognition, including findings from one study that rats forced to exercise had better behavioral recovery on tests of motor function (i.e., cylinder and amphetamine-induced rotational tests), and better preservation of tyrosine hydroxylase immunoreactivity in both striatum and substantia nigra (Tajiri et al., 2010). The authors also found that exercise increased the migration of BrdU and doublecortin-positive cells as well as increased BDNF and GDNF in the striatum on the side of the lesion. In another study, exercise resulted in enhanced duration and velocity of running behavior, indicating that rats had learned to maintain a forward position on a treadmill (Fisher et al., 2004). Exercise in these MPTP- and saline-injected mice resulted in significant down-regulation of striatal dopamine transporter protein (DAT) as well as increased D2 (but not D1) receptor mRNA expression. Exercise attenuated the increase in striatal glutamate nerve terminal
labeling following MPTP, but there was no change in glutamate immunolabeling in CA1 in the hippocampus. In both of these studies exercise improved behavioral markers that were interpreted by the authors as indicative of enhanced cognitive function. It should be noted that these measures rely heavily on motor capacity and are typically associated more with motor than with cognitive function. The third study that showed neurobiological changes associated with corresponding behavioral changes tested effects of aerobic swimming in mice. The rodents showed improved long-term memory on a test of object recognition following exercise and had attenuation of impairments from exposure to 6-OHDA, including decreased pro-inflammatory cytokines, improved markers of oxidative stress and increased DA transmission (Goes et al., 2013). The last study that looked at forced exercise on a treadmill compared to no exercise conducted in a chronic model of PD found that endurance exercise improved only motor function related to gait ambulation and balance, with no improvement in cognitive measures (Pothakos et al., 2009). This study was also the only one of these four where exercise did not improve a neurobiological outcome following toxin administration, including no raise in striatal DA and no reversed loss of tyrosine-hydroxylase fibers in the substantia nigra pars compacta.

Two studies (one with MPTP and one with reserpine) looked at the effects of voluntary exercise (wheel running), compared to forced exercise on a treadmill or no exercise (Aguiar et al., 2009; Gorton et al., 2010). In one study, exercise was introduced prior to the administration of reserpine (Aguiar et al., 2009), while in the other (Gorton et al., 2010), exercise was not initiated until after the MPTP lesion. Both studies found that either form of exercise improved behavior underlying cognitive capacity in a PD-like model. Interestingly, only forced exercise, following the lesion, improved a test of motor learning (transfer of treadmill performance to Rotarod). The
authors suggested this finding reflects learning as the animals had presumably transferred skill from the treadmill to the Rotarod task (Gorton et al., 2010). The rodents forced to exercise on the treadmill showed a greater improvement on the Rotarod test, even though rodents on the wheel willingly spent more time exercising than those forced to run on the treadmill. Both voluntary and forced exercise had anxiolytic effects as assessed using the elevated plus maze, which the authors linked to cognition and memory, but neither type of exercise had any effect on depressive behavior as assessed by sucrose preference and tail suspension. These improvements were not associated with changes in the striatal DA or amygdalar serotonin (5HT) levels following the exercise intervention, as compared to saline-treated sedentary controls (Gorton et al., 2010). However, both MPTP- and saline-treated mice had a similar relative increase in striatal DA following forced or voluntary exercise compared to saline-treated sedentary controls. Forced exercise also increased 5HT in the nucleus accumbens in the MPTP-treated mice compared to saline controls. In the second study, when either forced or voluntary exercise was introduced prior to reserpine administration, both exercise paradigms resulted in improved motor learning on the Rotarod and open-field tasks (tests of exploratory activity), as well as improved social memory (Aguiar et al., 2009). Social memory improved with a low dose or reserpine which, unlike the high dose of the toxin, did not affect the animals’ motor function. Biomarkers were not assessed in this study.

Further details of study characteristics and results for pre-clinical studies are summarized in Table 2.1 Table 2.2.
<table>
<thead>
<tr>
<th>Study</th>
<th>Study title</th>
<th>Subjects</th>
<th>Intervention</th>
</tr>
</thead>
<tbody>
<tr>
<td>Goes et al., 2013</td>
<td>Neuroprotective effects of swimming training in a mouse model of Parkinson's disease induced by 6-hydroxydopamine</td>
<td>• 2 groups (n = 20 each): 6-OHDA, saline&lt;br&gt;• 2 treatment cohorts (n = 10 each): swimming training, no exercise</td>
<td>• 20–60 min/day, 5 days/week for 4 weeks&lt;br&gt;• Starting 4 days after toxin administration</td>
</tr>
<tr>
<td>Gorton et al., 2010</td>
<td>Exercise effects on motor and affective behavior and catecholamine neurochemistry in the MPTP-lesioned mouse</td>
<td>• 2 groups (n = 24 each): MPTP, saline&lt;br&gt;• 3 treatment cohorts (n = 8/group): forced exercise, voluntary exercise, no exercise</td>
<td>• Up to 1 hr/day, 5 days/week for 4 weeks&lt;br&gt;• Starting 5 days after toxin administration</td>
</tr>
<tr>
<td>Tajiri et al., 2010</td>
<td>Exercise exerts neuroprotective effects on Parkinson's disease model of rats</td>
<td>• 1 group (n = 60): 6-OHDA&lt;br&gt;• 2 treatment cohorts (n = 30 each): forced exercise, no exercise</td>
<td>• 30 min/day, 5 days/week for 4 weeks&lt;br&gt;• Starting 1 day after toxin administration</td>
</tr>
<tr>
<td>Aguiar et al., 2009</td>
<td>Physical exercise improves motor and short-term social memory deficits in reserpinized rats</td>
<td>• 4 groups (n = 24 each): high/low dose reserpine or high/low dose saline&lt;br&gt;• 3 treatment cohorts± (n = 8/group): forced exercise, voluntary exercise, no exercise</td>
<td>• 20–25 min/day, 5 days/week for 4 weeks&lt;br&gt;• Starting 4 weeks before toxin administration</td>
</tr>
<tr>
<td>Pothakos et al., 2009</td>
<td>Restorative effect of endurance exercise on behavioral deficits in the chronic mouse model of Parkinson's disease with severe neurodegeneration</td>
<td>• 2 groups (n = 29 each): probenecid/MPTP (model of chronic PD), probenecid only&lt;br&gt;• 2 treatment cohorts (n = 5-10/group): forced endurance exercise, no exercise – for probenecid/MPTP group only</td>
<td>• 40 min/day, 5 days/week for 8–12 weeks&lt;br&gt;• Starting 1 week before, 5 weeks during, 8–12 weeks after toxin administration</td>
</tr>
<tr>
<td>Fisher et al., 2004</td>
<td>Exercise-induced behavioral recovery and neuroplasticity in the 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine-lesioned mouse basal ganglia</td>
<td>• 2 groups (n = 60 each): MPTP, saline&lt;br&gt;• 2 treatment cohorts (n = 20/group): forced exercise, no exercise*</td>
<td>• Up to 2x 30 min/day, 5 days/week for 4 weeks&lt;br&gt;• Starting 4 days after toxin administration</td>
</tr>
</tbody>
</table>

± Only rats able to maintain a forward position on the treadmill were assigned to the treadmill exercise group.
* Only rats able to maintain a forward position on the treadmill were randomized to either exercise or no exercise cohort.
Table 2.2 Outcomes and risk of bias for pre-clinical studies on rodent models of Parkinson’s disease.

<table>
<thead>
<tr>
<th>Study</th>
<th>Behavioral outcomes</th>
<th>Neurobiological outcomes</th>
<th>Major sources of risk of bias</th>
</tr>
</thead>
<tbody>
<tr>
<td>Goes et al., 2013</td>
<td><em>Forced exercise following onset of experimental PD:</em></td>
<td><em>Changes in the striatum from forced exercise following onset of experimental PD:</em></td>
<td>Performance bias: sedentary control animals were not exposed to the swimming training program, the warm water or handled to be dried off following each session.</td>
</tr>
<tr>
<td></td>
<td>• Decreased marker of depression (tail suspension)</td>
<td>• Decreased interleukin 1-beta levels (proinflammatory cytokines)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>• Improved motor coordination (decreased falls on Rotarod test)</td>
<td>• Attenuated inhibition of glutathione peroxidase activity, decreased glutathione reductase and glutathione S-transferase activity (all markers of oxidative stress)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>• Improved long-term memory, but not short-term memory in object recognition test</td>
<td>• Increased dopamine, homovanillic acid, and 3,4-dihydroxyphenylacetic acid levels</td>
<td></td>
</tr>
<tr>
<td>Gorton et al., 2010</td>
<td><em>Forced and voluntary exercise following onset of experimental PD:</em></td>
<td><em>Forced and voluntary exercise following onset of experimental PD:</em></td>
<td>Performance bias: each animal was only evaluated on one test.</td>
</tr>
<tr>
<td></td>
<td>• Improved motor learning (Rotarod)</td>
<td>• Had no effect on levels of DA in the striatum and serotonin in the amygdala compared to saline controls</td>
<td></td>
</tr>
<tr>
<td></td>
<td>• Reduced anxiety in elevated plus maze (passive avoidance task, authors linked to cognition/memory)</td>
<td>• Forced and voluntary exercise increased DA in the striatum to similar levels following MPTP or saline administration</td>
<td></td>
</tr>
<tr>
<td></td>
<td>• Had no effect on markers of depression, sucrose preference and tail suspension (MPTP lesion also had no effect)</td>
<td>• Forced exercise increased 5HT in the nucleus accumbens in MPTP-treated mice compared to saline controls</td>
<td></td>
</tr>
<tr>
<td>Tajiri et al., 2010</td>
<td><em>Exercise following onset of experimental PD:</em></td>
<td><em>Exercise following onset of experimental PD:</em></td>
<td>Information bias: exercise was started soon (24 hrs) after toxin administration, so the lesion may not represent a complete PD-like model.</td>
</tr>
<tr>
<td></td>
<td>• Improved cylinder test, amphetamine-induced rotational test (authors linked to cognitive-related behavior)</td>
<td>• Preserved nigrostriatal dopamine neurons (increased tyrosine hydroxylase-positive fibers)</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>• Increased migration of new-born neural stem/progenitor cells toward striatum</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>• Up-regulated neurotrophic factors, BDNF and GDNF, in the striatum</td>
<td></td>
</tr>
<tr>
<td>Study</td>
<td>Behavioral outcomes</td>
<td>Neurobiological outcomes</td>
<td>Major sources of risk of bias</td>
</tr>
<tr>
<td>---------------------------</td>
<td>-------------------------------------------------------------------------------------</td>
<td>----------------------------------------------------------------</td>
<td>---------------------------------------------------------------------------------------------</td>
</tr>
<tr>
<td>Aguiar et al., 2009</td>
<td><em>Forced and voluntary exercise before onset of experimental PD:</em></td>
<td>Neurobiological outcomes not assessed</td>
<td>Information bias: behavioral testing was soon (24 hrs) after the reserpine administration, so the lesion may not represent a complete PD-like model.</td>
</tr>
<tr>
<td></td>
<td>• Improved motor deficits following a high dose of reserpine</td>
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<tr>
<td></td>
<td>• Improved short-term social memory (tested through olfactory discrimination), with no deficit on motor or olfactory function from the low dose of reserpine</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pothakos et al., 2009</td>
<td><em>Endurance exercise before and following onset of experimental chronic PD:</em></td>
<td><em>Endurance exercise before and after onset of experimental chronic PD:</em></td>
<td>Selection biases: there was not a group that received exercise and probenecid. There was also not a control group with only a saline injection. The effects of the control solution, probenecid, on cognition are not known.</td>
</tr>
<tr>
<td></td>
<td>• Reversed balance and gait performance, restored regular movement</td>
<td>• Did not raise striatal DA (n = 6)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>• Had no effect on learning (cued Morris water maze), amphetamine-stimulated locomotion or motor coordination</td>
<td>• Did not reverse loss of tyrosine-hydroxylase fibers in substantia nigra (pars compacta)</td>
<td></td>
</tr>
<tr>
<td>Fisher et al., 2004</td>
<td><em>Exercise following onset of experimental PD:</em></td>
<td><em>Exercise following onset of experimental PD:</em></td>
<td>Information bias: the learning paradigm for behavioral results (learning to stay on the treadmill) relied substantially on motor capacity.</td>
</tr>
<tr>
<td></td>
<td>• Improved velocity and endurance on treadmill</td>
<td>• Had no effect on tyrosine hydroxylase</td>
<td></td>
</tr>
<tr>
<td></td>
<td>• Sensory feedback not needed over time for behavioral response</td>
<td>• Up-regulated dopamine D2 receptor mRNA expression</td>
<td></td>
</tr>
<tr>
<td></td>
<td>(i.e., maintaining a forward position on treadmill), authors suggested indicative of learning</td>
<td>• Down-regulated striatal DAT</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>• Reversed increased nerve terminal glutamate in striatum (as a result of MPTP)</td>
<td></td>
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</tbody>
</table>
2.4.3 Major sources of risk of bias for pre-clinical studies

One of the six pre-clinical animal studies was at risk of selection bias because the investigators did not include a saline-only control group or an exercise and probenecid group, although it was still a randomized controlled trial (Pothakos et al., 2009). Three other pre-clinical studies were at risk of information biases; in one case the cognitive assessment relied on motor capacity (Fisher et al., 2004), in another case the behavioral testing began soon after administration of the toxin, which may have interrupted the lesion process (not measured) and made the model less comparable to PD in human subjects (Aguiar et al., 2009) and in the third case the exercise was started soon after the toxin was administered, which may also have interrupted the lesion process (Fisher et al., 2004). Two studies had performance biases, specifically that each animal was only evaluated on one test (Gorton et al., 2010) and that exercised mice were trained for swimming for two weeks before toxin administration and handled each session, whereas the sedentary animals were not trained or handled (Goes et al., 2013). Additionally, the social interaction (i.e., housing environment) the animals experienced was different in each of the six studies, making it difficult to compare results across studies. The number of rodents housed in a cage ranged from one to ten, and one study did not specify the housing environment. Paired housing compared to single housing in rodents has been shown to mediate the effects of MPTP on nigrostriatal degeneration and motor behavior (Goldberg et al., 2012) and may have a substantial effect on behavioral and histological outcomes. The risk of bias for each pre-clinical study is summarized in Table 2.2.
2.4.4 Study characteristics and results for clinical studies

All eight studies showed that exercise improved a marker of cognition (Table 2.3). Based on the GRADE ranking system, the quality of the evidence and strength of recommendations for the four studies on executive function were greater (“moderate” and “strong”) than for studies testing unspecified aspects of cognitive function (“low” and “weak”).

<table>
<thead>
<tr>
<th>Study</th>
<th>Can exercise improve a marker of cognitive function?</th>
<th>Quality of evidence</th>
<th>Strength of recommendation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Studies that specifically measured executive function (n = 4)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>McKee et al., 2013</td>
<td>Yes</td>
<td>Moderate</td>
<td>Strong</td>
</tr>
<tr>
<td>Cruise et al., 2011</td>
<td>Yes</td>
<td>Moderate</td>
<td>Strong</td>
</tr>
<tr>
<td>Ridgel et al., 2011</td>
<td>Yes</td>
<td>Moderate</td>
<td>Strong</td>
</tr>
<tr>
<td>Tanaka et al., 2009</td>
<td>Yes</td>
<td>Moderate</td>
<td>Strong</td>
</tr>
<tr>
<td>Studies that measured unspecified aspects of cognition (n = 4)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Dos Santos Mendes et al., 2012</td>
<td>Yes</td>
<td>Low</td>
<td>Weak</td>
</tr>
<tr>
<td>Pompeu et al., 2012</td>
<td>Yes</td>
<td>Low</td>
<td>Weak</td>
</tr>
<tr>
<td>Müller et al., 2010</td>
<td>Yes</td>
<td>Low</td>
<td>Weak</td>
</tr>
<tr>
<td>Baatile et al., 2000</td>
<td>Yes</td>
<td>Low</td>
<td>Weak</td>
</tr>
</tbody>
</table>

1. Quality of evidence and strength of recommendations based on the Grades of Recommendations, Assessment, Development, and Evaluation (GRADE) ranking system (Higgins et al., 2011a; Higgins et al., 2011b).
2. The GRADE system offers four levels for the quality of evidence: high, moderate, low and very low.
3. The GRADE system offers three levels for the strength of a recommendation: strong, weak, or no recommendation.

The sample size for the eight clinical studies varied from six to 60 subjects, with a mean of 15 subjects per group. The PD participants had a mean age between 60 to 70 years and had mild to moderate PD according to the Hoehn & Yahr (H&Y) scale (Hoehn and Yahr, 1967), and were compared to age-matched healthy control subjects. All of these studies examined subjects while they were taking their regularly prescribed medication. These studies did not report when
subjects took their regular medication and which medications they continued to take during the study.

Each of the four studies that tested the effect of exercise on unspecified aspects of cognition found benefits on general markers of overall function which the authors related to cognition, including the cognition component of PDQ-39, MoCA, memory, reaction time and peg insertion time (requiring visual and spatial cognition, sorting and planning) (Baatile et al., 2000; Muller and Muhlack, 2010; dos Santos Mendes et al., 2012; Pompeu et al., 2012). The evidence from two of the studies is limited as they were pilot trials designed to test feasibility (Baatile et al., 2000; Muller and Muhlack, 2010). Additionally, the measures of cognition these studies used, the cognition component of PDQ-39, reaction time and peg insertion, are not clear measures of cognitive function. However, the two other studies clearly demonstrated that exercise had an effect on cognitive capacity (dos Santos Mendes et al., 2012; Pompeu et al., 2012). Of these two trials, both tested the Wii Fit™ program. They showed that after training, PD subjects can retain and transfer learning, depending on the cognitive demands of the game, (dos Santos Mendes et al., 2012) but Wii Fit™ may not provide additional advantage in comparison to balance exercises without cognitive stimulation or feedback (Pompeu et al., 2012).

Across studies, exercise interventions varied significantly in terms of the intensity, mode and duration of the program. One study involved an individualized walking program (PoleStriding) using Nordic poles three times per week for eight weeks (Baatile et al., 2000). Another study assessed outcomes before and after a single bout of high-intensity cycling (Muller and Muhlack, 2010). The two studies that used the Wii Fit™ program for their exercise intervention included
two sessions per week for seven weeks and follow-up after 60 days (dos Santos Mendes et al., 2012; Pompeu et al., 2012). One of the two studies using Wii Fit™ compared PD subjects to healthy controls (dos Santos Mendes et al., 2012) and the other study looked at differences between PD subjects participating in the combined Wii Fit™ program with balance-based and cognitive training compared to multimodal global exercises (Pompeu et al., 2012).

