IMPLICIT SEQUENCE LEARNING IN PEOPLE WITH PARKINSON’S DISEASE

A Dissertation submitted to the Faculty of the Graduate School of Arts and Sciences of Georgetown University in partial fulfillment of the requirement for the degree of Doctor of Philosophy in Psychology

By

Katherine Gamble, M.S.

Washington, DC

January 15th, 2014
IMPLICIT SEQUENCE LEARNING IN PEOPLE WITH PARKINSON’S DISEASE

Katherine R. Gamble, M.S.

Thesis Adviser: Darlene V. Howard, Ph.D.

ABSTRACT

People with Parkinson’s disease (PD) have known deficits in striatal dopamine, and compared to age-matched healthy older adults, increased striatal deficits, and difficulties with maintaining postural stability and complex language comprehension, behaviors that involve sequencing. Because sequencing abilities are important for daily life and for various types of rehabilitation, this decline with PD should be well understood, yet the literature on the effects of PD in sequence learning is mixed. Some of the confusion in the literature may be due to variation in participant characteristics, but also to task differences, such as the extent of motor movement required. The present studies examined implicit sequence learning using the Triplets Learning Task (TLT) in a PD group. There were 29 medicated PD participants, and 30 healthy older adults were recruited as a control group. Study 1 compared learning over several blocks of training in the TLT. Two learning measures indicated that people with PD learned, but they showed less implicit sequence learning than Controls. This group difference in one analysis was carried largely by late training, when learning is thought to be particularly dependent on the striatum. Additionally, when first and second halves of blocks were examined separately, the PD group showed less learning than Controls in the second halves of blocks, when learning is thought to be striatal-dependent. In Study 2, we examined relationships between learning and PD-related characteristics. Using learning in the second half block, we found that in our PD
group, learning positively correlated with Age, Disease duration and Levodopa levels. These three variables were positively correlated with one another, and therefore it was difficult to disentangle their individual effects. Together, these studies suggest that people with PD were able to learn implicit sequences, though to a lesser degree than Controls, and Levodopa medication may have helped moderate this sequencing deficit. Findings of group differences in learning in sections of training suggest that particular points in training may be more sensitive to sequencing or striatal deficits. Thus, the TLT and these learning analyses could be used to better characterize clinical populations that may have hippocampal or striatal deficits.
I would like to say a huge thank you to my advisor, Darlene Howard. There are not enough words of thanks for you accepting me into your lab and teaching me all that you have. You have allowed me such independence, but even when I was not aware of it, always provided a guiding hand and a support system if things got difficult. I can look back and see how I have transformed as a researcher and a mentor, and gained confidence in myself and my work, and I have you to thank for that. You are not only an incredible teacher and mentor, but lead by example in all that you do. Almost on a daily basis, I realize that so many of my thoughts and behaviors are a reflection of what you have taught, and I hope that continues for the rest of my career, because there is no better example to follow or person to try to emulate.

A thank you to Darlene does not come without a thank you to Jim Howard. I truly got a two-for-one deal in the two best advisors there are. Your brilliance and passion for research has been an inspiration, Thank you for teaching me to think about work in a greater level of detail and connection than I knew how. And thank you for being the picture of never stopping learning or being afraid to try new techniques or ideas.

To my entire lab, past and present members, each of you were amazing to work with. Lani Bennett and Jess Simon, you were the best lab big sisters I could ask for, and you gave me big shoes to fill and set a high standard that I was always reaching for. Thank you for being such tremendous guides. Chelsea Stillman, lab little sister, thank you for your daily support, and for letting me pretend to lead the lab! To the lab managers, Lauren Westbay, Halley Feldman and Eileen Rasmussen, I didn’t know how much I needed you until the first day you weren’t at work. You kept my life and work organized and running. Thank you to the undergraduate research assistants, Marcie King, Catie Profaci, Jessie Schwab, Alyssa Coffin, Caroline Wambach, RJ
Marchese, Erica Rabinovich, Joanna Lee, Maija Paegle, without whom I would not have been able to complete any of my projects. Thank you for allowing me to be your mentor, your colleague, and your friend. You made my life easier, and made it a joy for me to go to work every day.

The Psychology department at Georgetown has been an incredible environment to grow up as a researcher and get my Ph D. The faculty is a fantastic group of researchers and teachers. I learned so much from everyone I had contact with, whether through classes or being a teaching assistant, and constantly felt supported by everyone. The graduate students, past and present, made the program a phenomenal experience. The level of support and welcoming made a challenging few years so much easier.

To my cohort, Eric Murphy, Natalie Brito, Cristina Novoa and Sarah Vidal, you are my strongholds. I could not have asked for a better group of people to have gone on this journey with. I am so grateful for all that you have taught me, and for the endless support you gave me in all aspects of life. I cannot imagine what Georgetown would have been without you four, and feel so lucky to have you in my life.

Finally, to my family, thank you. I would not be who I am without you, I would not have had the confidence or drive to be where I am today were it not for your confidence in me and examples of always reaching for more and working harder. You not only pushed me to always be better, but provided three incredible examples of what hard work and success look like. This degree is for you three, because it would not be without you.
TABLE OF CONTENTS

Chapter 1: Introduction .........................................................................................1

Parkinson’s disease ......................................................................................2

What is Parkinson’s disease? .................................................................2

Direction of dopamine denervation relates to the progression of cognitive decline .................................................................8

Differential effects of dopaminergic medication on task performance ..........10

Changes in the prefrontal cortex in Parkinson’s disease .........................12

Learning in tasks with phasic vs. tonic dopamine exposure .....................14

Compensation of other brain regions for striatal declines .......................18

Implicit sequence learning ..........................................................................21

Implicit learning .........................................................................................21

Sequence learning and dopamine ............................................................22

Role of the striatum in implicit sequence learning ...................................23

Sequence learning in Parkinson’s disease .................................................26

The importance of sequence learning in Parkinson’s disease .................26

Sequence learning in a motor reaching task ............................................27

Sequence learning in performance-adjusted tasks ....................................29
Varied ways of examining learning in the Serial Reaction Time task ………..30

Mixed findings of sequence learning in Parkinson’s disease …………………31

Potential problems with using the SRT in Parkinson’s disease ………………35

The Triplets Learning Task with PD participants ...............................36

The present studies .............................................................................38

Chapter 2: Method ........................................................................... 43

Participants .........................................................................................43

Parkinson’s disease ...........................................................................43

Healthy controls ...............................................................................44

Tasks ..................................................................................................44

Triplets Learning Task .......................................................................44

TLT Recognition task .........................................................................46

Grooved Pegboard Test .....................................................................46

Scales for Outcomes of Parkinson’s disease – Cognition ....................46

Mini Mental State Examination .........................................................47

Montreal Cognitive Assessment .......................................................47

Geriatric Depression Scale ..............................................................47
SCOPA-COG and learning ................................................................. 64

Intraindividual variability and disease related characteristics ..................64

Study 2 Conclusions ........................................................................ 65

Chapter 5: General Discussion ..........................................................66

Summary ........................................................................................ 66

Study design .................................................................................. 66

Medication status .......................................................................... 67

Findings and Implications ...............................................................68

Group differences in sequence learning ...........................................68

Group differences in the Half Block comparison ..............................71

Correlations with learning in the PD group .....................................73

Group differences in Intraindividual variability ..............................75

Limitations and Future directions ..................................................76

Medication vs. disease effects .......................................................76

Limited range of disease severity .................................................76

Effects of medication fluctuations ................................................77
Grooved Pegboard effects .................................................................77

Implications .......................................................................................78

References ..........................................................................................81

Tables ..................................................................................................97

Figures and ANOVAS .........................................................................102

Appendix .............................................................................................123
CHAPTER 1: INTRODUCTION

Parkinson’s disease is the second most common neurodegenerative disorder behind Alzheimer’s disease (Brockmann et al., 2013), and while its symptoms can often be controlled through medication, there is no cure for the disease nor known cause for most people living with it (National Parkinson Foundation, 2012). While Parkinson’s disease itself is not fatal, deficits that occur with the disease can result in problems that can be fatal, such as an increased incidence of falls (The Michael J. Fox Foundation, 2013; Loftus, 2009). Increased falls in people with Parkinson’s disease are often attributed to an overall deficit in sequencing abilities (Loftus, 2009) which are necessary for more motor-based functions such as speaking and walking (Abbruzzese, Trompetto, & Marinelli, 2009). People with Parkinson’s disease also have problems with comprehension of complex sentences (Grossman et al., 2001) and language production (Illes, Metter, Hanson, & Iritani, 1988), suggesting that the disease affects overall sequencing abilities, not only those that rely on motor movement. It is important to better understand sequencing deficits in Parkinson’s disease because sequencing is involved in rehabilitation (Abbruzzese et al., 2009) as well as everyday activities, such as taking medication at the correct times or in the correct doses.

Sequencing has been examined in people with Parkinson’s disease using various sequence learning tasks, but results have been mixed, with some studies showing deficits in learning and others not. The present studies examined implicit sequence learning in medicated Parkinson’s disease participants, who have striatal deficits due to a loss of dopamine (Kish, Shannak, & Hornykiewicz, 1988). We compared implicit sequence learning in our Parkinson’s disease group to healthy controls, and also examined individual differences in learning within the Parkinson’s group. Learning in the task we used has been shown to relate to dopamine
availability (Simon, Stollstorff et al., 2011) and activation in the striatum (Simon, Vaidya, Howard Jr., & Howard, 2012). In Study 1, we examined how learning in the Parkinson’s group compared to Controls, and if there were group differences, if they were greater during points in training that have been shown or suggested to rely on brain regions affected by Parkinson’s disease. Group differences would provide additional evidence to better understand the neural bases of implicit sequence learning. In Study 2, we examined how learning at different parts of training in the Parkinson’s disease group related to disease severity and other neuropsychological measures.

In this Introduction, I will first give a background of what Parkinson’s disease is, what we know about deficits that occur with the disease, and explore a variety of studies with Parkinson’s disease participants that do not involve sequencing, to better understand some cognitive effects of the disease. I will then briefly discuss sequence learning, and how dopamine and the striatum, both of which show deficits in Parkinson’s disease, are important to learning. I will then review some of the findings of sequence learning in people with Parkinson’s disease, and suggest possible reasons for the mixed findings. I will end by discussing the goals of the present studies, and how they go beyond what has previously been studied in people with Parkinson’s disease.

**Parkinson’s disease**

**What is Parkinson’s disease?** In 1817, James Parkinson described a “shaking palsy,” a disease he characterized by motor deficits and a tremor (Parkinson, 1817). He distinguished this particular palsy from other types of palsies or shaking disorders through causes and symptoms. In 1877, Jean-Martin Charcot changed the name of the shaking palsy to Parkinson’s disease (Lees, 2007), and further described some of the symptoms, such as bradykinesia (slowed
movement), resting tremor, and difficulty with balance (International Parkinson and Movement Disorder Society, 2013). Almost a century and a half later, these are the motor deficits still used to classify Parkinson’s disease. Additional symptoms include rigidity (an increased stiffness or resistance to passive limb movement), hypokinesia (reduced amplitude of movement), and akinesia (an absence of normal, unconscious movements, such as the swinging of one’s arms while walking), as well as effects on facial movements and handwriting, as the disease progresses (Dauer & Przedborski, 2003).

Parkinson’s disease (PD) is a neurodegenerative disorder that is characterized by the motor deficits mentioned above (Cools, 2006), as well as difficulty initiating movement, and maintaining postural control (Hoehn & Yahr, 1967). This collection of motor symptoms is used for classification into one of five stages on the Hoehn and Yahr scale:

Stage 1: “unilateral involvement with minimal or no functional impairment,”

Stage 2: “bilateral or midline involvement, without impairment of balance,”

Stage 3: “first sign of impaired righting reflexes…unsteadiness…somewhat restricted in activities…physically capable of leading independent lives, and their disability is mild to moderate,”

Stage 4: “fully developed, severely disabling disease; the patient is still able to walk and stand unassisted but is markedly incapacitated,”

Stage 5: “confinement to bed or wheelchair unless aided” (Hoehn & Yahr, 1969, p. 133).

Stage classification changes as motor deficits progress, moving from unilateral to bilateral involvement, or as motor impairments become severe enough to affect daily life.

The Unified Parkinson’s Disease Rating Scale (UPDRS) is another, more sensitive scale for classifying symptoms and deficits in people with Parkinson’s disease (Fahn & Elton, 1987).
The UPDRS has six sections, (1) Mentation, Behavior and Mood, (2) Activities of Daily Living, (3) Motor Examination, (4) Complications of Therapy, (5) Hoehn and Yahr Staging, and (6) Schwab and England Activities of Daily Living Scale (Gancher, 2002). Each of these has a list of items on which a person with PD can be rated from “normal” to “severely affected,” such that each section can be scored separately, and together they give an overall UPDRS score, with higher scores indicating more impairment. The UPDRS motor score (section III) has been shown to negatively correlate with the levels of striatal dopamine, such that ratings on this scale are related to the most prominent biological marker of PD, with greater dopamine decline relating to more motor impairment (Ishikawa et al., 1996). The motor section of the UPDRS has additionally been found to be sensitive to disease progression with time, and to a greater degree than Hoehn and Yahr classification (Evans et al., 2011). Inter-rater reliability is poor in the UPDRS, particularly the motor section (Post, Merkus, de Bie, de Haan, & Speelman, 2005), but individual rater reliability is high (Siderowf et al., 2002), suggesting that although it may be difficult to compare these ratings across studies, the UPDRS is a good scale for measuring motor impairment in people with PD within a study when the rater is the same for all participants.

One million people in the United States and four to six million worldwide are currently living with PD, while 50,000-60,000 people are newly diagnosed in the U.S. each year (National Parkinson's Foundation, 2012). Based on the current prevalence of the disease within specific age groups and the projection of growth of these age groups, these numbers in the U.S. are expected to nearly double by the year 2030 (Dorsey et al., 2006). Complications from the disease make up the 14th leading cause of death, and the disease has costs of approximately $25 billion a year in the U.S. alone (Parkinson's Disease Foundation, 2013). An individual’s medication costs may be as high as $2,500 per year, and treatment using bilateral deep brain
stimulation can cost up to $100,000. The incidence of hospital visits also increases in people with PD, which leads to both decreased well-being and increased health care costs (Hassan, 2011). Among the leading reasons for hospital visits in people with Parkinson’s disease is a higher incidence of falls, which can occur as a result of motor impairments or of cognitive effects, such as impaired sequencing (Loftus, 2009). Up to 70% of people with PD report falling at least once a year (Bloem, Steijns, & Smits-Engelsman, 2003), and these falls lead to a 130% increased chance of hospitalization compared to non-PD patients, as PD patients’ falls often result in more serious injury, such as hip fractures (c.f., Grimbergen, Munneke, & Bloem, 2004).

Most cases of Parkinson’s disease are idiopathic, or do not have a known cause, but about 15 – 25% of people diagnosed with the disease have a relative with the disease, which has led to the investigation of possible genetic links (Parkinson's Disease Foundation, 2013). People with early-onset PD (before the age of 40) more frequently have an underlying genetic cause, such as mutations in the Parkin, PINK-1 and DJ-1 genes, though studies have more recently shown that the parkin mutation sometimes also occurs in people who are older at disease onset (Dauer & Przedborski, 2003). Recent genome wide studies have allowed a far-reaching examination of genetic correlations with idiopathic PD to examine additional genes that may play a role in the disease. The α-synuclein (SNCA) single nucleotide polymorphism (SNP) on chromosome 4q22, for example, has been linked to familial forms of PD, but has been shown to have involvement in idiopathic PD, as well (Simon-Sanchez et al., 2009). The intracellular Lewy Bodies that are the hallmark of idiopathic PD are composed of alpha-synuclein deposits (Dauer & Przedborski, 2003), although the role of Lewy Bodies in the disease process is not clear (Libow, Frisina, Haroutunian, Perl, & Purohit, 2009). Another SNP, MAPT, on chromosome 17q21, was also found to be associated with idiopathic Parkinson’s disease. Multiple SNPs within LRRK2 are
also associated with idiopathic PD (Simon-Sanchez et al., 2009), which had previously been associated with genetically-linked PD (Zimprich et al., 2004). Despite how much knowledge has recently been gained regarding genes that may play a role in PD, much more work is needed to determine the extent of genetic underpinnings in this disease, particularly those that are currently classified with an unknown cause.

Physiologically, PD manifests through a decrease in dopamine in the striatum (Kish et al., 1988), a region of the brain involved in motor control (Strafella, Paus, Fraraccio, & Dagher, 2003). This decrease in striatal dopamine results from the death of dopamine-generating cells in the substantia nigra (Marsden, 1992), with motor symptoms not appearing until there is a 50% loss of nigral neurons (Fearnley & Lees, 1991) and an 80% loss of striatal dopamine (Marsden, 1992). These findings from post-mortem studies in humans are supported by work in animals; when striatal dopamine neurons were lesioned in rats, there was an all-or-none motor impairment in forepaw adjustments, such that impairment only occurred after 80% of neurons were lesioned (Chang, Wachtel, Young, & Kang, 1999). Thus, dopamine decline begins long before PD can be diagnosed via motor symptoms. Pre-symptomatic declines of dopaminergic nigral neurons have been found to occur approximately 4.7 years before PD motor symptoms appear (Fearnley & Lees, 1991). Thus, anyone who is diagnosed with PD has a loss of at least 80% of their striatal dopamine, and has spent years living with a progressive dopamine decline.

Parkinson’s disease has been associated with advanced aging, as the progressive dopamine loss characteristic of the disease has been seen as an acceleration of typical age-related dopamine loss that occurs over the course of a lifespan (Kish, Shannak, Rajput, Deck, & Hornykiewicz, 1992). Although striatal dopamine loss also occurs in healthy aging (in the absence of PD), it has become clear that the **pattern** and **magnitude** of dopamine loss in PD
differs from that in healthy aging (e.g., Fearnley & Lees, 1991; Huang et al., 2007). In PD, the
loss of striatal dopamine begins in the putamen, moving in a caudal (back) to rostral (front)
direction (Kish et al., 1988), while dopamine denervation in healthy controls moves rostral to
caudal (Kish et al., 1992). Post-mortem measures of dopamine levels comparing people who
died with PD to those who died in good health, showed a 97.8% difference in dopamine levels
between the two groups in the putamen (Kish et al., 1988). In contrast to the putamen, dopamine
denervation in the caudate moves in a rostral to caudal direction in both people with PD and
healthy controls (Kish et al., 1988; Kish et al., 1992), though denervation occurs to a greater
extent with PD, as post-mortem studies showed that people with PD had 81.5% less dopamine
compared to deceased healthy controls (Kish et al., 1988). Thus, there is a greater loss of
dopamine in both the putamen and the caudate in people with PD compared to controls, but the
loss in the putamen is greater than that in the caudate (Kish et al., 1988).

Post-mortem studies of healthy controls showed an average dopamine loss of 60% in
both the putamen and caudate, from ages 14 to 92 (Kish et al., 1992), and in the substantia nigra,
a loss of 33% of dopamine neurons from 20 to 90, or a 4.7% neuronal loss each decade (Fearnley
& Lees, 1991). In contrast, people with PD have a 45% loss of substantia nigra neurons in the
first decade of the disease, showing a much more rapid initial dopamine decline in PD compared
to a ten year period in healthy controls. Because of this initial substantia nigra cell loss,
dopamine loss in PD is initially exponential, being ten times greater than that in healthy older
adults (Fearnley & Lees, 1991). As the disease progresses, however, this loss becomes more
linear, as a longitudinal study showed a linear decrease in dopamine transporter binding in the
caudate and putamen over the course of four years in people with PD, with a 5.2% and 3.9%
yearly decrease for the two brain regions, respectively (Huang et al., 2007).
A positron emission tomography study examined dopamine transporter binding, which is a proxy for dopamine denervation (less binding suggests more denervation), and found the least amount of binding in the posterior putamen, followed by the anterior putamen, ventral putamen, caudate head, middle caudate, and the most binding (least denervation) in the anteroventral striatum (Kwak, Bohnen, Müller, Dayalu, & Seidler, 2013), providing additional support to post-mortem studies showing the direction of dopamine denervation in the striatum (Kish et al., 1988). Dopamine declines in the putamen and caudate in people with PD were negatively correlated with scores on the UPDRS motor section (Huang et al., 2007), and neuronal loss in the substantia nigra was found to be positively correlated with PD symptom duration (Fearnley & Lees, 1991), showing that dopamine declines are closely related to manifestations of the disease through motor deficits and symptoms.