Each of the four studies that specifically tested executive function showed that exercise improved performance on some measure of executive function, such as tests of abstraction, mental flexibility, spatial working memory, verbal fluency, mental imagery, and cognitive processing speed (Tanaka et al., 2009b; Cruise et al., 2011a; Ridgel et al., 2011; McKee and Hackney, 2013). The tools used to test executive function include the Wisconsin Card Sorting Task (WCST), Trail-Making Test A and B (TMT A & B), Cambridge Neuropsychological Test Automated Battery, and tests of verbal and semantic fluency. Two studies assessed mood as a potential confounding factor affecting executive function; one study found that exercise improved executive function independent of improvements in mood, attention, disease-specific quality of life or reduced anxiety (Tanaka et al., 2009b). Another study found that exercise possibly improved mood, but did not affect quality of life (Cruise et al., 2011a). This lack of impact on quality of life is interesting given that this study did not control for the benefits of social interaction received by the exercise group in comparison to the control group, who were instructed to continue with their normal routine. Both low-intensity passive aerobic cycling (Ridgel et al., 2011) as well as moderate-intensity aerobic and anabolic exercise (Tanaka et al., 2009b; Cruise et al., 2011a) were found to improve executive function in PD. The fourth study differed from the other exercise interventions because it assessed 20 sessions of tango classes
compared to education over 12 weeks. The subjects were assessed for cognitive function 10–12 weeks following the intervention. Subjects in the tango arm improved on the Brooks Spatial Task, a test of spatial cognition (i.e., mental imagery) (McKee and Hackney, 2013). The authors interpreted this as an improvement in executive function.

There was substantial variety in the intensity, mode and duration of the exercise interventions in these four studies. One study included an exercise intervention involving low-intensity passive cycling once per week for four weeks. Another study included moderate-to-high intensity anabolic and aerobic exercise 60 minutes per session twice per week for 12 weeks. The intensity and the work load of the sessions were increased over time. Each session involved a short low-intensity aerobic warm-up, six resistance exercises for both upper and lower body muscle groups, and then 25–30 minutes of aerobic exercise on a stationary bicycle, rowing machine or treadmill. A third study included moderate-intensity multimodal exercise training involving aerobic exercise with the addition of resistance, coordination and balance training 60 minutes per session, three times per week for 24 weeks. The 24-week intervention was divided into six phases and the load was increased after each phase. A session included five components: warm-up, stretching before exercise, exercise, cool-down, and stretching after exercise. The fourth study on tango implemented a standardized structured tango program for 90-minute sessions twice per week for 12 weeks.

Further details of study characteristics and results for clinical studies are summarized in Table 2.4 Table 2.5.
Table 2.4 Study characteristics of clinical trials on human Parkinson’s disease.

<table>
<thead>
<tr>
<th>Study</th>
<th>Study title</th>
<th>Subjects</th>
<th>Intervention</th>
<th>Study design</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Studies that specifically measured executive function (n = 4)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
| McKee et al., 2013 | The Effects of Adapted Tango on Spatial Cognition and Disease Severity in Parkinson’s Disease | Total n = 33 PD  
• n = 15 tango  
• n = 13 control | • Tango or education lessons  
• Sessions 90 minutes long, 20 sessions over 12 weeks, follow-up after 10–12 weeks | Randomized controlled trial |
| Cruise et al., 2011 | Exercise and Parkinson's: benefits for cognition and quality of life | Total n = 28 PD  
• n = 15 exercise  
• n = 13 control | • Moderate-to-high-intensity anabolic and aerobic exercise or usual care  
• Sessions 1 hr/day, 2x/week for 12 weeks | Single-blind randomized controlled trial |
| Ridgel et al., 2011 | Changes in executive function after acute bouts of passive cycling in Parkinson's disease | Total n = 19 PD  
• n = 10 exercise  
• n = 10 control | • Low-intensity passive aerobic exercise (cycling)  
• Sessions 1/week for 4 weeks | Randomized controlled trial, crossover |
| Tanaka et al., 2009 | Benefits of physical exercise on executive functions in older people with Parkinson's disease | Total n = 20 PD  
• n = 10 exercise  
• n = 10 control | • Moderate-intensity multimodal exercise training (aerobic, resistance, coordination and balance) or usual care  
• Sessions 1 hr/day, 3x/week for 24 weeks, intensity increased every 4 weeks | Controlled trial* |

| **Studies that measured unspecified aspects of cognition (n = 4)**                                                                 |
| Dos Santos Mendes et al., 2012 | Motor learning, retention and transfer after virtual-reality-based training in Parkinson's disease - effect of motor and cognitive demands of games: a longitudinal, controlled clinical study | Total n = 27 PD  
• n = 16 PD  
• n = 11 healthy control | • Low-intensity Wii Fit™ training, involving motor shifts and cognitive skills  
• Sessions 1 hr/day, 2x/week for 7 weeks, follow-up at 60 days | Longitudinal pre-post trial |
Subjects were assigned into the training group based on previous participation as a control in another study and upon referral by their physician. Baseline characteristics did not differ between the groups.

<table>
<thead>
<tr>
<th>Study</th>
<th>Study title</th>
<th>Subjects</th>
<th>Intervention</th>
<th>Study design</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pompeu et al., 2012</td>
<td>Effect of Nintendo Wii\textsuperscript{TM}-based motor and cognitive training on activities of daily living in patients with Parkinson’s disease: A randomised clinical trial exercise no Wii</td>
<td>Total n = 32 PD • n = 16 exercise &amp; Wii • n = 16 control</td>
<td>•Both groups: low-intensity stretching, strengthening •Experimental group: Wii Fit\textsuperscript{TM}-based motor/cognitive training Control group: balance exercises without feedback or cognitive stimulation •Sessions 1 hr/day, 2x/wk for 7 weeks, follow-up at 60 days</td>
<td>Single-blind randomized controlled trial</td>
</tr>
<tr>
<td>Müller et al., 2010</td>
<td>Effect of exercise on reactivity and motor behaviour in patients with Parkinson’s disease</td>
<td>Total n = 22 PD</td>
<td>•Single bout of high-intensity endurance aerobic exercise (heart rate-targeted cycling) or rest following L-dopa administration •Randomized order 1 day apart</td>
<td>Randomized controlled feasibility trial, cross-over</td>
</tr>
<tr>
<td>Baatile et al., 2000</td>
<td>Effect of exercise on perceived quality of life of individuals with Parkinson’s disease</td>
<td>Total n = 6 PD</td>
<td>•Low-intensity aerobic exercise program with Nordic walking poles (PoleStriding) •Sessions 3x/week for 8 weeks</td>
<td>Nonrandomized feasibility trial, no control</td>
</tr>
</tbody>
</table>

*Subjects were assigned into the training group based on previous participation as a control in another study and upon referral by their physician. Baseline characteristics did not differ between the groups.

Table 2.5 Outcomes and risk of bias of clinical trials on human Parkinson’s disease.

<table>
<thead>
<tr>
<th>Study</th>
<th>Behavioral outcomes</th>
<th>Major sources of risk of bias</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Studies that specifically measured executive function (n = 4)</em></td>
<td></td>
<td></td>
</tr>
<tr>
<td>McKee et al., 2013</td>
<td>Tango improved disease severity (UPDRS-III) and spatial cognition/mental imagery (Brooks Spatial Task) more than education group, maintained gains 10–12 weeks post-intervention</td>
<td>Detection bias: study was underpowered (n = 23 tango, n = 8 education) to evaluate some main effects within groups, so main effect of time was evaluated</td>
</tr>
<tr>
<td>Cruise et al. 2011</td>
<td>Exercise improved verbal fluency and spatial working memory on Cambridge Neuropsychological Test Automated Battery •Exercise was of “possible benefit” on semantic fluency and mood •Exercise did not benefit spatial or pattern recognition, quality of life, had no negative impact</td>
<td>Selection bias: the control group received usual care, no control for the effect of social interaction with exercise •Information bias: the variable intensity level of the intervention could have affected outcomes</td>
</tr>
<tr>
<td>Study</td>
<td>Behavioral outcomes</td>
<td>Major sources of risk of bias</td>
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<td>------------------------------</td>
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| Ridgel et al., 2011          | • Time to complete Trail-Making Test A & B (tests executive function) decreased after passive cycling  
  • Performance improved on Trail-Making Test B following passive cycling | • Selection bias: no control  
• Information bias: the same test pattern was used pre- and post-intervention, although practice effects were attempted to be controlled through pre-test training with the task |
| Tanaka et al., 2009          | • Exercise improved executive function for “Categories Completed” (i.e., capacity for abstraction) and “Preservative Errors” (i.e., mental flexibility) on the Wisconsin Card Sorting Task  
  • No interactions for confounding variables: concentrated attention, trait or state anxiety, depression | • Selection bias: small sample size, no long-term follow-up, not purely randomized  
• Information bias: no mention of medication administration; only one participant in the group had a heart rate monitor, so the intensity was targeted towards the group and not the individual |
| Dos Santos Mendes et al., 2012 | • PD showed no deficit in learning or retention on 7/10 games, deficits related to cognitive demands of tasks  
  • PD had worse performance than healthy individuals on 5 tests  
  • PD could transfer learning to an untrained motor task at follow-up | • Selection bias: the baseline physical fitness of the subjects was not compared  
• Performance bias: no PD controls not performing intervention, no control for enjoyment or motivation |
| Pompeu et al., 2012          | • Both groups improved UPDRS-II, MoCA and balance, no additional advantage from Wii Fit™ games  
  • Improved scores on Wii Fit™ games, maintained at follow-up  
  • No differences in outcomes between groups pre-to post-intervention or in follow-up | • Information bias: the baseline physical fitness of the subjects was not compared, so potential for differences between groups |
| Müller et al., 2010          | • Reaction time increased after rest and decreased after exercise, movement time decreased after exercise  
  • Number of correct answers decreased after rest  
  • Tapping rate increased after exercise  
  • Peg insertion interval time decreased after exercise (complex movement sequences, visual and spatial cognition, sorting and planning) | • Selection bias: no PD control group, no healthy controls  
• Information bias: one-day washout period (24 hours) may not have been long enough; pilot trial  
• Detection bias: unclear how reactivity was measured |
| Baatile et al., 2000         | • Improved UPDRS score (only total score significant)  
  • Improved PDQ-39 score, most improved in cognition component | • Selection bias: limited sample size, no control group; pilot trial  
• Information bias: exercise intensity not standardized |

*Studies that measured unspecified aspects of cognition (n = 4)*
2.4.5 Major sources of risk of bias for clinical studies

Most of the studies had a selection bias from not adequately standardizing control subjects. More specifically, the subjects’ physical fitness levels and their concomitant medications before and during the study period were not documented and may have impacted their ability to exercise and the ability to compare their potential benefits on cognition. Additionally, information bias resulted from variability with the timing and intensity of the exercise intervention between subjects. Individuals may have received different amounts and types of exercise which could have affected the impact of exercise on cognition.

Of the five randomized controlled clinical trials, four studies included control groups that received the same social interaction as the exercise intervention group. One study (Cruise et al., 2011a) included a control group receiving usual care, which did not control for the potential benefits to the subjects’ mood and cognition from increased social interaction through participation in the exercise intervention. Overall, both the pre-clinical and clinical studies showed a trend of selective reporting for only significant outcomes. A risk of bias across studies includes a publication bias, as studies with insignificant or negative findings are less likely to be published. The risk of bias for each clinical study is summarized in Table 2.5.

2.5 Discussion

2.5.1 Pre-clinical evidence

The rodent studies identified in this review have suggested potential mechanisms for the benefits of exercise on cognitive improvements in PD, particularly related to learning and memory. The mechanisms include:
1) enhanced availability of DA in projections to the dorsal and/or ventral striatum;
2) enhanced expression of neurotrophic factors BDNF and GDNF, which could promote plasticity for learning and memory; and
3) decreased oxidative stress and/or neuroinflammation in the basal ganglia.

Two of the five studies that assessed biomarkers did not find effects from exercise following toxin administration. However, in one case forced and voluntary exercise similarly elevated striatal DA in both MPTP- and saline-treated groups, although there was no difference compared to sedentary saline-treated controls (Gorton et al., 2010). The other study used a model of chronic PD where the toxin was administered with probenecid over five weeks; while exercise restored regular motor function, it had no effect on learning (Pothakos et al., 2009). DA levels following the chronic toxin administration were low and potentially more difficult to increase with exercise due to the substantial neurodegeneration.

The obvious caveat to all studies using rodents to assess cognition is that the tasks of cognition, learning and memory require motor function to evaluate their performance. The models used in these studies (i.e., MPTP, 6-OHDA and reserpine) are widely accepted, although none of them recapitulates the indolent and progressive nature of PD (Colpaert, 1987; Gerlach and Riederer, 1996; Schwarting and Huston, 1996b, a, 1997; Jakowec and Petzinger, 2004). It is unknown how the time of onset or intensity of the exercise paradigm initiated during or following the lesion/toxin may affect the severity of parkinsonism. More promising models for the future would include transgenic or knock-in rodents characterized by expression of mutant or overexpression of wild-type α-synuclein. A related model is the injection of synthetic α-
synuclein fibrils into the rodent brain. This model shows progressive and selective loss of DA neurons in the substantia nigra pars compacta, as well as cell-to-cell transmission in anatomically connected regions (Luk et al., 2012). As cortical Lewy body pathology is a major contributing factor to dementia in PD (Horvath et al., 2013), future research with models of abnormal α-synuclein deposition may translate better into clinical research and practice than models that create basal ganglia lesions.

The potential for exercise to improve cognition by reducing the impact of neuroinflammation is promising. Another study in a rodent model not dependent upon a selective dopaminergic neurotoxin found that forced treadmill and voluntary wheel exercise in rats can also alleviate impairments from brain inflammation on long-term potentiation and spatial learning (Kim et al., 2013). The authors injected rats with lipopolysaccharides into the cerebral ventricles to induce brain inflammation. They found that both treadmill and wheel training improved the resulting deterioration in spatial learning. The effects of exercise on learning were attributed to enhanced expression of BDNF, tyrosine kinase B and phosphorylated cyclic AMP response element binding protein in the hippocampus. The impact of the injection on neuroinflammation was not documented. This model results in preferential but not entirely selective nigral DA cell degeneration and the findings may thus provide insight into potential mechanisms of exercise in humans to reduce brain inflammation and improve cognition.

Overall, the results from these studies on rodent models of PD offer promising support for exercise to improve cognition in humans with PD through the promotion of neuronal proliferation, neuroprotection, neurogenesis, and potentially reduction in brain inflammation.
These results support recent work that highlights how exercise likely promotes neurorestoration through activation of signaling cascades by neurotrophic factors (Zigmond et al., 2012b). Exercise has been shown to affect regulation of DA function, including upregulated DA D₂ receptor mRNA and down-regulated striatal DAT in rodents (Fisher et al., 2004). High-intensity exercise also increased DA D₂ receptor availability in a recent feasibility study by the same group using five human subjects (n = 2 PD exercise, n = 2 PD no exercise, n = 1 healthy control) and PET with [¹⁸F]fallypride (Fisher et al., 2013a). While this small human trial does not assess cognition, these findings encourage more clinical trials based on rodent outcomes in this field. Overall, the evidence from rodent studies in this systematic review cannot be directly applied to mechanisms in humans yet, but they suggest that PD patients would likely experience a meaningful improvement in cognition in response to exercise.

2.5.2 Clinical evidence

Clinical studies in humans demonstrate that various modalities and intensity levels of exercise can improve cognitive capacity in PD, and especially executive function, although the mechanisms have not yet been determined. Cognitive dysfunction in PD is commonly associated with impaired executive function (Higginson et al., 2003). However, evaluating research on cognition, and particularly executive function, in PD is challenging because MCI in PD has only recently been defined and formal diagnostic criteria are still in development (Litvan et al., 2012). Selection and interpretation of measures of executive function in PD have been challenging and the clinical implications are not yet fully appreciated (Kudlicka et al., 2011). Executive function is generally related to goal-directed behaviors processed by the frontal lobes of the brain. Executive function has been categorized into four components: planning, purposive action,
effective performance and volition (Lezak, 1995). It is not known what frequency, intensity, type or timing of exercise might be most effective to improve executive function in PD, but there is evidence in older adults without PD that light aerobic exercise (walking), and not anaerobic exercise (stretching and toning), selectively improves executive functions processed in the frontal and prefrontal areas of the brain (Kramer et al., 1999). The potential different effects of aerobic compared to anaerobic exercise on cognition in PD have not yet been studied.

Exercise in animal models of PD may induce DA release and enhance DA transmission via up-regulation of DA D_2 receptors (Petzinger et al., 2007; Vuckovic et al., 2010b). A systematic review on the effects of exercise in the elderly showed that moderate-intensity exercise can effectively increase peripheral BDNF (de Melo Coelho et al., 2013). Serum BDNF crosses the blood–brain barrier so these results may have implications for brain neurotrophin levels (Sartorius et al., 2009). One of the animal studies reviewed in this paper also found that exercise increased BDNF and GDNF in the striatum (Tajiri et al., 2010). The findings from this review support the theory of potential neuroprotective benefits of exercise for human PD. A recent review on the benefit of exercise to improve cognition emphasized the potential neuroprotective effects of vigorous exercise in PD (Ahlskog, 2011). The authors provided guidelines including vigorous exercise, structured programs for cognitively impaired patients, and therapies that replenish DA to provide the maximum capability and motivation to exercise (Ahlskog, 2011). In addition to these guidelines, the current systematic review shows that any exercise should be encouraged as it may benefit numerous aspects of patients’ cognitive function and these effects could be transferrable to other tasks. Importantly, the effects of vigorous exercise can last up to 60 days (dos Santos Mendes et al., 2012; McKee and Hackney, 2013). In these studies there was
overall a high retention rate for subjects committing to a twice or three times weekly exercise program, suggesting that these interventions could be feasibly implemented as treatment programs. A recent meta-analysis showed that very light to vigorous exercise seems to have a small effect on cognition in the acute phase following exercise, but larger longer-lasting effects are possible with more intense exercise (Chang et al., 2012).

The benefits of exercise on cognition in PD are comparable to those seen in healthy older adults. A recent review showed that endurance and resistance exercise can improve cognition in healthy seniors (Muller and Kuhn, 2009). There is less research on the effects of exercise in frail older adults, but recent evidence showed that a three-month physical activity intervention improved physical abilities, executive functions, processing speed and working memory (Langlois et al., 2013). The effects of exercise on cognition in older adults with MCI are less promising, as a recent meta-analysis showed only limited potential to improve cognition (Gates et al., 2013). However, the interpretation is constrained because many of the publications were deemed by the authors of this analysis to be of moderate quality and many of the studies were underpowered. It is possible that exercise may have an impact on dopaminergic signaling that renders it particularly valuable in PD. Whether cognition in PD is improved due to dopaminergic mechanisms of exercise or other mechanisms such as increased neurotrophic factor availability or reduced neuroinflammation remains to be determined.

2.5.3 Limitations

Limitations at the level of each study include risk of information, performance and/or selection biases (Table 2.2 and Table 2.5) as well as confounds inherent with the limitations of non-
randomized trials. There was overall a lack of reporting of concomitant medications (i.e., PD therapies and antidepressants) as well as the medical condition of the subjects, which could have affected the results. The limitations of this review include potential for incomplete retrieval of information given the search strategy and inclusion criteria. Additionally, it is not known whether there are genetic factors underlying response to exercise in PD.