Neurons in the substantia nigra primarily project to the putamen (Dauer & Przedborski, 2003), where dopamine denervation is seen first in the striatum (Kish et al., 1988), while the ventral tegmental area, adjacent to the substantia nigra, projects to the caudate and has less cell loss in Parkinson’s disease (Dauer & Przedborski, 2003). These different areas of initial dopamine projection help explain why dopamine denervation occurs later in the caudate than in the putamen in PD (Kish et al., 1988). The ventral tegmental area also has dopaminergic projections to the prefrontal cortex, which, as with the caudate, likely explains why impairment in tasks that rely on the prefrontal cortex is seen later in disease progression compared to initial motor deficits (Dauer & Przedborski, 2003).

**Direction of dopamine denervation relates to the progression of cognitive decline.** The direction in which dopamine denervation occurs in different regions of the striatum helps to explain the progression of cognitive decline seen in Parkinson’s disease. That is, dopamine
denervation begins in the putamen, which is involved in the motor-corticostriatal loop, and then moves to the caudate, involved in the executive-corticostriatal loop (Seger, 2006), a progression of decline that is consistent with motor impairments occurring before cognitive impairments in Parkinson’s disease. The roles of these loops in people with PD and symptom development are further supported by a study that used repetitive transcranial magnetic stimulation, where dopamine release occurred in the caudate following stimulation of the dorsolateral prefrontal cortex (Strafella, Paus, Berrett, & Dagher, 2001), and in the putamen following stimulation of the primary motor cortex (Strafella et al., 2003). Thus, there are close connections between the putamen and the primary motor cortex, and between the caudate and the dorsolateral prefrontal cortex, such that dopamine changes in one region should have an effect on the connected regions. The relationship between dopamine levels in different regions of the brain and their effects on task performance will be discussed further, below (Cools, 2006).

One study used positron emission tomography to measure dopamine levels within the brains of people with PD, and found that dopamine projections to the caudate predicted individual behavioral differences on a spatial sequence learning task, a relationship not seen in healthy controls (Carbon et al., 2004). While the reason for this predictive relationship seen in PD and not in healthy older adults is not clear, the authors suggest that it may be due to a smaller range of striatal dopamine transporter binding across healthy control participants (Carbon et al., 2004), which is in comparison to high variability both across individuals and within the brains of people with PD as dopaminergic denervation increases and binding decreases (Bezard et al., 2001; Olanow, Stocchi, & Lang, 2011). Thus, the degree of dopaminergic denervation is related to behavioral impairment in tasks that rely on the caudate, and it varies widely across individuals with Parkinson’s disease.
Differential effects of dopaminergic medication on task performance. All of the participants with Parkinson’s disease in the present studies were on anti-Parkinsonian medication, so it is important to discuss the effects of medication on task performance, and how these effects can differ by task and disease characteristics in PD participants. Levodopa is the gold standard of medication for people with Parkinson’s disease, and it not only controls motor symptoms, but also significantly improves quality of life (Olanow et al., 2011). Although dopaminergic medication given to people with PD acts as dopamine replacement for regions of the brain where there are dopamine declines (Parkinson’s Disease Foundation, 2013), it does not improve PD-related dysfunctions that are unrelated to dopaminergic function (e.g., insomnia, depression; Olanow et al., 2011). It has it has been suggested that dopaminergic medication may actually impair the function of brain regions where there is little or no dopamine decline by flooding, or “overdosing” them, and thus impairing their functions (Cools, 2006). Specifically, in early PD, the dorsal striatum has the most dopamine denervation, so dopaminergic medication improves performance in tasks that rely on this region of the brain, while other areas of the brain that are not yet affected by the disease, such as the ventral striatum (Cools, 2006), and even the prefrontal cortex (Mattay et al., 2002), may be impaired by dopaminergic medication.

This idea of a dopamine overdose is based on an inverted-U curve of dopamine and its relation to performance. This curve is well demonstrated by Bäckman and colleagues, who presented multiple lines of evidence, through aging, genetic, and neuroimaging research to show that there is an optimal level of dopamine to achieve optimal performance on a given task, represented as the tip of an inverted-U (Bäckman, Nyberg, Lindenberger, Li, & Farde, 2006). Where participants lie along this curve and thus where this optimal dopamine point is, varies across individuals depending on their level of dopamine. In this inverted-U, dopamine levels are
typically on the x-axis and performance on the y-axis. Thus, low dopamine levels will match with low performance (left side of the inverted-U), while at the mid-point of each axis dopamine and performance levels will be optimal, and then on the right side of the curve dopamine levels become too high, and performance declines. This theory has been supported through aging and genetic research (c.f., Bäckman et al., 2006; Nagel et al., 2008), and has even been shown in healthy young participants, where researchers compared the effects of Levodopa on performance on a combination Simon/Stroop task in healthy young and older adults (Onur, Piefke, Lie, Theil, & Fink, 2011). In this task, older adults showed similar behavior and functional activation following both a placebo and a dose of Levodopa, while young adults who received Levodopa had worse performance in the conflict adaptation task, such that higher dopamine levels were related to worse performance, as well as more activation in the anterior cingulate. The authors suggest that in young adults, the prefrontal cortex was overdosed by Levodopa, putting them onto the right side of the dopamine inverted-U curve, such that the prefrontal cortex was no longer the best region to support learning, causing the anterior cingulate cortex to activate more in an attempt to improve performance. Thus, even in young adults, a group without dopamine declines, Levodopa affects performance; by adjusting dopamine levels, performance can be positively or negatively affected depending on which side of the dopaminergic inverted-U people lie.

Because the effects of varying dopamine levels have been shown through both genetic differences and administration of dopamine replacement drugs in healthy participants, it is not surprising that these differences are true of people with dopaminergic deficits, such as those that occur in Parkinson’s disease. People with mild to moderate PD were tested ON and OFF medication in a Stroop-like task that was designed to measure conflict adaptation (Duthoo et al.,
When ON medication, participants did not show conflict adaptation, that is, they were not able to learn how to use inhibition in response to incongruent stimuli, and thus remained slower to incongruent than congruent trials over practice. However, when these same individuals were tested OFF their medication, they were able to improve their behavior in this task, such that their response times to incongruent trials became faster, and more similar to those in congruent trials with practice. The authors suggested that impaired performance ON medication was due to the overdosing effect of dopaminergic medication, such that brain regions which are involved in conflict adaptation that do not have dopaminergic declines with PD, such as the anterior cingulate cortex, may have been negatively affected, or “overdosed,” by medication, thus impairing performance on the Stroop task.

**Changes in the prefrontal cortex in Parkinson’s disease.** People with PD often exhibit deficits in tasks that rely on the prefrontal cortex; one group of medicated PD participants had similar performance to that of people with frontal lobe damage on the frontal-based Tower of London task (Owen et al., 1992). In a n-back task which involves working memory, when people with PD (Hoehn and Yahr stages [H&Y] 1 and 2) were OFF their medication, they had more activation in the prefrontal cortex and the parietal and cingulate regions, but equivalent behavior compared to when they were ON their medication (Mattay et al., 2002). Despite behavioral equivalence in the two medication states, in the OFF state, performance on the n-back was negatively related to cortical activation, suggesting that the increased cortical activation was a sign of inefficiency, rather than compensation. This deficit in a prefrontal-related task was seen in people with early PD, when dopamine denervation is still isolated to the striatum, which suggests that prefrontal deficits may occur as a result of a disruption of dopaminergic projections to the prefrontal cortex from the striatum (c.f., Cools, Miyakawa, Sheridan, & D'Esposito, 2010).
This disruption could occur in two ways, either through disrupted mesocortical connections from the ventral tegmental area, or through projections of the D1 receptors from the striatum (Mattay et al., 2002).

It has also been shown that dopamine in the striatum and the prefrontal cortex have an inverse relationship, such that high levels of dopamine in one region correspond to low levels in the other (c.f., Cools et al., 2010). Thus, when people with PD have dopamine denervation only in the striatum, when OFF medication, there is an increase in the level of dopamine in the prefrontal cortex, which may reflect compensation for decreased levels of striatal dopamine. The inverse is true, however, when people are on dopaminergic medication, where being ON medication would increase dopamine binding in regions of the striatum that have dopamine denervation, which may cause a decrease in dopamine levels in the prefrontal cortex. People with PD were tested both ON and OFF medication and compared to healthy controls in a prefrontal-based delayed response task in the presence of a distractor (Cools et al., 2010). When people with PD were OFF their medication, they showed significantly better distractor resistance than when they were ON and when compared to healthy controls. The authors suggested that having less striatal dopamine (no dopamine replacement when OFF medication) caused dopamine levels to be higher in the prefrontal cortex, thus allowing people with PD who were OFF medication to inhibit the distractor even beyond that of healthy older adults. These results fit with models suggesting a facilitating effect of dopamine in the prefrontal cortex to improve target detection in the presence of distractors.

One study examined performance in the Tower of London task in controls and people with PD OFF medication (H&Y 2 and 3), and found not only the involvement of the prefrontal cortex in the absence of striatal activation, but also additional activation in the hippocampus in
the PD group (Dagher, Owen, Boecker, & Brooks, 2001). Controls and the PD group had equal
task performance, and both groups showed activation in the prefrontal cortex, rostral anterior
cingulate cortex and posterior parietal areas, but healthy controls additionally activated the right
dorsal caudate, which was positively correlated with task complexity. The PD group did not
have caudate activation, but did show task complexity-related hippocampal activation during the
task that was not seen in controls. These results suggest that both the prefrontal cortex and the
hippocampus may have been compensating for a lack of striatal involvement.

This collection of studies shows that dopamine denervation and declines in the striatum
do not occur in isolation. That is, the prefrontal cortex can compensate for striatal deficits
through increased dopamine levels that can not only improve performance in prefrontal-based
tasks (Cools et al., 2010), but also in tasks that rely on the striatum in healthy adults (Dagher et
al., 2001). This will be explored more, below, with other brain regions.

**Learning in tasks with phasic vs. tonic dopamine exposure.** The striatal deficits that occur in
people with PD allow this population to be used to examine how performance differs in striatal-
based tasks as a result of the disease in comparison to healthy controls. Results yield not only
information about task performance in PD participants, but also insights into the neural bases of
specific tasks.