2.6 Conclusions

Overall, this systematic review found that exercise can improve cognitive function in animal models and human PD. Pre-clinical studies showed exercise results in behavioral and corresponding neurobiological changes in the basal ganglia related to cognition. Specifically, learning and memory improved after exercise in the rodents, although the exact mechanisms remain unclear and merit further research. Pre-clinical studies also showed that any exercise is better than inactivity and that forced exercise has a greater impact than self-paced voluntary exercise. Exercise resulted in structural, neurochemical and molecular changes in rodents, which may be of relevance to the human disorder. The clinical studies showed that various types of exercise, including aerobic, resistance and dance can improve cognitive function, especially executive function in PD patients. However, the best type, amount, mechanisms, and duration of exercise are not yet known. The evidence from clinical studies suggests that a more intensive aerobic exercise program including strength and balance training can promote greater cognitive gains. However, low-intensity exercise and balance-based exercises also showed benefits. Research on the effects of exercise on cognition in PD is a relatively new area. As outlined in this review, there are several limitations with the current studies in terms of study design and risks of bias. Questions that remain to be addressed include the prescription of exercise, if any,
which elicits the most gains as well as the duration of effects. Future research on the effects of exercise on cognition in PD should include a longitudinal randomized controlled clinical trial examining neurobiological mechanisms *in vivo*, including neuroimaging. Understanding the mechanism of benefit from exercise could help us to harness its potential neuroprotective effects. Patients should use these findings as further rationale to increase their daily physical activity. With growing support for exercise to improve not only motor symptoms, but also cognitive impairments in PD, health care providers and policy makers should recommend exercise as part of routine management and neurorehabilitation for PD.
**Bridging statement**

This systematic review provides the appropriate background to next describe more broad effects of exercise on two cohorts of PD subjects that have not previously been investigated: habitual exercisers and non-regular exercisers. While this systematic review found that exercise can benefit cognition in PD, there are few studies on this topic and most have not investigated the mechanism of benefit from exercise. There is especially a lack of research on the mechanisms of benefit from exercise in human PD on both motor and non-motor outcomes, as well as the role of dopaminergic systems. Furthermore, many studies have been inconclusive and few studies have included exercise interventions longer than 3 months, but it is likely long-term participation elicits the best outcomes. Our study described in Chapter 3 helps to start filling the void in the literature on the effects of habitual exercise in human PD and potential mechanisms of benefit.
Chapter 3: The effects of habitual exercise on Parkinson’s disease

3.1 Introduction

Motor and non-motor symptoms of PD stem from the loss of dopaminergic projections in the basal ganglia. Benefits from exercise in PD may arise primarily from dopaminergic mechanisms, but may also include complex interplay with other neurotransmitters. As understanding the mechanism of benefit from exercise in PD is a relatively new area of research, this investigation naturally begins by studying how dopaminergic systems in the basal ganglia may be affected by exercise in PD. Anecdotal evidence indicates that habitual exercisers have better clinical outcomes than those who are generally sedentary; however, neurological differences between these two groups have not been studied previously. Most quality research on exercise in PD involves exposing individuals with PD to exercise interventions between three and six months long and thus does not capture the effects of long-term participation in exercise. Given that dopaminergic medication and their adjuncts are the most effective pharmacological therapy for PD and that exercise improves symptoms of PD, it is possible that those who exercise regularly experience increased levels of endogenous dopamine from their participation in exercise. Some evidence suggests that participation in exercise could cause dopamine release in PD (Ouchi et al., 2002; Nozaki et al., 2013), although these studies used simple foot movements performed during PET and do not capture the sustained effects of exercise. It is possible that habitual exercisers may over time have neurological changes leading to increased dopamine release in response to exercise in comparison to non-exercisers. Increased dopamine availability could then facilitate activity in the basal ganglia and improve both motor and non-motor function. However, the mechanism of dopamine release from exercise is unknown and merits further investigation.
As highlighted in previous chapters, exercise improves clinical measures of both motor and non-motor function, which indicates exercise may impact many pathways in the brain. Mesolimbic pathways originating in the mid-brain have been more associated with reward whereas nigrostriatal pathways have been more associated with motor function (Schultz, 2002; Wise, 2002). The ventral striatum is part of the mesolimbic pathway and has been demonstrated to initially process sensory, emotional and motivational stimuli through inputs from the ventromedial prefrontal cortex, orbitofrontal cortex and dorsal anterior cingulate (Haber, 2011). The ventral striatum contains dopaminergic nerve terminal responsible for mediating action and output from reward (Haber, 2011). Dopamine depletion reduces responsiveness to reward, and particularly to effort-based reward (Salamone et al., 2007). As participation in exercise requires effort, exercise could enhance dopamine release in the ventral striatum. Exercise could also increase the individual’s response to general rewards, and to exercise as a type of rewarding stimulus, which could further facilitate their continued participation in exercise. One of the studies suggesting that exercise causes dopamine release supports this theory. This study found that subthalamic nucleus DBS facilitated dopamine release in the ventromedial striatum during exercise (right foot movement) measured with RAC binding during PET (Nozaki et al., 2013). Therefore, investigating how habitual exercise affects dopaminergic activity must include studying how exercise impacts mechanisms in the ventral striatum as well as response to reward in the ventral striatum.

The overarching objective of the study presented in this chapter was to investigate how habitual exercise may benefit PD. Specifically, this project aimed to compare dopamine release in the
dorsal and ventral striatum, as well as response to reward in the ventral striatum in PD between habitual exercisers and non-regular exercisers. Supplementary measures to these neuroimaging outcomes included clinical assessments of motor and non-motor outcomes between these two cohorts. As characteristics of PD can be somewhat discretely categorized into motor and non-motor features, two hypotheses were developed accordingly.

First, it was hypothesized that all subjects would release dopamine in the dorsal striatum in response to exercise, and that habitual exercisers would have higher dopamine release than non-exercisers. Furthermore, these differences would be associated with improved motor function. The primary outcome for this hypothesis was to measure dopamine release in the dorsal striatum using PET and displacement of RAC binding. Secondary outcome measures for this hypothesis were to compare measures of motor function, including finger tapping rates, Purdue Pegboard rates and Timed Up and Go (TUG) speed.

Second, it was hypothesized that all subjects would release dopamine in the ventral striatum in response to exercise, and that habitual exercisers would have: 1) increased dopamine release in the ventral striatum, and 2) increased blood-oxygen-level-dependent (BOLD) percent signal change in the ventral striatum in response to rewarding stimuli measured using a gambling card task during fMRI. Furthermore, these differences would be associated with improved non-motor function, including measures of mood, apathy and cognition. The primary outcome for this hypothesis was to measure dopamine release in ventral striatum using PET and displacement of RAC binding. Secondary outcome measures included measuring fMRI BOLD percent signal change at 0%, 50% and 75% and 100% probability of winning during the gambling card task.
Additional secondary outcome measures included comparing mood using Beck Depression Inventory (BDI) and Positive and Negative Affect Schedule (PANAS), apathy using Starkstein Apathy Scale (Starkstein), and cognition using Wisconsin Card Sorting Task (WCST), Trail-Making Test A & B (TMT A & B) and a computerized test of simple reaction time.

### 3.2 Measuring dopamine release with PET

PET was used in this study because it allows *in vivo* measurement of dopamine release in humans. PET is a type of functional imaging that allows visualization of molecular and cellular biological processes. PET data cannot demonstrate actual biological activity, but can infer that activity. In the case of estimating dopamine release following a bout of exercise, PET can be used to measure binding potential (BP) of a radioactive ligand (i.e., radiotracer), which provides an inference of receptor occupancy.

[^11]C]Raclopride (RAC) is a PET radiotracer that has a modest affinity for the dopamine D₂ and D₃ receptors and is accordingly subject to competitive displacement by endogenous dopamine released in response to a variety of stimuli (Seeman et al., 1989; Laruelle, 2000), including levodopa (Tedroff et al., 1996; de la Fuente-Fernandez et al., 2001b), amphetamine (Piccini et al., 2003), repetitive transcranial magnetic stimulation (rTMS) (Strafella et al., 2001; Strafella et al., 2003; Strafella et al., 2005a), primary reward (Koepp et al., 1998; Small et al., 2001; Small et al., 2003) placebo (de la Fuente-Fernandez et al., 2001a; Lidstone et al., 2010) and exercise (Ouchi et al., 2002; Nozaki et al., 2013). The studies on dopamine release in exercise assessed the effects of continuous foot extension/flexion during PET scanning (Ouchi et al., 2002; Nozaki et al., 2013). In these studies, the authors evaluated dopamine release during a relatively simple
and isolated foot exercise, and in one study subjects had DBS (Nozaki et al., 2013), whereas our study looks at the acute response to a bout of high intensity aerobic exercise and how this response may differ based on an individual’s regular participation in exercise. As with previous studies, change in RAC BP before and after an intervention is used as an index of dopamine release. A decrease in RAC BP has been demonstrated to indicate dopamine release (Drevets et al., 1999; Martinez et al., 2003).

3.3 Study methodology

3.3.1 Subjects

The UBC Research Ethics Board approved this study. Informed consent was obtained for each subject prior to all study procedures. Eligible subjects were recruited from PD patients seen at the Pacific Parkinson's Research Clinic at UBC and through community outreach. Twelve subjects completed the study (n = 6 habitual exercisers, n = 6 non-regular exercisers). Subjects had to have mild to moderate (Hoehn & Yahr stage ≤ III) idiopathic PD according to United Kingdom Brain Bank criteria (modified to permit inclusion of subjects with a family history), aged 40-75. The two cohorts were matched for age and length of disease. Disease duration was measured from the date of first motor symptom onset, as documented by the subject’s neurologist.

Subjects were selected based on the level of regular exercise in their daily life for at least the previous six months and were allocated to one of the two cohorts. To quantify each subject’s level of regular exercise, a questionnaire was used to determine weekly participation in exercise and length of exercise per week (Appendix C). The scoring for this questionnaire was based on
evidence that moderate exercise (3-6 metabolic equivalents, METs) requires about twice as much energy as mild activities (1-2 METs) and vigorous exercise (>6 METs) requires about three times the energy of a mild exercise (Jette et al., 1990; Ainsworth et al., 2000). Subjects with controlled depression and taking antidepressants were eligible for participation in this study. Subjects were excluded if they met any of the following criteria:

1. not cleared to exercise per the subject’s physician’s notes or based on screening with the Physical Activity Readiness Questionnaire (Appendix D),
2. atypical Parkinson syndrome, (e.g. multiple system atrophy, progressive supranuclear palsy, etc.)
3. significant osteoporosis or arthritis,
4. another neurological disease,
5. self-report of claustrophobia,
6. history of cancer within five years of study participation,
7. high dose of radiation from another procedure within the year,
8. not able to tolerate being off PD medication for up to 24 hours,
9. body mass index of 35 or more,
10. female subjects who are breast-feeding or pregnant, and
11. any factor excluding MRI scanning such as an electrical brain stimulator (i.e., DBS)
12. significant or unstable cardiovascular or respiratory disease identified on the exercise stress test,
13. significant cognitive impairment (Montreal Cognitive Assessment (MoCA) score < 24) while medicated for PD,
14. significant depression (BDI score > 18) while medicated for PD and depression.
3.3.2 Study design

Each subject’s study procedures took place on three separate days across one to three weeks. All data were collected between September 1, 2013 and April 11, 2014. Each study visit (described below) was held on a separate day at UBC in Vancouver and lasted between 2-4.5 hours. Study visit 1 included an aerobic capacity test and assessments of motor and non-motor function; study visit 2 comprised two PET scans two hours apart with a 30-minute cycling exercise stimulus in between scans; and study visit 3 included an fMRI scan during which the subject played a card task, as well as assessments of motor and non-motor function. Several validated clinical tools were used during each study visit to assess motor function, cognition, mood and apathy. These clinical measures were chosen based on their validation in the PD literature and their ease of implementation. Two study investigators administered all clinical assessments (DM and MS), except for MDS-UPDRS III, which was administered by two research coordinators. Each investigator followed the same procedures, providing the same instructions to each subject. Many of the clinical measures were repeated at another study visit to gather data when subjects were taking (i.e., “on”) their regular medication for PD (study visit 1) and while they were not taking their regular medication for PD (i.e., “off”) (study visits 2 and 3). When subjects were off their medications for PD, their last dose was administered 12-18 hours prior to testing.

Study visit 1 involved determination of peak aerobic capacity (VO\textsubscript{2} peak) on a stationary bike and was conducted at the UBC Sports Medicine Centre. VO\textsubscript{2} peak is the gold standard assessment of aerobic capacity (Whaley et al., 2006) and has been validated for use in PD (Katzel et al., 2011). VO\textsubscript{2} peak is representative of cardiorespiratory fitness and is commonly
used to prescribe exercise programs recommended by the American College of Sport Medicine (Bradshaw et al., 2005; Whaley et al., 2006). VO$_2$ peak is dependent on metabolically active tissue (Albouaini et al., 2007). Thus, measuring VO$_2$ peak relative to body mass (mL/kg·min) allows for a comparison of aerobic capacity between groups and standardization of the exercise stimulus in study visit 2 to the subject’s aerobic capacity. Gender, age and physical activity level are predictors that are highly correlated with VO$_2$ peak scores; VO$_2$ peak is typically higher in men, declines with age, and higher in more physically active individuals (Bruce et al., 1973). VO$_2$ peak testing of aerobic capacity can be conducted in many ways, but the most common is in a supervised laboratory setting on a treadmill or cycling. Cycling is well tolerated by people with PD, including those with freezing of gait, in whom it is less likely to result in loss of balance (Alberts et al., 2011; Snijders et al., 2012). Therefore, cycling was also used as the exercise stimulus in study visit 2. During the VO$_2$ peak test we recorded heart rate (HR, (Polar T31 Heart Rate Monitor; Polar Electro Inc, Lake Success, NY), power output (watts, W) and rate of perceived exertion (RPE) using the Borg RPE scale (Scherr et al., 2013). The VO$_2$ peak protocol we used was a 15-30 W ramped protocol using an electrically braked stationary bicycle that increased the resistance by 15-30 W over one minute. This allowed for a direct comparison of power to use for the exercise between PET scans in study visit 2. Clinical tests completed during this study visit included measures of motor function (Unified Parkinson’s Disease Rating Scale motor section (MDS-UPDRS III)), finger tapping, and Purdue Pegboard), tests of cognition (TMT A & B, reaction time) and a written questionnaire to assess affect (PANAS). Subjects were on their regular medication for PD for testing during this study visit.

*Study visit 2* included two 60-minute RAC PET scans with an exercise stimulus performed in
The exercise stimulus was 30 minutes of cycling on a stationary bike in the PET scanning room and began within 30 minutes following completion of the first RAC scan. The subject’s second PET scan began within 15 minutes following the cycling session. The 30-minute cycling session was conducted at a workload (watts) of approximately 60% of the subject’s VO$_2$ reserve determined using the Karvonen formula ($\%$VO$_2$ reserve = $\%$(VO$_2$ peak – VO$_2$ rest) + VO$_2$ rest) and the subject’s VO$_2$ peak from study visit 1 in order to regulate the intensity of the cycling across subjects (Karvonen et al., 1957). Percentage of VO$_2$ was used instead of HR to standardize the exercise intensity because autonomic dysfunction may accompany PD and presumably impair the heart rate response to exertion (DiFrancisco-Donoghue et al., 2009). The cycling session involved 3-5 minutes of a warm-up, and then 25-30 minutes of cycling at as close as possible to 60% VO$_2$ peak and 60 revolutions per minute. During the cycling stimulus heart rate and RPE was recorded every 5 minutes. Clinical measures performed during this study visit included MDS-UPDRS III and PANAS. PANAS was conducted twice, once before the first PET scan and once after the second PET scan. Subjects were off their regular medication for PD for testing during this study visit. A more detailed description of PET data acquisition and analysis is in the sub-sections below.

Study visit 3 included an approximately 60-minute MRI/fMRI scan. The first 6 minutes of the scan captured MRI structural data, which was used for determining regions of interest (ROIs) for both MRI and PET. Following the MRI, subjects were then assessed with fMRI while playing a card game that lasted 30 minutes. Clinical measures performed during this study visit included tests of motor function (MDS-UPDRS III, finger tapping, Purdue Pegboard and TUG), tests of cognition (MoCA, TMT A & TMT B, reaction time, and WCST), a test of mood (BDI), a test of
affect (PANAS), and a test of apathy (Starkstein). Subjects were off their regular medication for PD for the testing during this study visit. A more detailed description of fMRI acquisition and analysis and the clinical tests are in the sub-sections below.

3.3.3 PET image acquisition

PET was conducted at UBC Hospital on the CTI/Siemens High Resolution Research Tomograph (HRRT) with in-plane resolution of 2.4 mm, in order to permit adequate visualization of the ventral striatum. The PET image acquisition and analysis techniques used were developed and described previously by our research group (Lidstone, 2008). The HRRT captures 206 planes with a plane thickness of 1.2 mm over 16 time frames per 60-minute scan. Subjects were positioned in a standard fashion using external lasers so the scanner gantry was parallel to the inferior orbital-external metal line. Head movement was minimized using custom fitted thermoplastic masks that were also used to assist in repositioning for the second scan.

Attenuation correction was accomplished before each emission scan by performing a 10-minute transmission scan with $^{137}$Cs rods. Each scan was started following intravenous bolus RAC administration of approximately 296 MBq (8 mCi) per scan, with effective dose of 1.85 mSv per scan. The two 60-minute RAC PET scans were performed at approximately 9:00AM and 11:00AM, thus always separated by two hours (approximately six 20-minute half-lives of $^{11}$C), to permit decay of injected radioactivity between the first and second scans.

3.3.4 PET image analysis

The second (post-exercise, “post”) PET scan was analyzed by realigning to that individual’s baseline (pre-exercise, “pre”) PET scan. Binding potential (BP) was determined in regions of
interest (ROIs) positioned over the caudate, anterior, intermediate and posterior putamen, and ventral striatum using Logan analysis with a cerebellar reference region (Logan et al., 1996), as a single elliptical ROI (2520.85 mm$^2$). As realignment between pre- and post-scans is very good, but not perfect, the position of the dorsal striatum ROIs were occasionally adjusted between the pre- and post-PET scans to capture the most radioactivity within the dorsal striatum. These ROI adjustments are common practice and were based on visualizing the activation and anatomy in that region and were only moved by at most one pixel in any direction per ROI. We measured the effects of exercise on RAC BP in ten ROIs (circular, ellipse or half-circle shapes) in five separate areas in each hemisphere of the brain (Figure 3.1). These areas included the ventral striatum and four regions of the dorsal striatum: head of caudate nucleus and three sub-regions of the putamen (anterior, intermediate and posterior). The head of the caudate ROI was always 74.3 mm$^2$, the anterior and intermediate putamen ROIs were each 47.5 mm$^2$, and the posterior putamen was 50.5 mm$^2$.

**Figure 3.1** Example of ROIs placed on PET images of the dorsal and ventral striatum. Warmer colours indicate increased RAC binding. a) PET image of four ROIs per hemisphere hand-placed on the dorsal striatum (mean of 9 slices), positioned from the top of the figure: i) caudate, ii) anterior putamen, iii) intermediate putamen, and iv) posterior putamen. b) PET image on the right shows one ROI per hemisphere placed on the ventral striatum (mean of 5 slices) per the technique illustrated in Figure 3.2.