People with PD have been shown to have deficits in habit formation (de Wit, Barker,
Dickinson, & Cools, 2010), or the linking between stimuli and actions, which is related to the
integrity of projections from the substantia nigra to the dorsal striatum (Faure, Haberland, Condé,
& Massiou, 2005). A model of PD showed that phasic striatal dopamine, which is critical for
stimulus-response learning from feedback, is less potent in people with PD due to low tonic
dopamine levels (Frank, 2005). Compared to what is seen in healthy aging, the decreased size of
phasic dopamine bursts in PD participants likely results in a decreased association of representations following feedback in associative learning tasks (Frank, Seeberger, & O'Reilly, 2004). In a probabilistic classification task with feedback, people with PD (regardless of medication state) were impaired at associative learning, but had intact declarative knowledge of associations, showing a deficit in feedback-driven stimulus-response learning (learning that relies on the striatum), but no effects on more explicit learning, which is generally thought to be hippocampal-dependent (de Wit et al., 2010).

Frank’s (2005) model suggests that dopaminergic medication increases tonic dopamine levels in people with PD when they are ON compared to OFF medication, such that being ON medication will cause the brain to be less sensitive to smaller phasic dopamine increases than during an OFF state. This model was empirically supported in a study using the Weather Prediction Task (WPT), a feedback-driven probabilistic classification task that relies on both the hippocampus and striatum in an interactive manner (Poldrack & Rodriguez, 2004). In the WPT, when people with PD were OFF (H&Y $M = 2.8$) their medication, learning was equivalent to that of healthy controls, and significantly better than when they were ON (H&Y $M = 2.2$) medication (Jahanshahi, Wilkinson, Gahir, Dharminda, & Lagnado, 2010). The amount of learning in people with PD when OFF medication was marginally positively related to the levels of medication they were prescribed (Levodopa equivalence). This suggests that intrinsic (OFF medication) dopamine levels were very low in people who were prescribed high levels of medication, such that when medication was withdrawn, they were very sensitive to phasic bursts of dopamine from feedback, which strengthened learning in the WPT. In contrast, when ON medication, there was a non-significant negative relationship between medication levels and learning, such that higher levels of medication were weakly related to less learning. Though not
significant, the direction of this relationship was likely due to the medication producing such a high level of tonic dopamine in the system, that learning-related phasic bursts of dopamine could not peak high enough to form associations within the striatum.

One study examined the association between dopamine transporter binding (less binding indicates more denervation) in the striatum and performance on a Stroop-like task paired with reward in people with PD (H&Y $M = 2.0$) who were OFF medication (Aarts et al., 2012). They found that less dopamine transporter binding in the posterior putamen was associated with a higher response to reward, i.e., more sensitivity to dopamine bursts. These findings fit with others, suggesting that when tonic dopamine levels in people with PD are low due to a withdrawal from dopaminergic medication, phasic dopamine bursts that occur following reward are effective in producing associations and good task performance (Frank, 2005).

The timing of feedback in a probabilistic classification task is essential, as the timing of phasic dopamine bursts is what allows associations between responses and rewards; if feedback occurs long enough after a response, dopamine bursts may result in incorrect associations (Frank, 2005). The effects of immediate versus delayed feedback in learning associations were tested in a probabilistic classification task (Foerde & Shohamy, 2011). A functional neuroimaging study with young adults showed the dorsal and ventral striatum and the hippocampus were sensitive to prediction error, such that they showed a greater response to correct feedback (expected outcome) than to incorrect feedback (unexpected outcome). There was a dissociation in feedback timing, however, where the ventral and dorsal striatum were sensitive to prediction error when feedback was immediate, while the hippocampus was sensitive to prediction error when feedback was delayed. This suggests that the striatal system is effective when rewards, and thus dopamine bursts, are immediate, while the hippocampus supports associative binding
over time and following longer delays. The authors suggest that the hippocampus may be more responsive to tonic dopamine levels, in contrast to the striatum that is more responsive to phasic bursts of dopamine for associative learning.

This same probabilistic classification task with delayed and immediate feedback was examined in people with PD (H&Y 1-3), comparing them to healthy older adult controls (Foerde & Shohamy, 2011) and to people with amnesia (Foerde, Race, Verfaellie, & Shohamy, 2013). Healthy controls learned equally well from both types of feedback, while people with PD had impaired learning from immediate feedback, but intact learning from delayed feedback (Foerde & Shohamy, 2011), and people with amnesia, who had hippocampal lesions, had intact learning from immediate, but impaired learning from delayed feedback (Foerde et al., 2013). Thus, results from two clinical populations show a dissociation, with the hippocampus underlying immediate feedback and the striatum underlying delayed feedback. Together, the neuroimaging study with young adults and the behavioral studies with healthy older adult controls, people with amnesia and people with PD, converge to suggest that with delayed feedback in a probabilistic learning task, both the hippocampus and the striatum underlie associative learning; the hippocampus integrates information across time, while the striatum is involved in more immediate associations and relational binding (Foerde et al., 2013; Foerde & Shohamy, 2011). Most participants (13 of 17) in Foerde and Shohamy’s study were medicated during test, while participants in the study by Foerde et al. withheld medication during the task, but medication did not change the results between the two studies, similar to the lack of a medication effect on results from de Wit et al. (2010).

The collection of studies reviewed above examined learning in people with PD in tasks that relied on either phasic (striatal-based) or tonic (hippocampal-based) bursts of dopamine. In
general, results showed that PD participants only had intact learning in tasks that relied on phasic bursts of dopamine, largely tasks or portions of tasks that relied on the striatum, when they were OFF medication (lower levels of striatal dopamine). In contrast, when tasks were responsive to tonic levels of dopamine or could be completed by the hippocampus, such as declarative knowledge of associations or immediate feedback learning, performance was intact in PD regardless of medication status.

**Compensation of other brain regions for striatal declines.** The ability of the brain to reorganize in light of injury or decline has been examined in various clinical populations, and is a particularly pervasive idea in the aging literature (Cabeza, 2002; Park & Reuter-Lorenz, 2009). For example, with healthy aging, it has been shown that older adults will activate homologous regions in both hemispheres to successfully complete a task in which younger adults may only activate one hemisphere (Cabeza, 2002), or may additionally recruit the prefrontal cortex to aid in tasks that rely on brain regions that show age-related decline, such as the hippocampus (Park & Reuter-Lorenz, 2009). This plasticity has also been shown in people with Parkinson’s disease. For example, one study trained participants in a balance task over a six week period, and found a linear increase in gray matter in the cerebellum in people with PD (H&Y 1-3) that was not seen in controls (Sehm et al., 2014). While this change was structural, it supports the hypothesis that brain plasticity is maintained in Parkinson’s disease, and that other regions may adapt to help compensate for disease-related declines.

A review examined the involvement of the cerebellum in task performance, and found evidence for it as a compensatory mechanism in Parkinson’s disease (Martinu & Monchi, 2012). In people with PD, two regions of the striatum which inhibit motor movements, the subthalamic nucleus and the globus pallidus internal, are over-active compared to controls. In contrast, the
globus pallidus external, which is excitatory for motor movements, is underactive in PD, leading to some of the movement deficits in PD, such as bradykenesia and rigidity. This increased inhibition of motoric movements causes people with PD to have difficulty self-initiating movements, but visual cues can help guide these voluntary movements. Martinu and Monchi suggest that this improvement from visual cues may be controlled by the cerebellum. While rarely thought of as a brain region that has dopaminergic involvement, the cerebellum is strongly modulated by visual feedback, has a high density of dopamine receptors, has underlying connections to motor and cognitive functions, and showed more activation during automatic motor movements in people with PD compared to controls.

The hippocampus is another brain region suggested to be compensatory in PD, as mentioned above in a study using the Tower of London task (Dagher et al., 2001). A role of the hippocampus as compensatory for striatal deficits in PD has also been shown in the Weather Prediction Task (Moody, Bookheimer, Vanek, & Knowlton, 2004). Healthy controls had significantly more task-related activation in the caudate and globus pallidus than people with PD who were ON medication (H&Y ON 1-2.5, OFF 1.5-3), while the PD group showed significantly more task-related activation in the prefrontal cortex. The PD group also had significant task-related activation in the hippocampus and parahippocampal gyrus, activation not seen in controls. Hippocampal activation increased over the course of the task in the PD group, while healthy controls showed hippocampal deactivation as the task continued, a finding that was consistent with previous studies of the Weather Prediction Task in healthy older adults. Thus, hippocampal activation in PD was not only greater than in controls, but it was related to better task performance, which also increased throughout the task, suggesting that it was beneficial and may have been compensating for a lack of activation in the striatum that was seen in controls.
Despite the fact that the hippocampus has been shown to be compensatory in PD (Dagher et al., 2001; Moody et al., 2004), hippocampal atrophy does occur with the disease. A study with healthy older adults, found that hippocampal atrophy does not begin until after the age of 65 (Raz et al., 2005), while a study comparing healthy older adults to people with PD found that atrophy began after the age of 70 in healthy controls, but began earlier in non-demented people with PD (Bouchard et al., 2008). Another study found that people with PD who were not demented showed atrophy in the hippocampus not only beyond that of healthy controls, but to a similar degree of that in people with Alzheimer’s disease and people with PD who had dementia (Camicioli et al., 2003). Additionally, even though non-demented people with PD in this study did not meet DSM-IV criteria for dementia, their performance on reading and recall tests was worse than that in healthy controls, and was similar to people with PD dementia on these and a recognition test. These results suggest that hippocampal atrophy may be related to declines in some cognitive processes.

In the previous two studies, all PD participants had an average disease duration of at least five years, and all were taking dopaminergic medication (Bouchard et al., 2008; Camicioli et al., 2003), so a third study investigated the possibility that this PD-related hippocampal atrophy may be a consequence of disease duration or medication by examining brain volume in medication naïve PD participants who had a short (M 1.7 years) disease duration (Brück, Kurki, Kaasinen, Vahlberg, & Rinne, 2004). Results showed that in this sample of unmedicated PD participants, there was significantly more atrophy in the hippocampus and the prefrontal cortex with PD than in healthy controls. Additionally, hippocampal atrophy was associated with impaired verbal memory, and prefrontal atrophy was associated with increased reaction time on a test of vigilance. Thus, both atrophy and related cognitive impairment occurred in people with PD who
were medication naïve and newly diagnosed with the disease; this suggests that not all cognitive
deficits seen in people with PD are due to effects of long-term dopamine denervation or to a
dopaminergic medication overdose to certain regions of the brain.

As previously mentioned, plasticity is maintained in the brains of people with PD, such
that other brain regions (e.g., prefrontal cortex and cerebellum) are able to compensate for
decreases. The studies reviewed in this section additionally show that the hippocampus can help
compensate for striatal declines (Dagher et al., 2001; Moody et al., 2004), and that it is able to do
so despite hippocampal declines that occur in people with PD beyond that seen in healthy aging
(Camicioli et al., 2003).

**Implicit Sequence learning**

**Implicit learning.** Implicit learning occurs without intent to learn or awareness of having
learned (Reber, 1989). This type of learning allows us to be sensitive to regularities in our
environment, and adapt to changes in physical and social cues. Implicit learning contributes to
countless behaviors, from learning new languages (Kuhl, 2004), to understanding and
interpreting social cues (Lieberman, 2000), to various types of rehabilitation (Abbruzzese et al.,
2009). Compared to more explicit forms of learning and memory, implicit learning has been
relatively understudied, particularly in terms of how learning changes with age and neurological
disturbance.

The current research focuses on one form of implicit learning, implicit *sequence* learning,
which is often studied using the Serial Reaction Time (SRT) task (Nissen & Bullemer, 1987). In
the SRT, participants use keypresses to respond to the location of a series of dots on a computer
screen that, unbeknownst to participants, follows a repeating pattern. Sequence learning is
assessed by determining the extent to which participants respond more quickly and/or more
accurately to events that conform to this repeating pattern compared to randomly determined sequences. This is the type of implicit learning task that will be used in the present studies, so here, we examine what is known about the behavior and mechanisms that underlie sequence learning.

While the studies we present here with our PD group in the Triplets Learning Task (TLT; Howard Jr., Howard, Dennis, & Kelly, 2008) measure implicit learning, not all sequence learning is implicit. Some sequence learning tasks give explicit instruction to find a sequence, and once it is acquired, to use knowledge of it to perform (e.g., motor reaching tasks and some SRT tasks), while some SRT tasks do not give explicit instructions about a sequence, but explicit awareness of the pattern occurs during training (this will be discussed further, below). Therefore, we will indicate whether sequence learning tasks are explicit or implicit; the TLT (discussed in greater detail, below) will always involve implicit learning (Howard Jr. et al., 2008; Simon, Howard Jr., & Howard, 2011).

**Sequence learning and dopamine.** Sequence learning has been suggested to rely on striatal dopamine (Seger, 2006). This conclusion is supported by research in groups who have depleted dopamine levels, such as people with Parkinson’s disease (Shohamy, Myers, Grossman, Sage, & Gluck, 2004), as well as healthy older adults who have decreased dopamine levels and sequence learning compared to young adults (Rieckmann & Backman, 2009). One study examined implicit sequence learning in the SRT during PET imaging to determine striatal dopamine binding potential, and found that binding potential in the striatum and motor cortices was negatively related to sequence learning (Garraux, Peigneux, Carson, & Hallett, 2007). The authors suggested that this negative relationship was due to dopamine release in these areas.
during the task, such that there were higher levels of available dopamine in the synapse to bind to dopamine receptors, decreasing the binding potential for a tracer.

The influence of dopamine on implicit sequence learning was also examined indirectly in the Triplets Learning Task (described further, below; Howard Jr. et al., 2008; Simon, Stollstorff et al., 2011). This study found that in a group of college-aged adults, more sequence learning occurred in people who had a particular variant of a dopamine transporter gene that is related to higher levels of dopamine availability. These studies converge to suggest the importance of more dopamine availability for successful sequence learning. Healthy older adults and people with Parkinson’s disease can be studied with this type of learning to examine how dopamine, and therefore striatal, declines affect sequence learning.

**Role of the striatum in implicit sequence learning.** The neural bases of implicit forms of learning were discovered to be different from those underlying declarative forms of learning by Brenda Milner when examining the patient, H.M. (c.f., Squire, 2009). Milner discovered that H.M., a patient who had a bilateral medial temporal lobe lesion, was able to learn procedural motor skills, such as mirror-tracing, without any memory of having taken part in the task. These findings led to years of hypotheses about a dissociation between declarative and nondeclarative forms of learning and memory and the brain regions that underlie them, but more recent theories suggest an interactive relationship between the neural systems (Henke, 2010; Shohamy & Turk-Browne, 2013). For example, a review of one implicit learning task, a probabilistic classification task, concluded that the striatum, which is thought to underlie implicit learning, and the medial temporal lobe (MTL), largely involved in more declarative types of learning, have an interactive relationship during performance of this task (Poldrack & Rodriguez, 2004). A neuroimaging study examining learning-related activation in the SRT task found a *cooperative* relationship
between these two brain regions, as learning was related to activation in both regions across four runs, but there was more activity in the MTL in earlier training and more caudate activity later in training (Schendan, Searl, Melrose, & Stern, 2003). These results suggest that both brain regions were involved in learning in the SRT, but the region that was more dominant in supporting learning varied by amount of training.

More recently, studies have examined how the brain regions underlying implicit sequence learning differ in healthy older adults compared to young adults, as it has been shown that there are age-related declines in the striatum, while the hippocampus has relatively little decline with age in the absence of pathology (Raz et al., 2005). Behaviorally, age differences are often not seen in the SRT, but there are age-related differences in functional neuroimaging, with older adults showing more MTL activation during learning, and younger adults showing more striatal activation (Dennis & Cabeza, 2011). A similar difference was shown to relate to improvement in sequence learning in the SRT, such that older adults showed learning-related activation increases in both the MTL and the striatum, while in young adults, improved SRT learning related to decreased MTL activation and increased striatal activation (Rieckmann, Fischer, & Backman, 2010).

In contrast to the SRT, in the Triplets Learning Task (TLT), which was used in the present studies, age-related differences in learning typically appear in behavioral measures. A series of studies found consistent age differences in the TLT when the predictability of high frequency triplets was manipulated both within and across studies, when first-versus second-order (adjacent versus non-adjacent cues predicting the location of a target) predictability was manipulated (Howard Jr. et al., 2008), and even when learned triplets were arbitrarily determined (Simon, Howard Jr. et al., 2011). The neural bases of implicit learning in the TLT are similar to
those found in the SRT, with learning-related activation in the hippocampus in early and late training in older adults, and learning-related activation in the caudate, particularly in late training, in young adults (Simon et al., 2012). Though there were age group differences in the brain regions that were related to learning, both the hippocampus and caudate were involved in learning in young and older adults.

In the Alternating Serial Reaction Time (ASRT) task (Howard Jr. & Howard, 1997), a task similar to the TLT, Diffusion Tensor Imaging examined the relationship between the integrity of white matter tracts and learning in young and older adults (Bennett, Madden, Vaidya, Howard Jr., & Howard, 2011). Results showed that implicit learning was related to the integrity of white matter tracts between the dorsolateral prefrontal cortex and the hippocampus in early training, and the dorsolateral prefrontal cortex and the caudate in late training in both young and older adults. Together, these studies highlight the differences between the underlying neural bases of early and late sequence learning, with implicit sequence learning in the TLT and ASRT relying more on the hippocampus for early learning and the caudate for late learning (Bennett et al., 2011; Simon et al., 2012).

This distinction between the relative importance of these neural systems in early versus late learning suggests that compared to controls, people with PD should be more impaired late in training when optimal learning is more reliant on the striatum. In addition, by examining performance within each block throughout training, a recent paper has suggested that the reliance on these two neural systems for learning might change within short periods of time throughout training (Nemeth et al., 2013). Nemeth et al compared learning in the ASRT between a group of healthy young-older adults (M age 58) and an age-matched group of people with Mild Cognitive Impairment, a condition associated with declines in hippocampal structure and function. Overall
analyses revealed no differences in learning between the healthy and MCI groups. However, when they examined learning separately in the 1\textsuperscript{st} Half of Blocks (first 40 of 80 trials in each block) compared to the 2\textsuperscript{nd} Half of Blocks (second 40 of 80 trials in each block) group differences emerged; the MCI group showed less learning compared to the Controls in the 1\textsuperscript{st} Half of Blocks but not in the 2\textsuperscript{nd} Half of Blocks. Nemeth et al. hypothesized that the 1\textsuperscript{st} Half involved recall and reactivation of previously learned sequence, thus reflecting learning dependent on the MTL, while learning in the 2\textsuperscript{nd} Half of Blocks involved more automated and proceduralized behavior of learned sequences, and thus reflected striatal-based learning.

The present study provides an opportunity to test their hypothesis, because their interpretation would predict a double dissociation, such that, in contrast to Nemeth et al.’s (2013) MCI patients who showed impaired learning only in the 1\textsuperscript{st} Half of Blocks, our PD group should be impaired only in the 2\textsuperscript{nd} Half of Blocks when learning reflects striatal involvement.

**Sequence learning in Parkinson’s disease**

**The importance of sequence learning in Parkinson’s disease.** Sequence learning is important for many aspects of daily life, including language (Kuhl, 2004), social intuition (Lieberman, 2000), and walking (Abbruzzese et al., 2009). As discussed above, performance on sequence learning tasks has been shown to rely on the striatum for successful learning, even in older adults (Bennett et al., 2011; Simon et al., 2012) who have striatal declines (Raz et al., 2005). People with PD not only have declines in striatal dopamine (Kish et al., 1988), but have also been shown to have sequencing deficits, manifest in problems with language (Grossman et al., 2001; Illes et al., 1988) and falls (Loftus, 2009). Despite the importance of understanding sequencing deficits in PD, remarkably little is known about how implicit sequence learning is affected by this disease. Thus, a more in depth understanding of how sequence learning changes with PD would
ultimately contribute to developing both effective therapies for PD, and effective diagnostic tools for assessing the progression of dopamine denervation within the striatum in Parkinson’s disease.

**Sequence learning in a motor reaching task.** A number of studies examined explicit sequence learning in a motor reaching task, with all participants tested OFF of their medication. One study showed that learning in a sequence reaching task did not differ between people with PD (H&Y 1) who were tested OFF of their medication and people with PD who were medication naïve (Ghilardi, Eidelberg, Silvestri, & Ghez, 2003). While all PD participants were able to acquire the explicit sequence, the PD group (regardless of medication status) had slower learning rates and did not acquire as much information as Controls. The authors suggested that PD participants may have learned more slowly because early learning in this task (unlike more implicit versions of the SRT) has been shown to rely on the dorsolateral prefrontal cortex, putamen, and caudate, the latter two of which are affected early in Parkinson’s disease.