Limitations in the spatial resolution of the PET images make the slice selection and ROI
placement for the ventral striatum more variable than in the dorsal striatum. Therefore, a validated MRI-guided ROI placement technique was used, but with some minor modifications (Mawlawi et al., 2001). To use this technique the individual’s MRI was first co-registered to their PET image using the mutual information registration algorithm (Studholme et al., 1997). The modification of the Mawlawi et al (2001) technique included creating a mean image of the five selected slices and drawing one ventral striatum ROI in each hemisphere of the brain, instead of combining five ROIs per hemisphere, each drawn separately on each of the five selected slices (Figure 3.2).

![Figure 3.2 PET ROI drawing technique for the ventral striatum.](image)

*Image from Mawlawi et al 2001 paper showing the ventral striatum drawn on an MRI image registered to a PET image. In our study the ROI size was about $110 \pm 10 \text{ mm}^2$ per hemisphere, depending on the subject’s neuroanatomy.*

We additionally classified each hemisphere of the brain as either ipsilateral or contralateral to the more symptomatically affected clinical side, which is more appropriate than physiological classification (i.e., left/right) given the frequent unilateral presentation of PD. We assigned each hemisphere as “better” (i.e., ipsilateral to the more affected limbs) or “worse” (i.e., contralateral
to the more affected limbs) based on the side of the subject’s first motor symptoms documented in their clinical records. In the four subjects where the laterality of disease onset was indistinguishable based on their clinical record, we determined the more affected hemisphere based on their most recent clinical assessment (n = 3) or, if that was inconclusive (n = 1), based on the MDS-UPDRS III assessment during study visit 2.

In order to test the primary hypothesis that habitual exercisers release more dopamine in response to acute exercise than non-exercisers, we first analyzed whether acute exercise has an effect on RAC BP in our sample population of PD subjects. For this comparison, all twelve subjects were pooled together regardless of their participation in regular exercise and corresponding cohort allocation. As we did not have a control group for this pooled analysis, we calculated percent decrease in RAC BP to establish if the percent decrease in RAC BP from exercise was greater than possible change in RAC BP without a behavioral or pharmacological intervention. Greater percent decrease in RAC BP is indicative of less RAC binding in the post-scan and, thus, dopamine release. There is one study on the reproducibility of RAC BP in the basal ganglia with a cerebellar reference region without a pharmacological or behavioral intervention in between. This study showed intra-individual variability ranged from -9% to 7%, with -1.6 ± 6% test-retest variability in the group (Volkow et al., 1993). The authors suggested that percent change in RAC BP greater than ±10% could be attributed to more than intra-individual variability (Volkow et al., 1993). However, significant differences between groups less than ±10% would also be interesting. Percent decrease in RAC BP was calculated as follows: \[
\frac{(\text{RAC BP}_{\text{pre}} - \text{RAC BP}_{\text{post}})}{\text{RAC BP}_{\text{pre}}} \times 100.\]
3.3.5  fMRI acquisition

Functional magnetic resonance imaging (fMRI) uses BOLD signal to compute functional processes from changes in blood flow. fMRI is a technique that is commonly used to study reward (Reuter et al., 2005; Yacubian et al., 2006), and was accordingly used for this project. All of the MRI/fMRI scans for this study were performed on a Philips Achieva 3.0 Tesla scanner at the UBC MRI Research Centre at UBC Hospital.

To study response to reward in the ventral striatum, subjects completed a gambling card task during the fMRI scan. The object of the chance-based card task was to select the one winning card of the four cards dealt on a computer monitor from a deck of cards displayed facedown to the subjects while in the MRI gantry. The card task was designed to maintain the subject’s expectation of receiving a monetary reward at a consistent probability throughout a portion of the fMRI scan. This task was modified from the task used by Van der Vegt et al (2013). The task was modified to include only ‘win’ or ‘no win’ trials and to include more probabilities of winning. Responses associated with reward prediction could theoretically occur in healthy subjects at either 100% probability (maximum reward value) or at 50% probability (maximum uncertainty) (Fiorillo et al., 2003). However, reward processing is abnormal in those with PD and we have previously found that placebo-induced dopamine release was maximal when the stated probability of receiving active drug was 75% (Lidstone et al., 2010). We therefore wanted to ensure that all of these possibilities were assessed. Examples of the computer screens displayed at different stages during the card task are in Appendix E.

Subjects were trained on the card task approximately 30 minutes prior to starting the 45-minute
scanning session. Subjects selected a card by pressing a button on the hand-held controller during the scan. All subjects were able to learn the task with ease and completed the task successfully. The total time playing the card task in the scanner was 30 minutes. The task consisted of four separate 7.5-minute blocks, allowing for a short (~30 s) break for the subject in between blocks. The only difference between each block was the subject’s probability of winning on each hand (i.e., trial). The four probabilities of winning tested were 0%, 50%, 75% and 100%, where either zero, two, three, or four, respectively, of the four cards displayed being winning cards. The order of these probabilities was randomized for each subject using a randomization chart. At the beginning of the block, the subjects were informed verbally and in writing about the probability of winning a monetary reward ($0.50) on each trial, up to $30.00 total. The probability was then fixed for the duration of the block and displayed in the background of the computer screen. Each block consisted of 20 trials, each 20 seconds long. Each 20-second trial was comprised of 10 seconds before the card was turned over and revealed as winning or not winning (‘anticipation phase’), and 10 seconds from when the card was revealed to the start of the next trial (‘reward phase’). During the anticipation phase a timer appeared on the screen and instructed the subject to select a card during the first 5 seconds, and then the subject waited for 5 seconds before the card was revealed. The reward period consisted of 5-8 seconds where the card was displayed as winning or losing along with a corresponding smiling face or sad face. On winning trials a $0.50 coin was added to a tally showing the total winnings, which remained on the screen during both phases. Cheering sounds were played when the smiling face was presented and disappointed-sounding sighs were played when the sad face was shown. The last portion of the reward period was 2-5 seconds (depending on the time of the first portion of this period) as a jittered interlude before the start of the next 20-second trial.
Subjects were informed there was a relationship between the number of correct choices and the amount of the reward, but all subjects were paid $30 at the end of the study, regardless of their performance. Subjects also completed a 6-minute resting state fMRI (rs-fMRI) scan during the scanning session, which was not included in this thesis, but will be a part of future analyses by our research group.

3.3.6 fMRI analysis

Each subject’s structural MRI was registered to their fMRI. An atlas-based mask in Montreal Neurological Institute (MNI) space was created for the ROI (ventral striatum) for both brain hemispheres and was placed on the MRI. The left and right ventral striatum ROI masks were then transformed to the functional image and the non-zero voxels were determined. In order to bring these ROIs into the subject’s fMRI space, both linear (FSL FLIRT) and non-linear (FSL FNIRT) registrations were performed on each of the subjects’ T1 MRI images to register them to the MNI space. The inverse registration was calculated in order to transfer from the MNI space back to each subject’s T1 space. The inverse registration matrix was then applied to the atlas-based ROIs to convert them to the subject's T1 space. Each subject’s T1 image was brought into their fMRI space, so the transformation could be calculated for each by running FSL FLIRT on the T1 images. This transformation was then applied to the ROIs in order to bring them into the subject’s functional MRI space. The summation of the transformation was the mean value of BOLD signal voxel intensity for each ROI for each of the 218 fMRI files.

To analyze the card task data an ROI-based approach was used and measured differences in percent signal change (PSC) of the BOLD signal in the ventral striatum. PSC was chosen as the
analysis method because it has been more reliable than counting the most active voxel in healthy subjects and in stroke (Kimberley et al., 2008b; Kimberley et al., 2008a; Kokotilo et al., 2010).

3.3.7 Clinical measures of motor and non-motor function

The tests for motor function included MDS-UPDRS III (Goetz et al., 2008), finger tapping, Purdue Pegboard, and TUG. For the finger tapping test subjects tapped a clicker with one finger of one hand at a time for 30 seconds, alternating between the left and right clicker with lateral movements. For the Purdue Pegboard test (Pal et al., 2001), subjects placed pegs in the cribbage-like board using one hand at a time, and then together with both hands for 30 seconds each session. As hand dominance could be a factor in performance on these two hand-based motor tasks all subjects started finger tapping and Purdue Pegboard with their dominant hand. The TUG test is commonly used as a test of mobility in frail elderly people (Podsiadlo and Richardson, 1991). This test required the subject to stand up from sitting, walk three meters, turn around and sit back down in the chair without using their arms for support to get up or sit back down. Finger tapping and Purdue Pegboard were tested both on and off medication and TUG was only tested off medication.

There were five tests of cognition used in this study: MoCA (Zadikoff et al., 2008a), WCST (computer-based) (Brown and Marsden, 1988; Tien et al., 1996), TMT A & B (Reitan, 1955) and a computerized simple reaction time test (‘reaction time’). These measures are part of the latest recommended tests to assess cognition in PD (Lee et al., 2012; Marras et al., 2014). Mood was assessed with BDI (Beck, 1996), affect was tested with the PANAS (Watson et al., 1988), and apathy was tested with Starkstein (Starkstein et al., 1992). BDI and Starkstein were tested off
medications and the PANAS was performed both on and off medications and at different times. PANAS was assessed four times, once on medication during study visit 1, twice during study visit 2 (before the first PET scan and following the second PET scan) off medication, and once during study visit 3 off medication. Different versions of the PANAS were used for each testing period; the alternate versions included the same words, but in another order. The MoCA and WCST were tested off medications, whereas TMT A & B, and reaction time were tested both on and off medications for PD. TMT A & B tests cognitive flexibility and in particular performance on TMT B can predict instrumental activities of daily living (Cahn et al., 1998; Higginson et al., 2013). Different versions of the TMT A & B tests were used on/off medications to limit the practice effect. The alternate versions included the numbers and letters in a different order. Examples of the study forms used for measures of motor and non-motor function are in Appendices F-L.

3.4 Statistical analyses

All analyses were performed using STATISTICA (data analysis software system), version 8.0 (copyright © StatSoft, Inc, Dell, www.statsoft.com). Dependent variables were log transformed to make them more normally distributed. Variables that were log transformed include VO$_2$ peak, body mass index (BMI), weight, height, systolic blood pressure, diastolic blood pressure, resting heart rate, heart rate at VO$_2$ peak, TMT A & B, reaction time, TUG, and RAC BP. All variables that were measured as a rate are Poisson distributed and were therefore square root transformed (McCullagh and Nelder, 1989). These variables include exercise participation (counts/week), exercise duration (minutes/week), finger tapping and Purdue Pegboard. The significance level for all tests was predetermined at $\alpha \leq 0.05$. 
Unpaired two-tailed Student’s t-tests were used to compare all subject characteristics, and clinical measures of motor function, mood, apathy and cognition, except for MDS-UPDRS III which was measured using repeated measures analysis of variance (ANOVA). Repeated measures ANOVA was used to compare RAC BP between cohorts. Analysis of covariance (ANCOVA) was used to compare percent decrease in RAC BP between cohorts (RAC BP_{pre} – \text{RAC BP}_{post}/\text{RAC BP}_{pre} *100) using RAC BP_{pre} and disease duration as covariates. As percent decrease in RAC BP was compared as a ratio (i.e., percent), untransformed RAC BP values were entered into the above percent decrease equation instead of log transformed RAC BP values. Other covariates were also explored using Spearman rank order correlations, including age, disease severity, minutes since last dose of PD medication, exercise participation per week and exercise duration per week. Tukey’s post-hoc analysis was performed if there were significant F ratios.

The outcomes to the fMRI card task described are for response to all 20 trials for both anticipation and reward phases for all four probabilities, and only responses to winning trials during the reward phase for 50% and 75% probabilities. To analyze change in fMRI BOLD signal in the ventral striatum, PSC was calculated for all five ~2 s TRs (repetition times) during each of the anticipation and reward phases using the following formula: (BOLD value per TR – mean of BOLD values per phase)/mean of BOLD values per phase. For each subject the 20 trials per probability block were averaged at each of the 5 TRs per phase (anticipation/reward). The peak PSC of the averaged 5 TRs per phase was determined for each subject. To compare PSC to the subject’s baseline BOLD signal we also calculated PSC for all TRs that preceded the start of each probability block (n = ~4 per probability, therefore n = ~32 for four blocks per subject).
generate the upper limit of the 95% confidence interval of baseline PSC, we used MatLab (MATLAB 8.1, The MathWorks, Inc., Natick, Massachusetts, United States) bootstrap functions ‘bootci’ and ‘bootstat’ to develop a 95% confidence interval from the baseline pre-trial TRs (n= ~32), for each subject. The median baseline PSC per cohort was also calculated to compare to the mean PSC during each probability block. Repeated measures ANOVA was performed using peak PSC per subject for each probability (0%, 50%, 75%, 100%), hemisphere (better/worse) and response phase (anticipation/reward). Repeated measures ANCOVA was also performed, including baseline PSC and disease duration as covariates. As we were also interested in differences between 0% and 50%, 0% and 75%, as well as interactions at each probability, these comparisons were investigated separately with repeated measures ANCOVA.

3.5 Results

3.5.1 Subject characteristics

The subjects in each cohort were similar on all measured parameters except habitual exercisers had greater VO2 peak, more weekly participation (counts) in mild to vigorous exercise and higher weekly duration of participation (minutes) in moderate to vigorous exercise (Table 3.1). These cohort differences confirmed the cohort allocations were appropriate and that other characteristics were unlikely to cause differences in the outcome measures. Notably, variables such as age, disease duration, MoCA, daily levodopa dose-equivalent medications for PD, and disease severity (MDS-UPDRS III and Hoehn & Yahr, H&Y) could impact the findings of this study, but these measures were similar between cohorts.
Table 3.1 Subject characteristics.

<table>
<thead>
<tr>
<th></th>
<th>Non-regular (n=6), mean</th>
<th>Non-regular, SD</th>
<th>Habitual (n=6), mean</th>
<th>Habitual, SD</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>67.5</td>
<td>2.6</td>
<td>64.0</td>
<td>3.6</td>
<td>0.0809</td>
</tr>
<tr>
<td>Gender (male, female)</td>
<td>4, 2</td>
<td></td>
<td>6, 0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Right handed (right, left)</td>
<td>5, 1</td>
<td></td>
<td>5, 1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Height (cm)</td>
<td>169.5</td>
<td>8.8</td>
<td>176.2</td>
<td>8.6</td>
<td>0.2121</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>80.2</td>
<td>19.0</td>
<td>74.0</td>
<td>11.6</td>
<td>0.6193</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>27.6</td>
<td>4.8</td>
<td>23.8</td>
<td>3.0</td>
<td>0.1565</td>
</tr>
<tr>
<td>Rest systolic (mm Hg)</td>
<td>135.3</td>
<td>14.1</td>
<td>119.7</td>
<td>12.2</td>
<td>0.0729</td>
</tr>
<tr>
<td>Rest diastolic (mm Hg)</td>
<td>79.3</td>
<td>6.4</td>
<td>73.0</td>
<td>4.0</td>
<td>0.0647</td>
</tr>
<tr>
<td>Resting heart rate (bpm)</td>
<td>78.0</td>
<td>12.5</td>
<td>70.0</td>
<td>10.1</td>
<td>0.2653</td>
</tr>
<tr>
<td>Heart rate at 60% VO₂ peak</td>
<td>111.8</td>
<td>12.4</td>
<td>117.0</td>
<td>15.8</td>
<td>0.5134</td>
</tr>
<tr>
<td>Heart rate at VO₂ peak (bpm)</td>
<td>130.0</td>
<td>16.9</td>
<td>150.8</td>
<td>16.8</td>
<td>0.0619</td>
</tr>
<tr>
<td>VO₂ peak (mL/kg-min)</td>
<td>20.2</td>
<td>2.6</td>
<td>36.9</td>
<td>6.8</td>
<td>0.0001*</td>
</tr>
<tr>
<td>Participation in exercise per week (counts)</td>
<td>1.8</td>
<td>0.9</td>
<td>16.5</td>
<td>9.8</td>
<td>&lt;0.0001*</td>
</tr>
<tr>
<td>Duration of exercise per week (min)</td>
<td>79.9</td>
<td>47.9</td>
<td>566.6</td>
<td>338.9</td>
<td>0.0004*</td>
</tr>
<tr>
<td>Disease duration (years)</td>
<td>5.8</td>
<td>2.3</td>
<td>5.3</td>
<td>4.5</td>
<td>0.8134</td>
</tr>
<tr>
<td>H&amp;Y, on</td>
<td>2.2</td>
<td>0.4</td>
<td>1.8</td>
<td>0.8</td>
<td>0.3628</td>
</tr>
<tr>
<td>H&amp;Y, off</td>
<td>2.0</td>
<td>0.6</td>
<td>1.7</td>
<td>0.8</td>
<td>0.4475</td>
</tr>
<tr>
<td>MDS-UPDRS III, on</td>
<td>18.4</td>
<td>6.8</td>
<td>17.3</td>
<td>14.0</td>
<td>0.7932</td>
</tr>
<tr>
<td>MDS-UPDRS III, off</td>
<td>20.3</td>
<td>10.6</td>
<td>19.9</td>
<td>11.9</td>
<td>0.7932</td>
</tr>
<tr>
<td>MoCA</td>
<td>28.5</td>
<td>1.6</td>
<td>28.0</td>
<td>1.1</td>
<td>0.5490</td>
</tr>
<tr>
<td>Daily levodopa dose</td>
<td>604.2</td>
<td>220.5</td>
<td>443.8</td>
<td>287.0</td>
<td>0.3030</td>
</tr>
<tr>
<td>Daily dose PD medications (levodopa equivalents)</td>
<td>620.8</td>
<td>241.1</td>
<td>535.4</td>
<td>409.8</td>
<td>0.6692</td>
</tr>
<tr>
<td>More PD-affected brain hemisphere</td>
<td>4, right</td>
<td>4, right</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>2, left</td>
<td>2, left</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Cohorts only differed in terms of their regular participation in exercise. Only moderate to vigorous activities greater than 3 MET were considered for total length of exercise per week. The “off” scores for H&Y and MDS-UPDRS III were collected during study visit 2. MDS-UPDRS was analyzed using repeated measures ANOVA. The findings were not impacted by accounting for effects of covariates, including age, duration of disease and minutes since their last dose of PD medications. P-values at α ≤ 0.05 are denoted with an asterisk (*).

The variable ‘daily levodopa dose’ in Table 3.1 indicates the amount of immediate release levodopa/carbidopa. The ‘daily dose of PD medications (levodopa equivalents)’ indicates the
amount of daily immediate release levodopa/carbidopa as well as the levodopa equivalents of dopamine agonists and adjuncts based on established medication conversion factors (Tomlinson et al., 2010). One subject per cohort took a daily dopamine agonist and two subjects in the habitual exerciser cohort took a daily adjunct therapy. In terms of other medications for symptoms of PD, five non-regular exercisers and one habitual exerciser were taking an anti-depressant, although all had controlled depression during study participation, as well as prior to study participation based on their neurologist’s clinic notes (anti-depressant prescription for individual non-regular exercisers: 30 mg mirtazapine (n = 2), 100 mg desvenlafaxine, 75 mg venlafaxine, 20 mg paroxetine; and for habitual exercisers: 50 mg mirtazapine). One non-regular exerciser and three habitual exercisers were prescribed a cholinesterase inhibitor for cognitive decline at the time of study participation. Two non-regular exercisers and one habitual exerciser were taking medication for their blood pressure (all hydrochlorothiazide, except for one individual who was additionally taking ramipril). One subject in each cohort had previously used cocaine. Both subjects who had used cocaine indicated they had not used it in the past few years, and one of those individuals had used cocaine only infrequently.

The subjects in each cohort had similar levels of education. All subjects had graduated from high school and nearly all (n=11) had completed a post-secondary degree. Therefore, it is unlikely that level of education contributed to differences in outcome variables. It is important to note there were only two women who participated in this study, and both were in the non-regular exerciser cohort. While statistical differences between these individuals and other subjects cannot be compared due to the low sample size, a description of these individuals helps to understand whether their characteristics could have impacted the results of this study. These two women
were comparable to the male non-regular exercisers in terms of their handedness, blood pressure, heart rate at VO₂ peak, disease duration and H&Y. The women weighed less, had a higher resting heart rate and lower VO₂ peak compared to the men in their cohort. One of the women was two years older than the second oldest of all subjects and also had a lower MoCA score (26). The other woman had a lower BMI (19.3 kg/m²) than the male non-exercisers, but a comparable BMI to the males in the habitual exerciser cohort.

The subjects experienced a similar exercise stimulus during study visit 2 (Table 3.2). During this study visit, subjects exercised on a stationary bicycle for 30 minutes in between two PET scans. The second PET scan began within 15 minutes following the end of the exercise session. All subjects were able to keep their cycling rate within a few revolutions per minute (RPM) of 60 RPM. Some subjects in the habitual exerciser group found the cycling speed to be too low, whereas subjects in the non-exerciser group did not report the speed felt too slow.

<table>
<thead>
<tr>
<th></th>
<th>Non-regular, mean</th>
<th>Non-regular, SD</th>
<th>Habitual, mean</th>
<th>Habitual, SD</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Targeted wattage</td>
<td>56.7</td>
<td>28.2</td>
<td>152.7</td>
<td>27.9</td>
<td>0.0001*</td>
</tr>
<tr>
<td>(based on 60% VO₂ peak)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Average RPM</td>
<td>58.3</td>
<td>4.1</td>
<td>60.0</td>
<td>0.0</td>
<td>0.3409</td>
</tr>
<tr>
<td>RPE at 5 min</td>
<td>11.0</td>
<td>2.1</td>
<td>12.2</td>
<td>1.2</td>
<td>0.2759</td>
</tr>
<tr>
<td>Heart rate at 5 min</td>
<td>107.8</td>
<td>22.8</td>
<td>118.7</td>
<td>14.3</td>
<td>0.3593</td>
</tr>
<tr>
<td>RPE at 10 min</td>
<td>13.0</td>
<td>1.4</td>
<td>13.5</td>
<td>1.4</td>
<td>0.5490</td>
</tr>
<tr>
<td>Heart rate at 10 min</td>
<td>113.2</td>
<td>22.3</td>
<td>125.5</td>
<td>16.3</td>
<td>0.2992</td>
</tr>
<tr>
<td>RPE at 15 min</td>
<td>13.5</td>
<td>1.1</td>
<td>13.7</td>
<td>1.5</td>
<td>0.8284</td>
</tr>
<tr>
<td>Heart rate at 15 min</td>
<td>117.5</td>
<td>23.1</td>
<td>131.5</td>
<td>14.6</td>
<td>0.2382</td>
</tr>
<tr>
<td>RPE at 20 min</td>
<td>14.2</td>
<td>1.0</td>
<td>14.2</td>
<td>2.1</td>
<td>1.0000</td>
</tr>
<tr>
<td>Heart rate at 20 min</td>
<td>119.2</td>
<td>22.8</td>
<td>133.7</td>
<td>16.1</td>
<td>0.2321</td>
</tr>
<tr>
<td>RPE at 25 min</td>
<td>15.5</td>
<td>1.5</td>
<td>14.7</td>
<td>2.3</td>
<td>0.4807</td>
</tr>
</tbody>
</table>

Table 3.2 Characteristics of the 30-minute exercise stimulus between PET scans.
Other than the targeted watts, the stimulus was the same for each cohort. The time of cycling was not exactly 30 minutes as cycling was stopped when RAC was ready to be delivered to the UBC PET suite. P-values at $\alpha \leq 0.05$ are denoted with an asterisk (*).

In order to provide some warm-up for the cycling exercise, the session was started at 40-50% of the subject’s VO$_2$ peak for a few minutes and was then increased to the targeted 60% of their VO$_2$ peak for the remainder of the session. To keep the exercise stimulus as close as possible to 60% of the workload at the individual’s VO$_2$ peak, the resistance was adjusted on occasion during the exercise based on the individual’s reported RPE or their heart rate. One non-exerciser had his (her) watts increased by 10 W 7 minutes into the exercise, one non-exerciser had his (her) wattage decreased by 10 W 10 minutes into the exercise and a habitual exerciser had his (her) wattage decreased by 10-30 W in the last 5 minutes of the cycling session.

### 3.5.2 Neuroimaging outcomes

#### 3.5.2.1 $[^{11}]$C Raclopride binding potential measured with PET

First, percent decrease in RAC BP was calculated across all 12 pooled subjects, irrespective of allocated cohort. Percent decrease in RAC BP was assessed for each of the ten ROIs, including 5 primary striatal regions with one ROI per brain hemisphere (Table 3.3). Significant variability in percent decrease in RAC BP was found in each ROI.

<table>
<thead>
<tr>
<th></th>
<th>regular (n=6), mean</th>
<th>regular, SD</th>
<th>(n=6), mean</th>
<th>SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>Heart rate at 25 min</td>
<td>121.3</td>
<td>23.8</td>
<td>155.7</td>
<td>55.7</td>
</tr>
<tr>
<td>RPE at session end</td>
<td>16.8</td>
<td>1.1</td>
<td>15.4</td>
<td>2.2</td>
</tr>
<tr>
<td>Heart rate at session end</td>
<td>121.83</td>
<td>23.85</td>
<td>140.33</td>
<td>16.00</td>
</tr>
<tr>
<td>Total time cycling (min)</td>
<td>28.84</td>
<td>1.65</td>
<td>29.79</td>
<td>1.97</td>
</tr>
</tbody>
</table>

Table 3.3 Percent decrease in RAC BP across 12 study subjects pooled regardless of cohort.
<table>
<thead>
<tr>
<th>Region of interest</th>
<th>Mean % (n=12)</th>
<th>Median % (n=12)</th>
<th>± 95% Confidence interval</th>
</tr>
</thead>
<tbody>
<tr>
<td>Better ventral striatum</td>
<td>1.86</td>
<td>2.05</td>
<td>5.00</td>
</tr>
<tr>
<td>Worse ventral striatum</td>
<td>4.16</td>
<td>5.25</td>
<td>3.69</td>
</tr>
<tr>
<td>Better caudate</td>
<td>0.70</td>
<td>-0.63</td>
<td>3.74</td>
</tr>
<tr>
<td>Worse caudate</td>
<td>2.23</td>
<td>2.88</td>
<td>3.29</td>
</tr>
<tr>
<td>Better anterior putamen</td>
<td>-1.73</td>
<td>-0.73</td>
<td>4.65</td>
</tr>
<tr>
<td>Worse anterior putamen</td>
<td>1.94</td>
<td>2.44</td>
<td>2.73</td>
</tr>
<tr>
<td>Better intermediate putamen</td>
<td>-0.59</td>
<td>3.59</td>
<td>5.45</td>
</tr>
<tr>
<td>Worse intermediate putamen</td>
<td>-1.51</td>
<td>-0.68</td>
<td>3.21</td>
</tr>
<tr>
<td>Better posterior putamen</td>
<td>-0.90</td>
<td>-1.82</td>
<td>4.06</td>
</tr>
<tr>
<td>Worse posterior putamen</td>
<td>-0.99</td>
<td>-0.47</td>
<td>4.91</td>
</tr>
</tbody>
</table>

Greater percent decrease in RAC BP is indicative of dopamine release. Percent decrease in RAC BP is calculated using the following formula: \( \frac{(\text{RAC BP}_{\text{pre}} - \text{RAC BP}_{\text{post}})}{\text{RAC BP}_{\text{pre}}} \times 100. \)

Wilcoxon Signed-Rank Test was performed and found the worse ventral striatum, caudate and anterior putamen had percent change in RAC BP that was significantly different compared to the mean of the reproducibility sample (-1.6%) (p = 0.023, 0.049, and 0.034, respectively). Mean percent change in RAC BP values were less than 10% for all regions, although the median percent change in the worse ventral striatum reached 5.25%. These findings suggest that acute exercise does not have a unilateral effect on RAC BP in PD. However, positive and negative percent changes in RAC BP greater than 10% occurred in individual subjects in several regions in both cohorts. Given that the reproducibility study by Volkow and colleagues was based on five healthy males without PD, younger than our study population (21-46 years), and the scans in that study were performed 24 hours apart, these reproducibility data are important to consider but not appropriate for absolute comparison to this research (Volkow et al., 1993). Therefore, we next investigated if percent decrease in RAC BP in response to acute exercise differed between habitual and non-regular exercisers. We found that percent decrease in RAC BP was only significantly different by cohort in the anterior putamen, where there was greater percent
decrease in RAC BP (i.e., dopamine release) in non-regular exercisers compared to habitual exercisers in the better hemisphere ($F_{(1, 7)}=36.73, p = 0.0005$) (Figure 3.5). There was also an effect of hemisphere in the anterior putamen, with greater percent decrease in RAC BP in the worse hemisphere ($2.67 \pm 2.04\%$) compared to the better hemisphere ($-2.21 \pm 2.15\%$) (mean ± SE; $F_{(1, 7)} = 22.54, p = 0.002$). The mean percent decrease in RAC BP and standard error for each striatal region are shown below (Figures 3.3-3.7).

**Figure 3.3** Percent decrease in RAC BP in the ventral striatum.
There were no differences in the ventral striatum ($F_{(1, 7)} = 0.36, p = 0.567$). The 95% confidence intervals per hemisphere (B: better; W: worse) and cohort (NR: non-regular, H: habitual) are as follows: -7.7 to 11.6% B/NR, -8.5 to 10.8% B/H, -0.6 to 14.6% W/NR, -5.8 to 9.5% W/H.
There were no differences in the caudate (F(1, 7) = 0.45, p = 0.526). The 95% confidence intervals per hemisphere (B: better; W: worse) and cohort (NR: non-regular, H: habitual) are as follows: -6.6 to 7.2% B/NR, -6.4 to 7.5% B/H, -1.1 to 8.4% W/NR, -3.3 to 6.3% W/H.

There was a hemisphere-by-cohort interaction in the anterior putamen (F(1, 7) = 36.73, p = 0.0005). The difference between cohorts on the better side was significant with ANCOVA (F(2, 6) = 15.83, p = 0.004). Tukey post-hoc analysis showed that in the better hemisphere there was dopamine release in non-regular exercisers and the opposite, increased RAC BP, in habitual exercisers (p = 0.0005). There was also an interaction by hemisphere: the worse hemisphere had greater dopamine release compared to the better side (F(1, 7) = 22.54, p = 0.002). The 95% confidence intervals per hemisphere (B: better; W: worse) and cohort (NR: non-regular, H: habitual) are as follows: -4.2 to 11.1% B/NR, -15.5 to -0.2% B/H, -4.4 to 10.1% W/NR, -4.8 to 9.7% W/H. P-values at α ≤ 0.05 are denoted with an asterisk (*).
Figure 3.6 Percent decrease in RAC BP in the intermediate putamen. There were no differences in the intermediate putamen ($F_{(1, 7)} = 0.13, p = 0.730$). The 95% confidence intervals per hemisphere (B: better; W: worse) and cohort (NR: non-regular, H: habitual) are as follows: -23.6 to 14.2% B/NR, -17.9 to 19.9% B/H, -13.0 to 4.2% W/NR, -10.2 to 7.1% W/H.

![Intermediate putamen graph](image)

Figure 3.7 Percent decrease in RAC BP in the posterior putamen. There were no differences in the posterior putamen ($F_{(1, 7)} = 0.05, p = 0.827$). The 95% confidence intervals per hemisphere (B: better; W: worse) and cohort (NR: non-regular, H: habitual) are as follows: -14.6 to 5.4% B/NR, -8.3 to 11.8% B/H, -18.7 to 6.3% W/NR, -10.6 to 14.3% W/H.

![Posterior putamen graph](image)

As when all 12 subjects were pooled, when percent decrease in RAC BP was compared between cohorts there was significant variability for all ROIs in both hemispheres. Given the wide
confidence intervals, we next investigated if the variability in percent decrease in RAC BP may have been due to inherent differences between cohorts in terms of their inherent ability to bind RAC, in general, in either the pre- or post-PET scan. We found there was not a statistical difference in any ROI in RAC BP_{pre} or in RAC BP_{post} between cohorts (F(1, 9) = 0.139, p = 0.718, no time-by-cohort interaction). Interestingly, the mean RAC BP of habitual exercisers in the pre- and post-scan was descriptively higher (although not reaching statistical significance) than the mean of non-regular exercisers across all ROIs in the ventral striatum, caudate and anterior putamen. Figures 3.8-3.12 show the mean RAC BP and confidence intervals, in order to demonstrate the variability in RAC BP.

**Figure 3.8** RAC BP in the ventral striatum.
There were no differences in RAC BP in the ventral striatum (mean ± 95% CI).
Figure 3.9 RAC BP in the caudate. There were no differences in RAC BP in the caudate (mean ± 95% CI).

Figure 3.10 RAC BP in the anterior putamen. There were no differences in RAC BP in the anterior putamen (mean ± 95% CI).
3.5.2.2 Response to rewarding stimuli in the ventral striatum using fMRI

First, baseline PSC in BOLD signal per individual was calculated. The median baseline PSC per cohort was 0.156% for non-regular exercisers and 0.145% for habitual exercisers. Next, the differences in PSC between cohorts during the four probability blocks (0%, 50%, 75%, 100%)
were compared with three-way repeated measures ANOVA (probability/phase/cohort). A main effect of phase and a phase-by-hemisphere interaction was found (Figure 3.13). Both cohorts had greater PSC across probability blocks in the reward phase compared to the anticipation phase and there was a greater difference in response to reward compared to anticipation in the better hemisphere ($F_{(1, 9)} = 10.96, p = 0.009$). Importantly, these interactions were not present with repeated measures ANCOVA, accounting for covariates including each individual’s baseline PSC and disease duration ($F_{(3, 21)} = 0.33 p = 0.804$).

![Figure 3.13](image)

**Figure 3.13** Effects of phase and hemisphere on percent signal change (PSC). This analysis did not account for baseline PSC. Habitual exercisers and non-regular exercisers are combined as there was no effect of cohort. There was greater activation (PSC) in reward compared to anticipation ($F_{(1, 9)} = 9.78, p = 0.012$, denoted as *). Tukey post-hoc analysis showed a greater difference between anticipation and reward in the better hemisphere ($p = 0.0005$, denoted as **). PSC exceeded the median baseline PSC for both cohorts combined (0.149%, represented with a horizontal line). These effects were not present when baseline PSC and disease duration were accounted for as covariates ($F_{(3, 21)} = 0.33 p = 0.804$).
While there were no differences in PSC between cohorts in terms of probability of winning, phase or hemisphere, as with RAC BP\textsubscript{pre} versus RAC BP\textsubscript{post} comparison described above in section 3.5.2.1, there was an interesting non-significant trend in the amount of PSC between cohorts. The mean and standard errors are shown below using repeated measures ANCOVA with baseline PSC and disease duration as covariates. Whether the PSC values exceeded the baseline PSC (median per cohort, horizontal line across bars) is shown and described below each figure.

![Figure 3.14 Percent signal change at 0% probability of winning (mean ± SE). All PSC values exceeded the median baseline PSC (0.145%) for habitual exercisers. Only reward in the better hemisphere exceeded the median baseline PSC (0.156%) for non-regular exercisers.](image-url)
Figure 3.15 Percent signal change at 50% probability of winning (mean ± SE). All PSC values exceeded the median baseline PSC (0.145%) for habitual exercisers. Reward in the better and worse hemispheres exceeded the median baseline PSC (0.156%) for non-regular exercisers.

Figure 3.16 Percent signal change at 75% probability of winning (mean ± SE). All PSC values exceeded the median baseline PSC (0.145%) for habitual exercisers. Reward in the better and worse hemispheres exceeded the median baseline PSC (0.156%) for non-regular exercisers. N = 5 for non-regular exercisers at this probability as there was a technological issue with the MRI head coil.
Figure 3.17 Percent signal change at 100% probability of winning (mean ± SE). All PSC values exceeded the median baseline PSC (0.145%) for habitual exercisers. Anticipation in the worse hemisphere exceeded the median baseline PSC (0.156%) for non-regular exercisers.

While each of the four probabilities of winning should be analyzed together in repeated measures ANCOVA, we were specifically interested in any effects within each probability which could be hypothesis-generating for future studies. There were no effects in 0%, 50%, and 75%, but there was a phase-by-cohort interaction at 100% ($F_{(1, 8)} = 13.58, p = 0.006$). As can be seen in Figure 3.17, habitual exercisers have an increased response to reward compared to anticipation whereas non-regular exercisers have a similar response to anticipation and reward. With Tukey post-hoc analysis, the difference between anticipation and reward in habitual exercisers is significant ($p = 0.017$). We were also particularly curious about the difference in response at 0% compared to 50% and 0% compared to 75% chance of winning. While there was no difference between 0% and 75%, when 0% and 50% were compared it was found that habitual exercisers had greater PSC across phases compared to non-regular exercisers, and the p-value was improved using
baseline PSC and disease duration as covariates (mean ± SE: 0.24 ± 0.018% habitual, 0.16 ± 0.018% non-regular; \( F_{(1, 8)} = 9.69, p = 0.014 \)).

### 3.5.3 Clinical outcomes

#### 3.5.3.1 Measures of motor function

Tests of clinical motor function showed significant differences between cohorts, with habitual exercisers able to finger tap at a faster rate, place more pegs with their right hand on the Purdue Pegboard and walk faster in the TUG test (Table 3.4).