A second study showed the same behavioral result of slower motor reaching sequence acquisition in people with PD (H&Y 1; OFF or naïve to medication) compared to controls, and also showed that the “typical” acquisition-related activation topography in the brain did not correlate with learning in PD as it did in healthy controls (Nakamura et al., 2001). People with PD not only lacked the typical topography, but activated the pre-supplementary motor area that controls did not activate. These results suggested that people with PD have an altered activation topography for sequence acquisition in this task compared to healthy controls.

A third study of explicit learning in a motor reaching task found equal amounts of learning in healthy controls and people with PD (H&Y 1; OFF or naïve to medication), and found that learning correlated with dopamine transporter binding in the caudate (Carbon et al., 2004). Imaging results showed that learning-related activation in the dorsolateral prefrontal
cortex correlated with dopamine binding in the caudate in people with PD, while in healthy controls, learning-related activation in the cortico-striato-pallido-thalamacortical loop was related to binding in the caudate. These results suggest that people with PD have decreased functional connectivity within this larger cognitive network, such that dopamine binding in the caudate was still related to learning in the PD, but there was decoupling of the typical network that is related to caudate dopamine and learning.

Finally, one study examined longitudinal changes in sequence learning in this motor reaching task as well as a visual sequence task similar to the motor task, over a two year period (Carbon, Reetz, Ghilardi, Dhawan, & Eidelberg, 2010). Behaviorally, they found that both healthy controls and people with PD (OFF medication) had marginal declines in performance on the motor sequence from baseline to follow-up, but performance on the visual sequence did not change. Although the learning decrease in motor sequencing was not significant, there was less decline in healthy controls and in a subset of PD participants than other PD participants. In general, people with PD showed less task-related activation than healthy controls at follow-up, but the subset of people with PD who performed similarly to controls showed increased hippocampal activation with increased learning, activation not seen in controls.

This collection of studies examined a number of factors involved in sequence learning in people with PD, from behavior, to neuroimaging, to longitudinal follow-up studies. The neuroimaging studies showed that although there may be declines in “typical” networks within the brain in PD (Carbon et al., 2004), this plasticity is not always bad, where additional activation in other brain regions, such as the caudate (Carbon et al., 2004) or the hippocampus (Carbon et al., 2010), likely contributed to learning levels in PD being equivalent to that of controls. These studies only examined one type of sequence learning, however, using a task that
was explicit and required overt motor movements to complete. Because people with PD have motor deficits and slower response time (Cooper, Sagar, Tidswell, & Jordan, 1994) than controls, one way to more closely examine sequencing deficits is to use a task that attempts to control for these known deficits in Parkinson’s disease.

Sequence learning in performance-adjusted tasks. Some studies have adjusted task parameters based on participant’s individual initial performance on a task to then further examine group differences in learning between people with PD and controls. One study examined implicit learning in the Serial Interception Sequence Learning Task, a sequence learning task where timing of stimuli presentation was adjusted so that all participants were performing at the same overall accuracy level (Gobel et al., 2013). Results showed that medicated people with PD (H&Y ≤ 2.5) had impaired learning, and learning scores were related to UPDRS scores in people for whom they had ratings. These results need to be interpreted in light of the fact that people with PD were significantly slower than controls, which suggests that the nature of the task, due to the individually adjusted timing, was different for the two groups and may have affected the results and how learning occurred.

Another study examined explicit motor sequence learning, and first determined the longest sequence length that individuals could perform in a reaching task without making a mistake during three training blocks, and then used that maximum sequence length for test during positron emission tomography imaging (Mentis et al., 2003). Results showed that people with PD (H&Y 1; OFF medication) were able to learn sequences, but that the longest sequence they could learn was shorter than that of healthy controls (4-5 elements in PD compared to 6 elements in controls). When performing their personally-adjusted maximum length sequence, people with PD activated the same regions as controls, but additionally activated bilateral
homologues, an activation pattern similar to a control group that performed more difficult tasks. Thus, people with PD could not learn as much as controls, and they required more neural resources to perform at their maximum level than controls needed to perform the six element sequence, which was not maximally difficult for them.

**Varied ways of examining learning in the Serial Reaction Time task.** The SRT is one of the most widely used tasks to study sequence learning in people with Parkinson’s disease. A number of challenges exist in using this task that will be discussed below, but one of the largest challenges is that people with PD have slower motor responding than healthy controls (Cooper et al., 1994), and this can affect their ability to show learning in a task where learning requires rapid and continuous motor responses. Slowed response time in people with PD may make it difficult to see learning through the reaction measure in the SRT, so accuracy can be examined to detect learning (Seidler, Tuite, & Ashe, 2007). Seidler and colleagues tested implicit SRT learning in a group of PD participants who were OFF medication. Learning was examined both with and without a distractor present, which caused the task to be more attention-demanding, and limited the potential for explicit awareness of the sequence. Using an accuracy measure to examine learning showed that PD participants were able to learn the sequence both with and without the distractor present. Additionally, participants were able to transfer this sequence learning to the opposite hand, showing that learning was not specific to one hand, and thus was not effector specific. Another study examined explicit SRT learning through the accuracy measure in healthy controls and people with PD (H&Y 1.5 – 2.5) when they were ON and OFF medication, and found learning-related activation in the ventral putamen in early learning in PD participants when they were OFF medication (Kwak, Muller, Bohnen, Dayalu, & Seidler, 2012). Thus, despite being tested OFF medication in a task with a large motor component, and therefore
having to cope with disease-related motor impairments without the control of medication, people with PD who were OFF medication were still able to show learning in both explicit and implicit versions of the SRT.

One study used a second-order SRT task to compare learning in young and older healthy older adults to that in people with Parkinson’s disease (H&Y 2 – 3; ON medication; Schendan, Tinaz, Maher, & Stern, 2013). Learning was implicit for the older and PD groups, while some young adults were aware of the sequence. Results showed that young adults had more higher order sequence learning than both healthy older controls and people with PD; although both older adults and people with PD learned some associations within the sequence, controls learned slightly more than the PD group. During the task, both older adults and people with PD had more overall activation in the dorsolateral prefrontal cortex, medial temporal lobe and basal ganglia; there was more task-related activation in the two older groups, but these areas also showed learning-related reductions in activation in healthy older adults, with even greater reductions in the PD group. Because young adults had explicit awareness of the sequence that older adults and people with PD did not have, the task was different for the young adults than the two older adult groups, which may explain why there was more prefrontal and medial temporal lobe activation in young adults. This study adds to the two above by showing that people with PD who were ON medication were able to show sequence learning. Learning in this study was measured through reaction time, though, so it is also possible that the motor requirements made it a more difficult task for people with PD, which may have reduced their ability to learn, or at least show learning, to the same extent as healthy older controls.

**Mixed findings of sequence learning in Parkinson’s disease.** As can be seen from the studies reviewed, above, studies examining sequence learning in people with PD have shown mixed
results, and this may be due to a number of reasons. First, *Hoehn and Yahr stage* (1967) is often not controlled for or taken into account when investigating learning in PD participants (Deroost, Kerckhofs, Coene, Wijnants, & Soetens, 2006). Deroost and colleagues examined SRT learning (implicitness unknown) in a group of medicated PD participants all in Hoehn and Yahr stage 3 compared to healthy controls, and found that, even in a group assumed to be relatively homogenous, PD participants could be divided into subgroups of fast and slow performers. Fast PD participants learned the sequence as well as healthy controls, while slow PD participants did not learn the sequence at all. The two PD groups also had significantly different scores on the Scales for Outcomes of Parkinson’s disease – Cognition (SCOPA-COG; Marinus et al., 2003), and these scores for both the overall test and the Learning and Memory portion, alone, were positively related to the amount of learning in the SRT (Deroost et al., 2006). Thus, even within this group of participants in the same Hoehn and Yahr stage, people differed in their levels of sequence learning and PD-related cognition, with performance on these two tasks being related.

A second study also investigated participants in Hoehn and Yahr stage 3, but divided participants based on their scores on the SCOPA-COG into low, average and high scoring (Vandenbossche, Deroost, Soetens, & Kerckhofs, 2009). This study found that people who were high and average scoring showed significantly more implicit learning in the SRT than people who were low scoring, and this difference remained even after controlling for disease duration. The results of these two studies show a relationship between SRT learning and scores on the SCOPA-COG, such that people with better cognitive scores had more sequence learning.

A second possible reason for discrepancies in results of sequence learning studies in people with PD is the *medication status* of participants; that is, whether they are tested while they are ON or OFF of their medication, or whether or not they ever have been medicated, i.e.,
are medication naïve (Kwak et al., 2013; Muslimovic, Post, Speelman, & Schmand, 2007). In a set of motor tasks looking at baseline motor reaching and explicit sequence learning through motor reaching in people with PD when they were ON and OFF medication (H&Y M = 2.3), sequence learning was the same regardless of medication status (Argyelan et al., 2008). A difference emerged, however, when the group was split into good and poor learners as determined from performance OFF medication; being ON medication impaired learning in good learners, but improved learning in poor learners. Thus, these results supported the overdose hypothesis, such that medication state interacted with the amount of sequence learning, with medication improving learning in poor learners, and impairing learning in good learners.

Another study examined implicit learning in a SRT-like task, testing people with PD who were either ON medication or were medication naïve (H&Y 1-3; Stephan, Meier, Weber Zaugg, & Kaelin-Lang, 2011). While this study did not examine learning in these two groups of participants separately, results showed that worse learning was related to higher Hoehn and Yahr stages, higher ratings on the UPDRS motor section, and higher levels of medication. Not only did measures of disease severity (Hoehn and Yahr stage and UPDRS) affect sequence learning, but dopaminergic medication was also related, and therefore may have affected learning. Thus, medication levels should be considered when examining sequence learning in people with PD who are medicated, and if medication naïve participants are included, they should be examined separately, as well.