<table>
<thead>
<tr>
<th></th>
<th>Non-regular (n=6), mean</th>
<th>Non-regular, SD</th>
<th>Habitual (n=6), mean</th>
<th>Habitual, SD</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>TUG, off</td>
<td>10.2</td>
<td>1.3</td>
<td>7.3</td>
<td>1.4</td>
<td>0.0142*</td>
</tr>
<tr>
<td>Finger tapping, R hand, on</td>
<td>32.0</td>
<td>3.5</td>
<td>39.5</td>
<td>4.5</td>
<td>0.0088*</td>
</tr>
<tr>
<td>Finger tapping, L hand, on</td>
<td>28.8</td>
<td>3.0</td>
<td>37.9</td>
<td>8.9</td>
<td>0.0302*</td>
</tr>
<tr>
<td>Finger tapping, R hand, off</td>
<td>32.5</td>
<td>4.6</td>
<td>35.0</td>
<td>4.8</td>
<td>0.2590</td>
</tr>
<tr>
<td>Finger tapping, L hand, off</td>
<td>30.9</td>
<td>2.8</td>
<td>33.5</td>
<td>5.3</td>
<td>0.2552</td>
</tr>
<tr>
<td>Purdue Pegboard</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>R hand, on</td>
<td>9.5</td>
<td>2.2</td>
<td>12.2</td>
<td>1.6</td>
<td>0.0400*</td>
</tr>
<tr>
<td>L hand, on</td>
<td>9.8</td>
<td>2.9</td>
<td>10.8</td>
<td>1.9</td>
<td>0.4638</td>
</tr>
<tr>
<td>Both hands, on</td>
<td>15.5</td>
<td>5.9</td>
<td>17.5</td>
<td>6.8</td>
<td>0.6328</td>
</tr>
<tr>
<td>R hand, off</td>
<td>10.2</td>
<td>3.5</td>
<td>11.2</td>
<td>3.2</td>
<td>0.2907</td>
</tr>
<tr>
<td>L hand, off</td>
<td>10.0</td>
<td>3.3</td>
<td>11.5</td>
<td>2.7</td>
<td>0.2187</td>
</tr>
<tr>
<td>Both hands, off</td>
<td>13.6</td>
<td>6.2</td>
<td>18.5</td>
<td>5.1</td>
<td>0.1144</td>
</tr>
</tbody>
</table>

*On” and “off” indicate if subjects were taking their regular medication for PD. *n=4 for non-regular cohort. P-values at \( \alpha \leq 0.05 \) are denoted with an asterisk (*).

There was a main effect of cohort in the TUG as shown in Table 3.4; habitual exercisers were faster on average faster over the three trials compared to the non-regular exercisers (\( F_{(1, 8)} = 9.74, \))
p= 0.014). However, there was also a trial effect (p = 0.025), as the second and third trials in each cohort were faster than the first. As both cohorts had a slower first trial there was not a cohort-by-trial-effect (p = 0.784). Therefore, the mean and standard deviation reported in Table 3.4 are only for the second and third trials of the TUG as the first trial was considered a practice trial. The first trial was not deemed indicative of their average ability to stand up, walk a short distance, turnaround and sit back down. Only the main effect of cohort remained when just the second and third trials were compared (F(1, 8) = 8.52, p= 0.0193). The finger tapping and Purdue Pegboard results were analyzed for all subjects based on their hand-dominant side compared to their non-hand-dominant side. When better/worse hemispheres were compared instead of right/left, significant differences only remained for finger tapping on medications for both the better (p = 0.012) and worse (p = 0.049) hemispheres (mean ± SD: 30.25 ± 3.96 taps for non-regular exercisers, 41.08 ± 7.70 taps for habitual exercisers, hand contralateral to better hemisphere; 30.58 ± 3.44 taps for non-regular exercisers, 36.33 ± 5.26 taps for habitual exercisers, hand contralateral to hemisphere). There were no significant findings in the tests of motor function displayed in Table 3.4 when Bonferroni correction was applied.

3.5.3.2 Measures of mood and apathy

Habitual exercisers had improved mood and apathy scores and more positive affect off PD medication only in comparison to non-regular exercisers (Table 3.5). On both BDI and Starkstein, a higher score indicates worse mood or apathy, respectively. The apathy score is divided into two sub-scores because the questionnaire is split into the first eight questions and last six questions based on the rating scheme. Habitual exercisers also indicated they felt more positive affect on the PANAS when tested at several time points while off their regular
medication for PD. A higher score for words connoting positive emotions indicates more positive affect. Likewise, a higher score for words connoting negative emotions indicates more negative affect. There were no significant findings in the mood outcomes displayed in Table 3.5 when Bonferroni correction was applied.

Table 3.5 Outcomes of tests of mood and apathy.

<table>
<thead>
<tr>
<th>Test</th>
<th>Non-regular (n=6), mean</th>
<th>Non-regular, SD</th>
<th>Habitual (n=6), mean</th>
<th>Habitual, SD</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Beck Depression Inventory</td>
<td>13.3</td>
<td>7.0</td>
<td>5.2</td>
<td>4.6</td>
<td>0.0392*</td>
</tr>
<tr>
<td>Starkstein Apathy Scale, questions 1-8(^1)</td>
<td>8.0</td>
<td>4.9</td>
<td>2.5</td>
<td>1.1</td>
<td>0.0232*</td>
</tr>
<tr>
<td>Starkstein Apathy Scale, questions 9-14(^1)</td>
<td>6.0</td>
<td>2.6</td>
<td>4.8</td>
<td>2.3</td>
<td>0.4470</td>
</tr>
<tr>
<td>Starkstein Apathy Scale, total(^1)</td>
<td>14.0</td>
<td>7.2</td>
<td>7.3</td>
<td>3.1</td>
<td>0.0696</td>
</tr>
<tr>
<td>PANAS, on medication, positive words</td>
<td>32.8</td>
<td>7.0</td>
<td>40.3</td>
<td>5.5</td>
<td>0.0665</td>
</tr>
<tr>
<td>PANAS, on medication, negative words</td>
<td>13.0</td>
<td>3.5</td>
<td>11.0</td>
<td>1.7</td>
<td>0.2317</td>
</tr>
<tr>
<td>PANAS, off medication, pre-PET scan, positive words</td>
<td>32.7</td>
<td>5.7</td>
<td>41.2</td>
<td>5.1</td>
<td>0.0214*</td>
</tr>
<tr>
<td>PANAS off medication, pre-PET scan, negative words</td>
<td>11.8</td>
<td>1.3</td>
<td>11.5</td>
<td>2.3</td>
<td>0.8200</td>
</tr>
<tr>
<td>PANAS, off medication, post-PET scan, positive words</td>
<td>32.2</td>
<td>5.6</td>
<td>42.8</td>
<td>5.9</td>
<td>0.0196*</td>
</tr>
<tr>
<td>PANAS, off, post-PET scan, negative words(^2)</td>
<td>10.0</td>
<td>0.0</td>
<td>10.0</td>
<td>0.0</td>
<td>-</td>
</tr>
<tr>
<td>PANAS, off medication, pre-MRI scan, positive words</td>
<td>31.3</td>
<td>7.2</td>
<td>42.5</td>
<td>7.8</td>
<td>0.0269*</td>
</tr>
<tr>
<td>PANAS, off medication, pre-MRI scan, negative words</td>
<td>14.0</td>
<td>2.0</td>
<td>12.7</td>
<td>2.3</td>
<td>0.3134</td>
</tr>
</tbody>
</table>

"On" and "off" indicate if subjects were taking their regular medication for PD (tested at separate study visits). Beck Depression Inventory (BDI) and Starkstein were only tested off medication. PANAS was performed four times: before the VO\(_2\) peak test, before the MRI scan, and both before and after PET scanning. P-values at \(\alpha \leq 0.05\) are denoted with an asterisk (*). \(^1\)n=5 for non-regular cohort, \(^2\)n=5 for both cohorts.
3.5.3.3 Measures of cognitive function

There were no differences in measures of cognition between cohorts (Table 3.6). There was a trial effect \((p = 0.024)\) and trial-by-cohort effect \((p = 0.005)\) for reaction time off medication (Figure 3.18). Post-hoc analysis with Tukey showed the first trial for the non-regular exercise cohort was significantly different than all other trials for both the non-regular exercise cohort and the habitual exerciser cohort \((p\text{-values were}<0.018)\). The second, third and fourth trials were similar in each cohort. Therefore, the fastest reaction time was compared and was not found to be significant; the mean of the fastest trial is reported in Table 3.6.

Table 3.6 Outcomes of measures of cognitive function.

<table>
<thead>
<tr>
<th></th>
<th>Non-regular (n=6), mean</th>
<th>Non-regular, SD</th>
<th>Habitual (n=6), mean</th>
<th>Habitual, SD</th>
<th>(p)</th>
</tr>
</thead>
<tbody>
<tr>
<td>TMT A, on (s)(^1)</td>
<td>42.2</td>
<td>10.3</td>
<td>34.9</td>
<td>10.7</td>
<td>0.2750</td>
</tr>
<tr>
<td>TMT B, on (s)</td>
<td>148.5</td>
<td>94.3</td>
<td>92.7</td>
<td>59.0</td>
<td>0.1877</td>
</tr>
<tr>
<td>TMT A, off (s)</td>
<td>40.4</td>
<td>12.4</td>
<td>32.8</td>
<td>13.2</td>
<td>0.2475</td>
</tr>
<tr>
<td>TMT B, off (s)</td>
<td>121.0</td>
<td>67.9</td>
<td>78.1</td>
<td>36.1</td>
<td>0.2469</td>
</tr>
<tr>
<td>Reaction time, on (s)</td>
<td>0.3669</td>
<td>0.0549</td>
<td>0.3112</td>
<td>0.0381</td>
<td>0.0595</td>
</tr>
<tr>
<td>Reaction time, off (s)(^1)</td>
<td>0.3019</td>
<td>0.0239</td>
<td>0.2990</td>
<td>0.0806</td>
<td>0.7778</td>
</tr>
<tr>
<td>Wisconsin Card Sorting Task(^1)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Categories completed</td>
<td>2.8</td>
<td>1.9</td>
<td>3.0</td>
<td>2.8</td>
<td>0.8945</td>
</tr>
<tr>
<td>Perservative errors</td>
<td>12.8</td>
<td>9.8</td>
<td>8.5</td>
<td>4.6</td>
<td>0.3639</td>
</tr>
<tr>
<td>Failure to maintain set</td>
<td>10.4</td>
<td>7.2</td>
<td>8.7</td>
<td>6.9</td>
<td>0.6940</td>
</tr>
<tr>
<td>% total correct responses</td>
<td>58.4</td>
<td>12.9</td>
<td>66.8</td>
<td>11.4</td>
<td>0.2836</td>
</tr>
<tr>
<td>Global score</td>
<td>100.0</td>
<td>19.2</td>
<td>84.0</td>
<td>47.5</td>
<td>0.5007</td>
</tr>
</tbody>
</table>

“On” and “off” indicate if subjects were taking their regular medication for PD (tested at separate study visits). Wisconsin Card Sorting Task (WCST) was not tested on medication to avoid a practice effect. \(P\text{-values at } \alpha \leq 0.05\) are denoted with an asterisk (*).\(^1\)\(n=5\) for non-regular cohort.
Figure 3.18 Reaction time per trial.
Trial 1 for non-regular exercisers (denoted as *) was significantly slower than all other trials (mean ± SEM: 0.395 ± 0.031 s trial 1 non-regular, 0.319 ± 0.032 s trials 2-4 non-regular and trials 1-4 habitual; p = < 0.018). P-values at α ≤ 0.05 are denoted with an asterisk (*).

3.6 Discussion
The habitual exerciser and non-regular exerciser cohorts were well matched for demographic- and disease-related characteristics except for their regular participation in exercise per week and aerobic capacity, which indicated their cohort allocation and subsequent comparison on outcome measures were appropriate. The primary findings from the study described in this chapter are minor, but interesting. First of all, habitual exercisers had increased RAC BP in the anterior putamen of their better (less affected) hemisphere following acute exercise. In comparison, non-regular exercisers had decreased RAC BP in their better anterior putamen, indicative of dopamine release. This finding in the anterior putamen was contrary to the hypothesis that habitual exercisers would release more dopamine in response to acute exercise. While decrease in RAC BP is an index of dopamine release, increase in RAC BP following an intervention does not indicate decreased dopamine release and the significance of this finding is not known. Therefore, the reason for increased RAC BP in response to acute exercise in habitual exercisers in the better anterior putamen observed in this study is not known.
An unexpected observation in this study worth discussing was a non-significant trend that habitual exercisers had increased mean RAC BP compared to non-regular exercisers across the ventral striatum, caudate and anterior putamen at both pre- and post-scans in both hemispheres. However, these differences were not significant given the variability in RAC BP values between subjects. With a larger sample size there could be a detectable difference in RAC BP between habitual exercisers and non-regular exercisers. An observation, again not reaching statistical significance, which coincides with the differences between sub-regions of the putamen is that the mean difference between cohorts in both RAC BP	extsubscript{pre} and in RAC BP	extsubscript{post} decreased moving caudally in the striatum. While the posterior putamen is more affected in PD than more rostral regions, it was somewhat surprising that we observed these differences between cohorts in the sub-regions of the putamen. Whether the effect in the better anterior putamen and the trend of increased RAC BP in more rostral striatal regions in habitual exercisers are indicative of potential neuroprotection or compensation could not be determined from this study.

As with the minor differences seen between cohorts in RAC BP in response to acute exercise, there were also some differences between cohorts in response to rewarding stimuli in the ventral striatum measured with fMRI. There were no effects between cohorts when baseline BOLD PSC was accounted for as a covariate and all probabilities were compared. However, when each probability of winning (i.e., level of uncertainty) was compared independently, habitual exercisers had a significantly greater response to reward compared to anticipation at 100%. This finding could indicate that habitual exercisers are more prone to respond to reward when reward is certain. As with the RAC BP findings, there was also an interesting non-significant trend in
activation of the ventral striatum between cohorts. It was observed that BOLD PSC in habitual exercisers was consistently higher than baseline BOLD PSC during both anticipation and reward phases at all probabilities of winning. BOLD PSC in non-regular exercisers was infrequently higher than baseline BOLD PSC, and notably only during reward at 50% and 75%. Although the difference between cohorts was not significant, this trend is interesting and could be elucidated with a larger sample size.

We found that habitual exercise is associated with some differences in motor and non-motor clinical function. Habitual exercise seems to be associated with better mood, less apathy, more positive affect and increased motor function on TUG (walking test) and on specific measures of hand speed (finger tapping, peg insertion). Differences were not found in the measures of cognitive function used in this study. The differences in clinical measures of motor and non-motor function complemented the neuroimaging outcomes. Importantly, these clinical findings further justify research on the mechanism of habitual exercise in PD. With the minor neuroimaging findings in this study, it cannot yet be determined how potential neurobiological changes from habitual exercise impact clinical symptoms.

3.6.1 Limitations

The primary limitation of this study was the small sample size. While subject characteristics were similar between cohorts, there were two women in the non-regular exerciser cohort and no women in the habitual exerciser cohort. Additionally, more subjects in the non-regular exerciser cohort were taking anti-depressants. These observed characteristic differences should be noted, although these factors do not explain the cohort differences in RAC BP or BOLD PSC. While
depression is common in PD and all subjects had controlled depression at the time of study participation, the difference in use of anti-depressants could explain the cohort differences in mood, affect and apathy. A statistical limitation of this study was that multiple comparisons were performed with this small sample size. Many of the p-values did not remain significant with Bonferroni correction. However, these findings were still important to demonstrate in order to highlight trends and areas of potential future research. Other limitations with this project include technical limitations that are inherent with neuroimaging, including PET ROI placement, use of an atlas-based ROI for the fMRI analysis, and steps to reduce noise in PET and fMRI data. This project and other recent PET and fMRI studies on exercise will help to determine the unique challenges with studying exercise in PD, which will help to mitigate analysis limitations for future groups. As much as possible, standardized techniques should be developed and implemented.

3.6.2 Future directions

There are many future directions that arise from this research project, including the use of animal models with basal ganglia lesions and the use of other radiotracers in humans to measure dopaminergic mechanisms that may be different between these two cohorts. Other radiotracers may help to understand how dopamine-driven differences between cohorts relate to the motor and non-motor clinical improvements associated with habitual exercise. RAC has a low affinity for D2/D3 and is more sensitive to dopamine release. Radiotracers with a high-affinity for D2/D3 include [18F]fallypride and [11C]FLB457. These tracers could help to image dopamine receptor density related to exercise. One study has already used these tracers with a small sample of subjects with early PD randomized to an exercise intervention (Fisher et al., 2013b). The effects
of habitual exercise have not yet been investigated with these radiotracers. Another useful tracer that could identify the integrity of striatal dopaminergic neurons is $^{11}$C-dihydrotetabenazine (DTBZ), which images vesicular monamine transporter 2 (Goswami et al., 2006).

Future research could also use the fMRI card task that was implemented in this study. That habitual exercisers more often exceeded baseline BOLD PSC and that habitual exercisers had a greater response to reward at 100% indicates this task may be used to study reward in the future. However, habitual exercisers responded to anticipation and reward at 0% probability of winning. This finding was unexpected and may indicate playing the task is stimulating on its own even without expecting a reward. The significance of the response at 0% needs to be investigated further. Additionally, most studies, as with this one, measure amplitude changes in BOLD signal, but BOLD signal can change in other ways. Other ways to analyze the BOLD signal include looking at its shape (i.e., time course) and its connectivity with other brain regions, which could demonstrate other meaningful information about mesolimbic circuitry. Subsequent studies could also consider adjusting the ventral striatum ROI as it was placed used a normalized human brain atlas. Other groups have recently developed techniques that can improve inter-subject comparison in fMRI by aligning the brain using functional connectivity instead of anatomical landmarks (Conroy et al., 2013). This alignment could improve comparisons across subjects. In regards to future directions for the clinical measurements to supplement neuroimaging outcomes in this study, further studies could increase the number and variety of cognitive outcomes. Behavioral cognitive measures are typically inexpensive to implement compared to neuroimaging outcomes. If differences between cohorts are found, it could direct future research
using neuroimaging to understand how exercise may impact other neurotransmitters, brain areas and overall functional connectivity of the brain.

3.6.3 Conclusions

Overall, the study described in this chapter identified that habitual exercise does not elicit more dopamine release compared to non-regular exercisers. Habitual exercise is associated with increased RAC BP in the anterior putamen, increased activation to reward in the ventral striatum, and improved measures of mood and motor function. It is plausible that the differences between cohorts may be dopaminergic in origin. While the sample size was small, habitual exercisers and non-regular exercisers with PD are not the same, and their disease-related differences are worth further exploration. The broad implications from this study are that habitual exercise could confound differences between subjects in other research on PD. Participation in exercise should be measured as a demographic characteristic in order to control for the potential effects of exercise in PD research.
Chapter 4: Conclusions

First, this thesis described the effects of cognition in PD based on a systematic review of the literature (Chapter 2). This systematic review identified that exercise can improve cognition, but the mechanisms in humans remain undetermined. Research with animal models with basal ganglia lesions has identified that dopaminergic mechanisms and neurotrophic factors likely drive the effects of exercise on cognition in PD. These findings align with the evidence for how exercise can impact motor outcomes in PD.