Another study examined PD (H&Y 1-3) participants both ON and OFF medication in an explicit version of the SRT (Kwak et al., 2013). Dopamine denervation was also measured in participants, and results showed a dissociation of the effects of dopamine medication on learning based on the level of dopamine denervation in the putamen. Consistent with the overdose
hypothesis, the more dopamine denervation a participant had (i.e., less dopamine) in the putamen, the more dopaminergic medication improved learning, whereas participants who had less dopamine denervation showed negative effects of dopamine medication on learning in the SRT. Therefore, this study adds to the findings of Argyelan et al. (2008) and Stephan et al. (2011), highlighting the importance of not only considering disease severity, here measured by dopamine denervation, but also understanding how dopaminergic medication can affect individuals differently, and thus affect task performance in different ways.

The importance of the interaction between disease severity and medication was additionally shown in a study examining SRT learning (unknown implicitness) in people with PD (H&Y 1-3) ON medication and people with PD who were medication naïve (Muslimovic et al., 2007). This study found that PD participants who were medication naïve learned as much as controls, while people with PD who were ON medication had less sequence learning than controls. Additionally, there were marginal negative correlations between SRT learning and Hoehn and Yahr stage and UPDRS motor scores, suggesting that people who were more impaired by PD had worse learning, and dopaminergic medication did not alleviate these deficits in people with PD who were ON medication, or at least not enough to make learning similar to that in controls.

The effects of two different types of PD treatment on learning were examined in participants both ON and OFF each respective type of treatment (Carbon et al., 2003). Participants were either treated with deep brain stimulation (H&Y M = 3.2) at the globus pallidus internal, or with intravenous dopaminergic medication (H&Y M = 2.0). In an explicit sequenced reaching task, results showed that in people who were treated with deep brain stimulation, being ON treatment improved target retrieval compared to when participants were OFF their treatment.
In contrast, in people who received dopaminergic medication, being ON actually impaired target retrieval compared to when they were OFF medication. Neuroimaging results showed that deep brain stimulation was associated with increased brain activation during the task, while being ON dopaminergic medication was associated with less activation during the task compared to being OFF medication. The authors suggested that this dissociation between how the two treatment types affected learning was due to the different ways in which the treatments increased dopamine levels. Dopaminergic medication increases tonic levels of dopamine in the striatum, while deep brain stimulation increases phasic bursts of dopamine, the type of dopaminergic delivery that is necessary for new learning of associations, as previously mentioned from Frank’s (2005) model.

The studies reviewed above, converge to suggest the importance of taking into account differences within PD groups, not only characteristics of disease severity such as Hoehn and Yahr stage or disease duration, but also medication levels and performance differences on neuropsychological or other behavioral tests.

**Potential problems with using the SRT in Parkinson’s disease.** As previously mentioned, much of the sequence learning literature in people with PD uses the Serial Reaction Time task, and as reviewed above, results are mixed. At least some of the inconsistency regarding whether implicit sequence learning is impaired in PD is likely due to limits imposed by the SRT task itself.

First, the *SRT involves explicit as well as implicit learning*; SRT participants often become aware of the pattern, making the task more explicit, and thus less dependent on striatal function. Thus, any PD-related deficit in *implicit* learning may be masked if PD participants are able to compensate via explicit forms of learning. Second, the *SRT makes strong motor sequencing demands*; participants must respond as quickly as possible to every event. Thus, any
apparent learning deficits might reflect motor and coordination deficits in people with PD, rather than effects on learning itself.

Finally, and perhaps most important, typically the SRT enables assessing sequence learning at only one, or a very few, points in training. This is problematic because interactive-systems theories (e.g., Henke, 2010; Poldrack et al., 2001) suggest that two learning systems underlie implicit sequence learning, and while both are likely operating throughout training, their relative importance changes as training progresses; a MTL-based system dominates early in training and a striatal system later.

**The Triplets Learning Task with PD participants.** The TLT is an implicit sequence learning task developed in our lab that measures implicit probabilistic sequence learning (Howard Jr. et al., 2008). Figure 1 shows a schematic of the task. Typically, there is a second-order learning structure, such that the location of the first event (first red cue) predicts the location of the third event, the green target, and the second red cue occurs in a random location (r). This occurs in a probabilistic manner, such that 80% of the time the first red cue predicts the location of a high probability target, in Figure 1, the first cue in location 3 predicts the green target in location 1, making 3r1 a High Probability triplet. The remaining 20% of the time, the first cue in location 3 predicts the green target in location 2, 3 or 4, such that 3r2, 3r3 and 3r4 are all Low Probability triplets. Because of the probabilistic nature of the triplets and their random occurrence throughout training, participants do not become aware of the regularity, or even that there is a regularity.

The TLT overcomes some of the limitations in earlier studies with PD in the SRT. For example, sensitive measures are used to assess explicit awareness of the learned pattern, and people do not become aware of the pattern even after extended practice. Additionally, the motor
component is reduced, and in fact there is no motor sequence to be learned, because participants respond to only the last (target) event in each trial. Another advantage is that the TLT enables trial-by-trial tracking of learning, so that it is possible to determine the time course of any effects, e.g., whether PD participants differ more from controls in late than early training, and whether there are group differences within blocks. Thus, the TLT is a useful tool for studying implicit learning in people with Parkinson’s disease.

Data from three studies in our lab converge to show that early implicit sequence learning relies primarily on the hippocampus (Bennett et al., 2011; Simon, Vaidya, Howard Jr., & Howard, 2011), while later learning is related to individual differences in dopamine levels (Simon, Stollstorff et al., 2011) and relies on the caudate (Bennett et al., 2011; Simon, Vaidya et al., 2011). Because the putamen, which is part of the motor-corticostratal loop (Seger, 2006), is the most affected region of the striatum (Kish et al., 1988), a motor-based sequencing task, such as the SRT, may call more on the putamen, which may mask intact sequencing that may rely on the caudate, where there is less dopamine denervation. Therefore, because late learning in the TLT relies on the caudate, in both young and older adults (Bennett et al., 2011; Simon et al., 2012), it is an effective task to examine sequence learning in PD participants, since the caudate is less affected by the disease than the putamen (Kish et al., 1988), and the caudate is part of the executive-corticostratal loop (Seger, 2006), and thus involved in more perceptual than motor-based learning. Having an understanding of how healthy older adults perform in the TLT and what brain regions they rely on for learning allowed us to make predictions about how a group of people with PD would perform and learn.

The TLT has a reduced motor component and controlled timing of events, so that compared to the SRT, it is not as affected by group difference in overall response time. Thus,
the TLT is ideal for studying age-related differences in learning, as older adults have slower processing speed compared to young adults (Salthouse, 1996). This same reasoning makes the TLT an ideal task to compare learning in people with PD to healthy controls, as people with PD have slower processing speed than healthy older adults (Cooper et al., 1994).

The above review of learning in the SRT and a sequenced reaching task show the importance of considering differences within PD groups, not only diagnostic criteria such as Hoehn and Yahr stage, UPDRS scores or disease duration, but also medication levels and performance on neuropsychological or other behavioral tests. Given our knowledge of some the neural bases involved in particular points in training, the TLT is well suited to examine such individual differences in people with Parkinson’s disease. These studies add to the literature using other striatal-based tasks in PD participants, such as probabilistic classification, which generally involves explicit instructions and feedback; while learned associations in probabilistic classification may occur as a result of striatal dopamine bursts, learning is generally done explicitly, and associations are guided by explicit positive or negative feedback.

**The present studies**

In the two present studies, the Triplets Learning Task was used to measure implicit sequence learning in a group of people who had mild to moderate PD (Hoehn and Yahr stages 1-2.5) who were ON medication. Learning in the PD group was compared to a group of age- and education-matched healthy older adults. The TLT was an ideal task for this investigation because learning is related to dopamine levels (Simon, Stollstorff et al., 2011), the neurotransmitter most affected in PD (Kish et al., 1988), and late learning most optimally relies on the caudate (Bennett et al., 2011; Simon, Vaidya et al., 2011), a region of the brain affected...
by PD, particularly as the disease progresses (Kish et al., 1988), such that the TLT is a well understood task that relies on specific factors that decline in PD.

In Study 1, we examined whether people with Parkinson’s disease were able to learn implicit sequences in the TLT, and if so, how this learning compared to a group of age- and education-matched healthy controls. We examined group differences in various measures of learning, as well as differences in other aspects of TLT performance. We asked the following questions:

1) Can people with PD learn sequences implicitly in the TLT? The TLT has a much reduced motor component compared to the SRT, the task most studies use to examine implicit sequence learning in PD, so when this motor component is removed, can learning still occur? We predicted that people with PD would learn.

2) Does sequence learning in the TLT (Triplet type effect and AL scores) differ between PD and a healthy Control group? We know that sequence learning in the TLT is positively related to higher levels of dopamine availability in healthy young adults (Simon, Stollstorff et al., 2011), and healthy older adults have higher levels of dopamine than age-matched people with Parkinson’s disease (Kish et al., 1992), so we predicted that there would be better implicit sequence learning in the Control group than the PD group.

3) Do differences in learning between the Control and PD groups occur at points in training that are known to rely on the striatum? That is, can these analyses help us to better understand the neural bases of implicit sequence learning by testing a group of individuals who have known striatal deficits?

a. Given that there are larger dopamine declines in the striatum in people with PD than healthy older adults (Kish et al., 1992), we predicted less learning in the PD than the
Control group, particularly toward the end of training when the caudate becomes more involved in learning (Bennett et al., 2011; Simon et al., 2012).

b. Using the Half Block analysis, where learning in the 1\textsuperscript{st} Halves of Blocks has been shown to be impaired in people with MCI, and thus is thought to rely on the hippocampus, and the 2\textsuperscript{nd} Halves of Blocks on the striatum (Nemeth et al., 2013), we predicted that people with PD would show less learning in the 2\textsuperscript{nd} Halves of Blocks than in the 1\textsuperscript{st} Halves. We also predicted that there would be a larger difference between the PD and Control groups in the 2\textsuperscript{nd} than in the 1\textsuperscript{st} Halves of Blocks.

4) Do people with PD have more intraindividual variability (individual variability within a task) than Controls? Because people with PD have been shown to have more variability in their response time than healthy controls (Cooper et al., 1994), and because neural noise or dysfunction (i.e., dopamine denervation) is thought to cause more intraindividual variability (de Frias, Dixon, Fisher, & Camicioli, 2007), we predicted that people with PD would have more intraindividual variability than healthy controls.