Next, this thesis described our study on how habitual exercise may benefit both motor and non-motor outcomes in PD using neuroimaging and clinical outcomes (Chapter 3). This study did not find differences in behavioral measures of cognition between the two cohorts, but there were some other interesting findings that suggest habitual exercise is beneficial for PD and may cause neurobiological changes. Acute exercise did not elicit dopamine release in either cohort, but habitual exercisers had the opposite effect (increased RAC BP) in their less affected anterior putamen. However, there may be a sustained effect of participating in regular exercise as we found a trend for increased RAC BP in habitual exercisers, especially in more rostral striatal regions, both before and following acute exercise. This trend supports previous findings from a small pilot PET study on human PD that 8 weeks of exercise can upregulate D<sub>2</sub> receptors in the basal ganglia (Fisher et al., 2013b). The trend from our study also supports another recent study by the same group that showed exercise can increase the density of spines and arborization of MSNs in mice (Toy et al., 2014). If participation in exercise can upregulate D<sub>2</sub> receptors after only a short intervention, then habitual exercisers may benefit from increased D<sub>2</sub> receptor density. D<sub>2</sub> receptors exist primarily on the post-synaptic membrane, but also serve as auto-
receptors on the pre-synaptic membrane. With more D₂ receptors, an individual could have increased sensitivity and regulation of both endogenous and exogenous dopamine (i.e., levodopa). Therefore, it is possible that habitual exercisers may not need as great a therapeutic dose of dopaminergic replacement therapy as a non-regular exerciser if they have an increased ability to bind dopamine in their basal ganglia. Interestingly, in this study habitual exercisers were prescribed less daily levodopa and general medications for PD, including dopamine agonists, although the difference between cohorts was not significant. The findings in BOLD PSC showed habitual exercisers had greater response to reward in the ventral striatum in comparison to baseline across probabilities of winning, and especially when reward was certain. These results suggest habitual exercise could improve neuronal, and specifically dopaminergic, integrity in the striatum.

In summary, this thesis provides more groundwork on an important topic in PD research. This project raised many more questions than it answered, but highlighted habitual exercise can cause subtle changes in the striatum, but the extent and significance of these differences remain to be determined with a larger sample size and complemented with other PET tracers. A promising future direction for this thesis work may be to compare the effects and mechanisms of exercise to pharmacological therapies to further understand how habitual exercise affects PD, how it can be therapeutic, and how it could complement current therapies.
Bibliography


Proceedings of the National Academy of Sciences of the United States of America
101:3316-3321.


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# Appendices

## Appendix A  PRISMA Checklist

<table>
<thead>
<tr>
<th>Section/topic</th>
<th>#</th>
<th>Checklist item</th>
<th>*Reported on page #</th>
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<tbody>
<tr>
<td><strong>TITLE</strong></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>Title</td>
<td>1</td>
<td>Identify the report as a systematic review, meta-analysis, or both.</td>
<td>21</td>
</tr>
<tr>
<td><strong>ABSTRACT</strong></td>
<td></td>
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</tr>
<tr>
<td>Structured summary</td>
<td>2</td>
<td>Provide a structured summary including, as applicable: background; objectives; data sources; study eligibility criteria, participants, and interventions; study appraisal and synthesis methods; results; limitations; conclusions and implications of key findings; systematic review registration number.</td>
<td>19</td>
</tr>
<tr>
<td><strong>INTRODUCTION</strong></td>
<td></td>
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<tr>
<td>Rationale</td>
<td>3</td>
<td>Describe the rationale for the review in the context of what is already known.</td>
<td>20</td>
</tr>
<tr>
<td>Objectives</td>
<td>4</td>
<td>Provide an explicit statement of questions being addressed with reference to participants, interventions, comparisons, outcomes, and study design (PICOS).</td>
<td>21</td>
</tr>
<tr>
<td><strong>METHODS</strong></td>
<td></td>
<td></td>
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<tr>
<td>Protocol and registration</td>
<td>5</td>
<td>Indicate if a review protocol exists, if and where it can be accessed (e.g., Web address), and, if available, provide registration information including registration number.</td>
<td>22</td>
</tr>
<tr>
<td>Eligibility criteria</td>
<td>6</td>
<td>Specify study characteristics (e.g., PICOS, length of follow-up) and report characteristics (e.g., years considered, language, publication status) used as criteria for eligibility, giving rationale.</td>
<td>22</td>
</tr>
<tr>
<td>Information sources</td>
<td>7</td>
<td>Describe all information sources (e.g., databases with dates of coverage, contact with study authors to identify additional studies) in the search and date last searched.</td>
<td>22</td>
</tr>
<tr>
<td>Search</td>
<td>8</td>
<td>Present full electronic search strategy for at least one database, including any limits used, such that it could be repeated.</td>
<td>22</td>
</tr>
<tr>
<td>Study selection</td>
<td>9</td>
<td>State the process for selecting studies (i.e., screening, eligibility, included in systematic review, and, if applicable, included in the meta-analysis).</td>
<td>22</td>
</tr>
<tr>
<td>Data collection process</td>
<td>10</td>
<td>Describe method of data extraction from reports (e.g., piloted forms, independently, in duplicate) and any processes for obtaining and confirming data from investigators.</td>
<td>22</td>
</tr>
<tr>
<td>Data items</td>
<td>11</td>
<td>List and define all variables for which data were sought (e.g., PICOS, funding sources) and any assumptions and simplifications made.</td>
<td>22</td>
</tr>
<tr>
<td>Risk of bias in individual studies</td>
<td>12</td>
<td>Describe methods used for assessing risk of bias of individual studies (including specification of whether this was done at the study or outcome level), and how this information is to be used in any data synthesis.</td>
<td>22</td>
</tr>
<tr>
<td>Summary measures</td>
<td>13</td>
<td>State the principal summary measures (e.g., risk ratio, difference in means).</td>
<td>22</td>
</tr>
<tr>
<td>Section/topic</td>
<td>#</td>
<td>Checklist item</td>
<td>Reported on page #</td>
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<tr>
<td>Synthesis of results</td>
<td>14</td>
<td>Describe the methods of handling data and combining results of studies, if done, including measures of consistency (e.g., $I^2$) for each meta-analysis.</td>
<td>22</td>
</tr>
<tr>
<td>Risk of bias across</td>
<td>15</td>
<td>Specify any assessment of risk of bias that may affect the cumulative evidence (e.g., publication bias, selective reporting within studies).</td>
<td>28, 37</td>
</tr>
<tr>
<td>studies</td>
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<tr>
<td>Additional analyses</td>
<td>16</td>
<td>Describe methods of additional analyses (e.g., sensitivity or subgroup analyses, meta-regression), if done, indicating which were pre-specified.</td>
<td>22</td>
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<tr>
<td>RESULTS</td>
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<tr>
<td>Study selection</td>
<td>17</td>
<td>Give numbers of studies screened, assessed for eligibility, and included in the review, with reasons for exclusions at each stage, ideally with a flow diagram.</td>
<td>23</td>
</tr>
<tr>
<td>Study characteristics</td>
<td>18</td>
<td>For each study, present characteristics for which data were extracted (e.g., study size, PICOS, follow-up period) and provide the citations.</td>
<td>27, 35</td>
</tr>
<tr>
<td>Risk of bias</td>
<td>19</td>
<td>Present data on risk of bias of each study and, if available, any outcome level assessment (see item 12).</td>
<td>28, 37</td>
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<tr>
<td>within studies</td>
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<tr>
<td>Results of individual</td>
<td>20</td>
<td>For all outcomes considered (benefits or harms), present, for each study: (a) simple summary data for each intervention group (b) effect estimates and confidence intervals, ideally with a forest plot.</td>
<td>24</td>
</tr>
<tr>
<td>studies</td>
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</tr>
<tr>
<td>Synthesis of results</td>
<td>21</td>
<td>Present results of each meta-analysis done, including confidence intervals and measures of consistency.</td>
<td>NA</td>
</tr>
<tr>
<td>Risk of bias</td>
<td>22</td>
<td>Present results of any assessment of risk of bias across studies (see Item 15).</td>
<td>NA</td>
</tr>
<tr>
<td>across studies</td>
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<tr>
<td>Additional analysis</td>
<td>23</td>
<td>Give results of additional analyses, if done (e.g., sensitivity or subgroup analyses, meta-regression [see Item 16]).</td>
<td>NA</td>
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<tr>
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<tr>
<td>DISCUSSION</td>
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<tr>
<td>Summary of evidence</td>
<td>24</td>
<td>Summarize the main findings including the strength of evidence for each main outcome; consider their relevance to key groups (e.g., healthcare providers, users, and policy makers).</td>
<td>39</td>
</tr>
<tr>
<td>Limitations</td>
<td>25</td>
<td>Discuss limitations at study and outcome level (e.g., risk of bias), and at review-level (e.g., incomplete retrieval of identified research, reporting bias).</td>
<td>44</td>
</tr>
<tr>
<td>Conclusion(s)</td>
<td>26</td>
<td>Provide a general interpretation of the results in the context of other evidence, and implications for future research.</td>
<td>44</td>
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<tr>
<td>FUNDING</td>
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<tr>
<td>Funding</td>
<td>27</td>
<td>Describe sources of funding for the systematic review and other support (e.g., supply of data); role of funders for the systematic review.</td>
<td>iv</td>
</tr>
</tbody>
</table>


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Appendix B  Methodology on information sources, study search and collection

The following electronic databases were searched for articles: PubMed, Web of Knowledge, and EBM Reviews (OvidSP). The primary search parameter for each database used the keywords, “Parkinson’s disease” AND “exercise” AND “cognition” with the Boolean operator “AND.” Additional keywords related to cognition were also searched by replacing “cognition” in the primary keyword phrase one at a time with each of “dementia,” “Alzheimer,” “memory,” “executive function,” and “impulse.” Additional studies were identified by ancestry searches of the articles yielded from the electronic search. No limits were provided for any of the database searches. Study author DM performed all aspects of the search, screen and identification of eligible studies (Figure 2.1). All studies identified through the information sources were compiled on the citation manager, EndNote. The titles of each study were then screened to identify PD participants, including human subjects or PD-like animal models. The abstracts of each remaining study were then searched manually for the eligibility criteria. The full text was searched for articles deemed to meet the eligibility criteria based on their title and abstract. All data were extracted by the study author DM in their existing form from the articles. The collected data and risk of bias (Higgins et al., 2011b) was used to assess the clinical studies for their quality of the evidence and determine the strength of the recommendations (Table 2.3) based on the Grades of Recommendations, Assessment, Development, and Evaluation (GRADE) ranking system (Higgins et al., 2011a). The criteria used to determine the quality of the evidence were based on the methodological quality of the evidence, the possibility of bias and transferability of the findings. ‘Moderate’ evidence was reserved for randomized trials where reasonable control groups were used, and ‘low’ evidence designated studies that were not randomized and thereby carried significant risk of bias. The criteria used to determine the strength of the recommendations were based on balancing the benefits and downsides of the findings, including consideration of the quality of the evidence. ‘Strong’ recommendations were for studies that had at least ‘moderate’ evidence quality and where we were confident that the likelihood of benefit from the intervention outweighed the likelihood of undesirable effects or no effect (as exercise is a reasonably benign intervention). The clinical studies were assigned the same GRADE rankings by two independent raters (DM and MS).
Appendix C  Exercise level screening tool

<table>
<thead>
<tr>
<th>Examples of exercises</th>
<th>Mild (&lt;3 MET)</th>
<th>Moderate (3-6 MET)</th>
<th>Vigorous (&gt;6 MET)</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Light walking, yoga, gardening, stretching, balance exercises</td>
<td>Swimming, hiking, speed walking, rowing, dancing, jogging, cross-country skiing, bicycling</td>
<td>Running, hockey, mountain climbing, aerobics, jump rope</td>
<td>Combined participation and length in all types of exercise per week*</td>
<td></td>
</tr>
<tr>
<td>Participation in Exercise (counts): Number of times per week, exercise duration must be 20 minutes or greater for one point/count</td>
<td>_____/3</td>
<td>_____x2</td>
<td>_____x3</td>
<td>_____</td>
</tr>
<tr>
<td>Duration of exercise (minutes): Duration of exercise activities recorded in minutes by times per week</td>
<td>_____/4</td>
<td>_____</td>
<td>_____</td>
<td>_____</td>
</tr>
</tbody>
</table>

*Weekly estimates are based on exercise habits over at least the previous 6 months.

To qualify as a habitual exerciser, must meet both requirements:
1) Participation in exercise per week: Total score (counts) per week must be ≥ 6.
2) Duration of exercise per week: Weekly duration in participation in activities > 3 MET only must be > 120 min.

To qualify as a non-regular exerciser, must meet both requirements:
1) Participation in exercise per week: Total score (counts) per week must be < 4.
2) Duration of exercise per week: Weekly duration in participation in activities > 3 MET only must be < 120 min.

This exercise screening tool was created by our group. The scoring scheme for this questionnaire was based on evidence that moderate exercise (3-6 metabolic equivalents, METs) requires approximately twice as much energy as mild activities (1-2 METs) and vigorous exercise (>6 METs) requires about three times the energy of a mild exercise (Jette et al., 1990; Ainsworth et al., 2000).
Appendix D  Physical Activity Readiness Questionnaire

Subject ID:  Date:  Time:

PAR-Q & YOU
(A Questionnaire for People Aged 15 to 69)

Regular physical activity is fun and healthy, and increasingly more people are starting to become more active every day. Being more active is very safe for most people. However, some people should check with their doctor before they start becoming much more physically active.

If you are planning to become much more physically active than you are now, start by answering the seven questions in the box below. If you are between the ages of 15 and 69, the PAR-Q will tell you if you should check with your doctor before you start. If you are over 69 years of age, and you are not used to being very active, check with your doctor.

Common sense is your best guide when you answer these questions. Please read the questions carefully and answer each one honestly: check YES or NO.

<table>
<thead>
<tr>
<th>YES</th>
<th>NO</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Has your doctor ever said that you have a heart condition and that you should only do physical activity recommended by a doctor?</td>
<td></td>
</tr>
<tr>
<td>2. Do you feel pain in your chest when you do physical activity?</td>
<td></td>
</tr>
<tr>
<td>3. In the past month, have you had chest pain when you were not doing physical activity?</td>
<td></td>
</tr>
<tr>
<td>4. Do you lose your balance because of dizziness or do you ever lose consciousness?</td>
<td></td>
</tr>
<tr>
<td>5. Do you have a bone or joint problem (for example, back, knee or hip) that could be made worse by a change in your physical activity?</td>
<td></td>
</tr>
<tr>
<td>6. Is your doctor currently prescribing drugs (for example, water pills) for your blood pressure or heart condition?</td>
<td></td>
</tr>
<tr>
<td>7. Do you know of any other reason why you should not do physical activity?</td>
<td></td>
</tr>
</tbody>
</table>

If you answered YES to one or more questions

Talk with your doctor by phone or in person BEFORE you start becoming much more physically active or BEFORE you have a fitness appraisal. Tell your doctor about the PAR-Q and which questions you answered YES.

- You may be able to do any activity you want — as long as you start slowly and build up gradually. Or, you may need to restrict your activities to those which are safe for you. Talk with your doctor about the kinds of activities you wish to participate in and follow his/her advice.
- Find out which community programs are safe and helpful for you.

If you answered NO to all questions

- DELAY BECOMING MUCH MORE ACTIVE:
  - If you are not feeling well because of a temporary illness such as a cold or a fever — wait until you feel better; or
  - If you are or may be pregnant — talk to your doctor before you start becoming more active.

PLEASE NOTE: If your health changes so that you then answer YES to any of the above questions, tell your fitness or health professional. Ask whether you should change your physical activity plan.

No changes permitted. You are encouraged to photocopy the PAR-Q but only if you use the entire form.

Note: This physical activity clearance is valid for a maximum of 12 months from the date it is completed and becomes invalid if your condition changes so that you would answer YES to any of the seven questions.
**Get Active Your Way, Every Day—for Life!**

Physical activity does not have to be hard. Build physical activity into your daily routine.

- **Start with a 10 minute walk—gradually increase the time.**
- **Find out about walking and running programs in your community.**
- **Observe a physical activity class to see if you want to try it.**
- **Try one activity at a time—you don’t have to have a make a long-term commitment.**
- **Do the activities you enjoy, more often.**

**Benefit of regular activity: Health risks of inactivity:**

- Better heart health
- Improved brain health
- Better quality of sleep
- Better mental health
- Stronger muscles and bones
- Lower blood pressure
- Weight control
- Reduced risk of type 2 diabetes
- Reduced risk of some types of cancer
- Better mental health
- Reduced risk of depression
- Improved mood
- Higher self-esteem
- Increased confidence
- Increased energy

**You Can Do It—Getting Started Is Easier Than You Think**

Physical activity does not have to be hard. Build physical activity into your daily routine.

- **Start with a 10 minute walk—gradually increase the time.**
- **Find out about walking and running programs in your community.**
- **Observe a physical activity class to see if you want to try it.**
- **Try one activity at a time—you don’t have to have a make a long-term commitment.**
- **Do the activities you enjoy, more often.**

**Benefit of regular activity: Health risks of inactivity:**

- Better heart health
- Improved brain health
- Better quality of sleep
- Better mental health
- Stronger muscles and bones
- Lower blood pressure
- Weight control
- Reduced risk of type 2 diabetes
- Reduced risk of some types of cancer
- Better mental health
- Reduced risk of depression
- Improved mood
- Higher self-esteem
- Increased confidence
- Increased energy

**Conclusion:**

Physical activity is important for everyone. It helps to keep us healthy and improves our quality of life. Whether you work out or not, physical activity is something that we should all try to include in our daily routines.

**References:**


For more information, please contact the Canadian Society for Exercise Physiology

202-185 Somerset Street
Otawa, ON K2P 0G2
Tel: 1-877-451-3755 • FAX (613) 234-3565
Online: www.cscep.ca

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Supported by

Health Canada

Santé Canada

The original PAR-Q was developed by the British Columbia Ministry of Health. It has been revised by an Expert Advisory Committee of the Canadian Society for Exercise Physiology chaired by Dr. N. Gereau (2002).

Disponible en français sous le titre «Questionnaire sur l’aptitude à l’activité physique - Q-AP (rev. 2002)».


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Appendix E  Example of computer displays during the card task during fMRI

1. Subjects are shown the winning cards out of the four cards. Anticipation period (10 s duration).

2. The cards are shuffled. Anticipation period (10 s duration).

3. The cards are shuffled. Anticipation period (10 s duration).

4. The subject has 5 seconds to pick a card by pressing a button on the hand-held controller. Anticipation period (10 s duration).

5. The subject has 5 seconds to pick a card by pressing a button on the hand-held controller. Anticipation period (10 s duration).

6. The subject is shown whether they picked a winning card or they did not. Happy or sad sounds accompany the happy or sad face. A coin is then added to the winnings barometer on the right. Reward period (10 s duration).
Appendix F  MDS-UPDRS III

Subject ID:    Time:
Date:      Session:

Is the patient on medication for treating the symptoms of PD?  No   Yes

If the patient is receiving medication for treating the symptoms of PD, mark the patient’s clinical state using the following definitions:

On: On is the typical functional state when patients are receiving medication and have a good response

OFF: Off is the typical functional state when patients have a poor response in spite of taking medications

Is the patient on levodopa?  No   Yes
If yes, minutes since last levodopa dose: _________________________

<table>
<thead>
<tr>
<th>Speech</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>No speech problems</td>
</tr>
<tr>
<td>1</td>
<td>Loss if modulation, diction or volume, but still all words are easy to understand</td>
</tr>
<tr>
<td>2</td>
<td>Loss of modulation, diction, volume with a few words unclear, but the overall sentences easy to follow</td>
</tr>
<tr>
<td>3</td>
<td>Speech is difficult to understand to the point that some, but not most, sentences are poorly understood</td>
</tr>
<tr>
<td>4</td>
<td>Most speech is difficult to understand or unintelligible</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Facial Expression</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>Normal facial expression</td>
</tr>
<tr>
<td>1</td>
<td>Minimal masked facies manifested only by decreased frequency of blinking</td>
</tr>
<tr>
<td>2</td>
<td>In addition to decreased eye-blink frequency, masked facies present in the lower face as well, namely fewer movements around the mouth, such as less spontaneous smiling, but lips not parted</td>
</tr>
<tr>
<td>3</td>
<td>Masked facies with lips parted some of the time when the mouth is at rest</td>
</tr>
<tr>
<td>4</td>
<td>Masked facies with lips parted most of the time when the mouth is at rest</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Postural Tremor Of Hands</th>
<th>R</th>
<th>L</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>Notremor</td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>Tremor is present but less than 1cm in amplitude</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>Tremor is at least 1 but less than 3cm in amplitude</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>Tremor is at least 3 but less than 10cm in amplitude</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>Tremor at least 10cm in amplitude</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Kinetic Tremor Of Hands</th>
<th>R</th>
<th>L</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>Notremor</td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>Tremor is present but less than 1cm in amplitude</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>Tremor is at least 1 but less than 3cm in amplitude</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>Tremor is at least 3 but less than 10cm in amplitude</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>Tremor at least 10cm in amplitude</td>
<td></td>
</tr>
</tbody>
</table>
### Rest Tremor Amplitude

<table>
<thead>
<tr>
<th>Extremity ratings</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>RUE</td>
<td>No tremor</td>
</tr>
<tr>
<td>1</td>
<td>RUE</td>
<td>&lt; 1 cm in maximal amplitude</td>
</tr>
<tr>
<td>2</td>
<td>RUE</td>
<td>&gt; 1 cm but &lt; 3 cm in maximal amplitude</td>
</tr>
<tr>
<td>3</td>
<td>RUE</td>
<td>&gt; 3 - 10 cm in maximal amplitude</td>
</tr>
<tr>
<td>4</td>
<td>RUE</td>
<td>&gt; 10 cm in maximal amplitude</td>
</tr>
</tbody>
</table>

### Lip and Jaw Ratings:

| 0                  | RLE | No tremor |
| 1                  | RLE | < 1 cm in maximal amplitude |
| 2                  | RLE | > 1 cm but < 2 cm in maximal amplitude |
| 3                  | RLE | > 2 cm but < 3 cm in maximal amplitude |
| 4                  | RLE | > 3 cm in maximal amplitude |

### Hand Movements

| 0                  | RLE | No problems |
| 1                  | RLE | Any of the following: a) the regular rhythm is broken with one or two interruptions or hesitations of the movement; b) slight slowing; c) the amplitude decrements near the end of the task |
| 2                  | RLE | a) 3 to 5 interruptions during the movements; b) mild slowing; c) the amplitude decrements midway in the task |
| 3                  | RLE | a) more than 5 interruptions during the movement or at least one longer arrest in ongoing movement; b) moderate slowing; c) the amplitude decrements starting after the 1st open-and close-sequence |
| 4                  | RLE | Cannot or can only barely perform the task because of slowing, interruptions or decrements |

### Pronation-Supination Movements of Hands

| 0                  | RLE | No problems |
| 1                  | RLE | Any of the following: a) regular rhythm is broken with one or two interruptions or hesitations of the movements b) slight slowing; c) the amplitude decrements near the end of the sequence |
| 2                  | RLE | a) 3 to 5 interruptions during the movements; b) mild slowing; c) the amplitude decrements midway in the task |
| 3                  | RLE | a) more than 5 interruptions during the movement or at least one longer arrest in ongoing movement; b) moderate slowing; c) the amplitude decrements starting after the 1st supination-pronation sequence |
| 4                  | RLE | Cannot or can only barely perform the task because of slowing, interruptions or decrements |

### Constancy of Rest Tremor

| 0                  | RLE | No tremor |
| 1                  | RLE | Tremor at rest is present <25% of the entire examination period |
| 2                  | RLE | Tremor at rest is present 26-50% of the entire examination period |
| 3                  | RLE | Tremor at rest is present 51-75% of the entire examination period |
| 4                  | RLE | Tremor at rest is present > 75% of the entire examination period |

### Finger Tapping

| 0                  | RLE | No problems |
| 1                  | RLE | Any of the following: a) the regular rhythm is broken with one or two interruptions or hesitations; b) slight slowing; c) the amplitude decrements near the end of the the 10 taps |
| 2                  | RLE | a) 3 to 5 interruptions during tapping; b) mild slowing; c) the amplitude decrements midway in the 10-tap sequence |
| 3                  | RLE | a) more than 5 interruptions during tapping or at least one longer arrest in ongoing movement; b) moderate slowing; c) the amplitude decrements starting after 1st tap |
| 4                  | RLE | Cannot or can only barely perform the task |

---

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### Toe Tapping

<table>
<thead>
<tr>
<th>Score</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>No problems</td>
</tr>
<tr>
<td>1</td>
<td>Any of the following: a) 3 to 5 interruptions during the movements; b) slight slowing; c) the amplitude decrements near the end of the sequence</td>
</tr>
<tr>
<td>2</td>
<td>a) 3 to 5 interruptions during the movements; b) mild slowing; c) the amplitude decrements midway in the task</td>
</tr>
<tr>
<td>3</td>
<td>a) more than 5 interruptions during the movement or at least one longer arrest in ongoing movement; b) moderate slowing; c) the amplitude decrements starting after the 1st tap</td>
</tr>
<tr>
<td>4</td>
<td>Cannot or can only barely perform the task because of slowing, interruptions or decrements</td>
</tr>
</tbody>
</table>

### Leg Agility

<table>
<thead>
<tr>
<th>Score</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>No problems</td>
</tr>
<tr>
<td>1</td>
<td>Any of the following: a) regular rhythm is broken with one or two interruptions or hesitations of the movements; b) slight slowing; c) the amplitude decrements near the end of the sequence</td>
</tr>
<tr>
<td>2</td>
<td>Any of the following: a) 3 to 5 interruptions during the movements; b) mild slowing; c) the amplitude decrements midway in the task</td>
</tr>
<tr>
<td>3</td>
<td>Any of the following: a) more than 5 interruptions during the movement or at least one longer arrest in ongoing movement; b) moderate slowing; c) the amplitude decrements starting after the 1st tap</td>
</tr>
<tr>
<td>4</td>
<td>Cannot or can only barely perform the task because of slowing, interruptions or decrements</td>
</tr>
</tbody>
</table>

### Arising From Chair

<table>
<thead>
<tr>
<th>Score</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>No problems</td>
</tr>
<tr>
<td>1</td>
<td>Slow; or may need more than one attempt; or may need to move forward in the chair to arise. No need to use the arms of the chair</td>
</tr>
<tr>
<td>2</td>
<td>Pushes self up from arms of seat without difficulty</td>
</tr>
<tr>
<td>3</td>
<td>Needs to push off, but tends to fall back and may have to try more than one time, but can get up without help</td>
</tr>
<tr>
<td>4</td>
<td>Unable to arise without help</td>
</tr>
</tbody>
</table>

### Posture

<table>
<thead>
<tr>
<th>Score</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>No problems:</td>
</tr>
<tr>
<td>1</td>
<td>Not quite erect, but posture could be normal for older person</td>
</tr>
<tr>
<td>2</td>
<td>Define flexion, scoliosis or leaning to one side, but patient can correct posture to normal posture when asked to do so</td>
</tr>
<tr>
<td>3</td>
<td>Stooped posture, scoliosis or leaning to one side that cannot be corrected volitionally to a normal posture by the patient</td>
</tr>
<tr>
<td>4</td>
<td>Flexion, scoliosis or leaning with extreme abnormally of posture</td>
</tr>
</tbody>
</table>

### Freezing of Gait

<table>
<thead>
<tr>
<th>Score</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>No freezing</td>
</tr>
<tr>
<td>1</td>
<td>Freezes on starting, turning or walking through doorway with a single halt during any of theses events, but then continues smoothly without freezing during straight walking</td>
</tr>
<tr>
<td>2</td>
<td>Freezes on starting, turning or walking through doorway with more than one halt during any of theses events, but then continues smoothly without freezing during straight walking</td>
</tr>
<tr>
<td>3</td>
<td>Freezes once during straight walking</td>
</tr>
<tr>
<td>4</td>
<td>Freezes multiple times during straight walking</td>
</tr>
</tbody>
</table>

### Gait

<table>
<thead>
<tr>
<th>Score</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>No problems:</td>
</tr>
<tr>
<td>1</td>
<td>Independent walking with minor gait impairment</td>
</tr>
<tr>
<td>2</td>
<td>Independent walking but with substantial gait impairment</td>
</tr>
<tr>
<td>3</td>
<td>Requires an assistance device for safe walking but not a person</td>
</tr>
<tr>
<td>4</td>
<td>Cannot walk at all or only with another person’s assistance</td>
</tr>
</tbody>
</table>

### Postural Stability

<table>
<thead>
<tr>
<th>Score</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>No problems: Recovers with one or two steps</td>
</tr>
<tr>
<td>1</td>
<td>3-5 steps, but subject recovers unaided</td>
</tr>
<tr>
<td>2</td>
<td>More than 5 steps, but subject recovers unaided</td>
</tr>
<tr>
<td>3</td>
<td>Stands safely, but with absence of postural response; falls if not caught by examiner</td>
</tr>
<tr>
<td>4</td>
<td>Very unstable, tends to lose balance spontaneously or with just a gentle pull on the shoulders</td>
</tr>
</tbody>
</table>
Global Spontaneity of Movement (Body Bradykinesia)

0 = No problems:
1 = Slight global slowness and poverty of spontaneous movements
2 = Mild global slowness and poverty of spontaneous movements
3 = Moderate global slowness and poverty of spontaneous movements
4 = Severe global slowness and poverty of spontaneous movements

Rigidity

Judged on slow passive movement of major joints with patient in sitting position

0 = Absent
1 = Slight, only detected with activation maneuver
2 = Rigidity detected without the activation maneuver, but full range of motion is easily achieved
3 = Rigidity detected without the activation maneuver; full range of motion is achieved with effort
4 = Severe, full range of motion not achieved

Neck

RIE LUE
RLE LLE

DYSKINESIA IMPACT ON PART III RATINGS

A. Were dyskinesias (chorea or dystonia) present during examination? No Yes

B. If yes, did these movements interfere with your ratings? No Yes

Hoehn and Yahr Stage

0 = Asymptomatic
1 = Unilateral involvement only
2 = Bilateral involvement without impairment of balance
3 = Mild to moderate involvement; some postural instability but physically independent; needs
4 = Severe disability; still able to walk or stand unassisted
5 = Wheelchair bound or bedridden unless aided.
Appendix G  Beck Depression Inventory - II

Subject ID: ________________________________  Date/Time: ____________________

Instructions: This questionnaire consists of 21 groups of statements. Please read each group of
statements carefully, and then pick out the one statement in each group that best describes the
way you have been feeling during the past two weeks, including today. Circle the number beside
the statement you have picked. If several statements in the group seem to apply equally well,
circle the highest number for that group. Be sure that you do not choose more than one statement
for any group.

1. **Sadness**
   0  I do not feel sad.
   1  I feel sad much of the time.
   2  I am sad all the time.
   3  I am so sad or unhappy that I can’t stand it.

2. **Pessimism**
   0  I am not discouraged about my future.
   1  I feel more discouraged about my future than I used to be.
   2  I do not expect things to work out for me.
   3  I feel my future is hopeless and will only get worse.

3. **Past Failure**
   0  I do not feel like a failure.
   1  I have failed more than I should have.
   2  As I look back, I see a lot of failures.
   3  I feel I am a total failure as a person.
4. **Loss of pleasure**
0 I get as much pleasure as I ever did from the things I enjoy.
1 I don’t enjoy things as much as I used to.
2 I get very little pleasure from the things I used to enjoy.
3 I can’t get any pleasure from the things I used to enjoy.

5. **Guilty Feelings**
0 I don’t feel particularly guilty.
1 I don’t feel guilty over many things I have done or should have done.
2 I feel quite guilty most of the time.
3 I feel guilty all of the time.

6. **Punishment Feelings**
0 I don’t feel I am being punished.
1 I feel I may be punished.
2 I expect to be punished.
3 I feel I am being punished.

7. **Self-Dislike**
0 I feel the same about myself as ever.
1 I have lost confidence in myself.
2 I am disappointed in myself.
3 I dislike myself.

8. **Self-Criticalness**
0 I don’t criticize or blame myself more than usual.
1 I am more critical of myself than I used to be.
2 I criticize myself for all my faults.
3 I blame myself for everything that happens.

9. **Suicidal Thoughts or Wishes**
0 I don’t have any thoughts of killing myself.
1 I have thoughts of killing myself, but I would not carry them out.
2 I would like to kill myself.
3 I would kill myself if I had the chance.

10. **Crying**
0 I don’t cry anymore than I used to.
1 I cry more than I used to.
2. I cry over every little thing.
3. I feel like crying, but I can’t.

11. Agitation
0. I am not more restless or wound up than usual.
1. I feel more restless or wound up than usual.
2. I am so restless or agitated that it’s hard to stay still.
3. I am so restless or agitated that I have to keep moving or doing something.

12. Loss of Interest
0. I have not lost interest in other people or activities.
1. I am less interested in other people or things than before.
2. I have lost most of my interest in other people or things.
3. It’s hard to get interested in anything.

13. Indecisiveness
0. I make decisions as well as ever.
1. I find it more difficult to make decisions than usual.
2. I have much greater difficulty in making decisions than I used to.
3. I have trouble making any decisions.

14. Worthlessness
0. I do not feel I am worthless.
1. I don’t consider myself as worthwhile and useful as I used to do.
2. I feel more worthless as compared to other people.
3. I feel utterly worthless.

15. Loss of Energy
0. I have as much energy as ever.
1. I have less energy than I used to have.
2. I don’t have enough energy to do very much.
3. I don’t have enough energy to do anything.

16. Changes in Sleeping Pattern
0. I have not experienced any change in my sleeping pattern.
1. I sleep somewhat more/less than usual.
2. I sleep a lot more/less than usual.
3. I sleep most of the day. OR I wake up 1-2 hours early and can’t get back to sleep.
17. **Irritability**
0  I am no more irritable than usual.
1  I am more irritable than usual.
2  I am much more irritable than usual.
3  I am irritable all the time.

18. **Changes in Appetite**
0  I have not experienced any change in my appetite.
1  My appetite is somewhat less/greater than usual.
2  My appetite is much less/greater than usual.
3  I have no appetite at all.  OR  I crave food all the time.

19. **Concentration Difficulty**
0  I can concentrate as well as ever.
1  I can’t concentrate as well as usual.
2  It’s hard to keep my mind on anything for very long.
3  I find I can’t concentrate on anything.

20. **Tiredness or Fatigue**
0  I am no more tired or fatigued than usual.
1  I get more tired or fatigued more easily than usual.
2  I am too tired or fatigued to do a lot of the things I used to do.
3  I am too tired or fatigued to do most of the things I used to do.

21. **Loss of Interest in Sex**
0  I have not noticed any recent change in my interest in sex.
1  I am less interested in sex than I used to be.
2  I am much less interested in sex now.
3  I have lost interest in sex completely.
Appendix H  Montreal Cognitive Assessment

**MONTREAL COGNITIVE ASSESSMENT (MOCA)**
Version 7.1 Original Version

**VISUOSPATIAL / EXECUTIVE**
- Copy cube
- Draw C At Ten past eleven (3 points)

**NAMING**
- Copy cube
- Draw C At Ten past eleven (3 points)

**MEMORY**
- Read list of words; subject must repeat them. Do 2 trials, even if 1st trial is successful. Do a recall after 5 minutes.

**ATTENTION**
- Read list of digits (1 digit/sec.). Subject has to repeat them in the forward order.
- Subject has to repeat them in the backward order.

**LANGUAGE**
- Repeat: I only know that John is the one to help today.
- Fluency: Name maximum number of words in one minute that begin with the letter F.

**ABSTRACTION**
- Similarity between e.g. banana - orange = fruit
- train - bicycle = watch - ruler

**DELAYED RECALL**
- Has to recall words with no cue

**OPTIONAL**
- Category cue
- Multiple choice cue

**ORIENTATION**
- Date
- Month
- Year
- Day
- Place
- City

© Z. Nasreddine MD www.mocatest.org Normal ≥ 26 / 30
ADMINISTERED BY ____________________________ Points for UNCUED recall only

TOTAL ______ / 30  

ADD 1 POINT IF ≤ 12 yr edu
Appendix I  Starkstein Apathy Scale

Starkstein Apathy Scale.

Subject ID: __________________________ Date/Time: __________________________

Scoring:

For questions 1-8,
not at all = 3 points;
slightly = 2;
some = 1; alot = 0.

For questions 9 - 14,
not at all = 0;
slightly = 1;
some = 2;
alot = 3.

1. Are you interested in learning new things?
   not at all slightly some a lot
2. Does anything interest you?
   not at all slightly some a lot
3. Are you concerned about your condition?
   not at all slightly some a lot
4. Do you put much effort into things?
   not at all slightly some a lot
5. Are you always looking for something to do?
   not at all slightly some a lot
6. Do you have plans and goals for the future?
   not at all slightly some a lot
7. Do you have motivation?
   not at all slightly some a lot
8. Do you have the energy for daily activities?
   not at all slightly some a lot
9. Does someone have to tell you what to do each day?
   not at all slightly some a lot
10. Are you indifferent to things?
    not at all slightly some a lot
11. Are you unconcerned with many things?
    not at all slightly some a lot
12. Do you need a push to get started on things?
    not at all slightly some a lot
13. Are you neither happy nor sad, just in between?
    not at all slightly some a lot
14. Would you consider yourself apathetic?
    not at all slightly some a lot
Appendix J  Trail-Making Test A

Session ____________________ Trail Making Test Part A

Subject ID: ______________________  Date/Time:___________________________
Appendix K  Trail-Making Test B

Subject ID: ____________________________  Date/Time:___________________________

Session ________________Trail Making Test Part B
Appendix L  Positive and Negative Affect Schedule

The Positive and Negative Affect Schedule (PANAS):

PANAS Questionnaire
This scale consists of a number of words that describe different feelings and emotions. Read each item and then list the number from the scale below next to each word. Indicate to what extent you feel this way right now, that is, at the present moment.

<table>
<thead>
<tr>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
</tr>
</thead>
<tbody>
<tr>
<td>Very Slightly or Not at All</td>
<td>A Little</td>
<td>Moderately</td>
<td>Quite a Bit</td>
<td>Extremely</td>
</tr>
</tbody>
</table>

| 1. Proud | 11. Afraid |
| 2. Enthusiastic | 12. Active |
| 5. Guilty | 15. Determined |
| 7. Upset | 17. Inspired |
| 8. Excited | 18. Ashamed |
| 10. Interested | 20. Irritable |