There is a great deal of heterogeneity among people with PD, so in Study 2, we examined how individual characteristics, both disease-related and scores on neuropsychological tests, related to sequence learning. We ran a series of correlations on measures of disease severity, motor impairments, cognition and learning (Table 3), and asked the following questions:

1) Are disease-related characteristics (motor impairment, disease severity) related to implicit sequence learning in the TLT? We examined whether measures of motor deficits (Hoehn and Yahr stage and UPDRS scores), as well as other disease-related characteristics (disease duration, Levodopa and Levodopa Equivalence), which we used as proxies for disease severity, were related to one another and/or to learning.
a. We predicted that these characteristics would be positively related to one another, as they are all related to disease progression. We based this prediction on the idea that as the disease progresses, more dopamine denervation occurs, such that worse scores or levels of each of these variables should suggest that there is less striatal dopamine.

b. We predicted that greater motor impairment (higher Hoehn and Yahr stage and UPDRS ratings) would be negatively related to implicit sequence learning (Stephan et al., 2011) in the TLT. We also predicted that higher medication levels would help moderate this effect, such that higher medication levels of Levodopa and Levodopa Equivalence would be related to more learning. Although it has been shown that sequence learning in the SRT is worse in people when they are ON compared to OFF medication (Kwak et al., 2012; Muslimovic et al., 2007), learning was explicit in Kwak et al. and had unknown implicitness in Muslimovic et al., and the tasks contained a large motor component, so we predicted anti-Parkinsonian medication to have different, and positive effects on our implicit sequence learning which relies on the caudate (Bennett et al., 2011; Simon et al., 2012).

2) Does reaction time on the Grooved Pegboard correlate positively with medication levels? Implicit learning? The Grooved Pegboard, explained in more detail below, measures fine motor movement and skills, and reaction time has been shown to relate to levels of dopamine denervation in the striatum (Bohnen, Kuwabara, Constantine, Mathis, & Moore, 2007). Based on these findings, we predicted a positive correlation between RT on the Grooved Pegboard and 1) Levodopa, and 2) Levodopa equivalence levels, and 3) the same relationships to learning as those described above with disease-related characteristics.
3) Do scores on the SCOPA-COG, a measure of cognition in people with PD, positively correlate with implicit sequence learning? Based on previous findings showing a relationship between scores on the SCOPA-COG overall and the SCOPA-COG Learning and Memory section (Marinus et al., 2003), we predicted a positive correlation between Triplet type learning and cognition scores on these two scales.

4) Do disease-related characteristics relate to intraindividual variability in people with Parkinson’s disease? Intraindividual variability can increase as a result of dysfunction or neural noise in a system, such as declines in dopamine that, through inconsistent functioning, may add variability to a motor system and its functions. Therefore, in the PD group, there is dysfunction of the dopaminergic system as well as variability of dopamine levels within participants due to dopaminergic medication fluctuations, suggesting that medication levels should be positively related to intraindividual variability. That is, increased medication levels suggest an overall larger decrease in dopamine cells, so there is a wider range of dopamine levels within the brain, from being as close to an OFF medication state as participants may be (e.g., just before taking their next medication dose) to the most ON state, when medication (dopamine) is at its optimal level. This lack of consistency in dopamine levels likely introduces more variability into the system, both at the level of the striatum, and regions of the brain that are innervated by the striatum, such as the motor cortex. Therefore, we predict that measures of disease severity, Hoehn and Yahr stage, UPDRS, disease duration and medication levels, will be positively related to intraindividual variability.
CHAPTER 2: METHOD

Participants

Parkinson’s disease. Thirty-one people with idiopathic Parkinson’s disease, as diagnosed by a clinical neurologist, participated in this study. Parkinson’s disease (PD) participants were recruited for or referred to our study by their neurologist (Dr. Steven Lo) or psychiatrist (Dr. Thomas Cummings). All participants were in mild to moderate stages of the disease (Hoehn and Yahr stage 1-2.5; Hoehn & Yahr, 1967), and all were taking medication for their Parkinson’s symptoms at the time of testing. While Levodopa is the most widely used medication for treating motor deficits in PD, longer exposure can lead to loss of symptom control, so other medications are often used to help control PD symptoms (Tomlinson et al., 2010). In order to be able to compare medication levels across patients and studies, Levodopa equivalence can be calculated using daily doses of various medications. Tomlinson and colleagues investigated various versions of the Levodopa equivalence and combined them to create a standard formula.

Here, Levodopa equivalence = (Levodopa mg x 1) + (Levodopa mg extended release x .75) + (Ritogotine mg x 30) + (Selegiline mg x 10) + (Entacapone mg x 0.33) + (Pramipexole/Mirapex mg x 100) + (Amantadine mg x 1) + (Rasagiline/Azilect mg x 100) + (Ropinirole mg x 20). All measures were taken and testing was done while PD participants were ON medication.

The same group of PD participants was used for analyses in both studies reported here. Study 2 examined only PD participants, using disease-related characteristics to more closely examine learning within this group. Exclusion criteria for participants varied by study, whether PD participants were being compared to one another (Study 2) or to a healthy control group (Study 1). In Study 2, one PD participant scored below the cutoff for the MMSE (score of 21), and one had an average reaction time greater than 2.5 standard deviations from that of the group.
mean; these two participants were removed from all data analyses. The remaining 29 participants had a mean age of 64.55 (SD = 5.77), and there were 17 males and 12 females.

Individual participant characteristics can be seen in Table 1.

Of the 29 PD participants, two participants fell below the cutoff for the MoCA used for Controls (details below), thus, 27 PD participants were used as a comparison for Controls in Study 1 (M age: 64.59, SD = 5.75, ten females).

**Healthy controls.** Thirty healthy older adults (M age = 66.47, SD = 5.32, 20 females) served as Controls in Study 1. An additional ten participants were tested and removed from analyses due to low cognitive scores (MoCA < 24, MMSE < 27), low verbal intelligence scores (NAART-35, a score well below that of the normed values for people of a particular age and education level; Uttl, 2002), or a history of memory loss or antipsychotic drug use. Controls were recruited to be age- and education-matched to our sample of PD participants. Additional participant characteristics and comparisons can be found in Table 2.

All participants received monetary compensation for their participation, and all methods were approved by the Georgetown University Institutional Review Board.

**Tasks**

**Triplets Learning Task.** A schematic of the primary task, the Triplets Learning Task (TLT), can be seen in Figure 1. In the TLT, participants saw four evenly spaced open circles in the middle of a computer screen (Howard Jr. et al., 2008). Each trial consisted of three successively presented events, two cues and a target, which together made a *triplet*. The first cue consisted of one of the four circles filling in red, followed by a second cue (the same or a different circle), also filling in red. The target, to which participants were to respond, consisted of a third circle filled in green. Each of the two red cues remained filled in for 120 ms followed by a 50 ms
interstimulus interval. The third stimulus, the green target, remained lit until the participant responded correctly. Participants responded to targets by using their left and right middle and index fingers to press keys, “z,” “x,” “.”, and “/” on a standard keyboard in response to the first, second, third, or fourth circles, respectively. In case using two hands was too challenging, Parkinson’s disease participants were also offered the opportunity to use a stimulus-response box with whichever hand was most comfortable for them, with four keys across corresponding to the four circles. Twenty-five PD participants used the keyboard, and four used the SR box. There were 50 trials per block, 10 blocks per session, and three sessions for each participant, all completed in a single visit to the laboratory. Training was also calculated into blocks of time, or epochs, which consisted of 5 blocks per epoch, totaling six epochs for all of training.

The triplet sequence contained a second-order regularity, in that the location of the first red cue predicted the location of the target (the green event), with the location of the second red cue being random (Howard Jr. et al., 2008). Referring to the circles as 1, 2, 3 and 4 from left to right, one possible pattern is 1r2r3r4r, where r is one of the four circles chosen randomly. Participants receiving this pattern would see the high frequency triplets 1r2, 2r3, 3r4, and 4r1 on 80% of the trials, and the low frequency triplets (e.g., 1r3, 2r4) on the remaining 20% of trials. There were six unique triplet patterns counterbalanced across participants (1r2r3r4r, 1r2r4r3r, 1r3r2r4r, 1r3r4r2r, 1r4r2r3r, and 1r4r3r2r), so that a given triplet (e.g., 1r4) was high frequency for some participants, but low for others. With four possible positions and three circles being lit per trial, there are 64 possible triplets. Thus, for each pattern, there were 16 possible high frequency triplets, and 48 possible low frequency triplets\(^1\).

\(^1\) Unlike many of our previous studies with implicit sequence learning where we removed some types of Low probability triplets (Repetitions and Trills), we have not done so in the current studies in order to make them more comparable to the data in Nemeth et al. (2013) for the Half Block analyses. We examined learning with Repetitions and Trills removed, however, and the pattern of results remained the same.
End of block accuracy and reaction time feedback was given to direct participants to achieve 92% accuracy, with the aim of matching the groups on overall accuracy. Participants were told to focus more on speed if their accuracy was ≥ 94%, or to focus more on accuracy if it was ≤ 90%.

**TLT Recognition task.** In the TLT Recognition task following the three sessions of the TLT, participants were shown each of the 64 possible triplets on the computer screen, presented with the same timing as during the TLT. Participants were asked to observe each triplet without responding to the target, and then to report via a keypress if they thought each individual triplet occurred “more often” or “less often” during training.

**Grooved Pegboard Test.** The Grooved Pegboard Test (Lafayette Instruments, Lafayette, IN) uses a small metal board with grooved holes in it that requires participants to manipulate the direction of a grooved peg to fit into each hole using one hand at a time. Reaction time to fill all of the holes with pegs and the number of pegs dropped during the task are recorded, so that a higher score indicates poorer performance. This task measures fine motor movement and skills, and can be analyzed by each hand separately, allowing analysis of effects of handedness as well as the side of the body where people with Parkinson’s disease are most affected. Additionally, results from one study suggested that the Pegboard test could be a proxy for striatal dopamine denervation in people with Parkinson’s disease (Bohlen et al., 2007).

**Scales for Outcomes of Parkinson’s disease – Cognition.** The Scales for Outcomes of Parkinson’s disease – Cognition (SCOPA-COG; Marinus et al., 2003) is a scale that evaluates cognition specifically in people with Parkinson’s disease. This scale measures four different cognitive skills: learning and memory, attention, executive functions, and visuo-spatial functions. In the present study, we analyzed both the overall SCOPA-COG score, as well as the score for
the learning and memory portion (e.g., verbal recall, backward digit span, pattern recall, delayed recall), as these scores have previously been shown to relate to sequence learning (Deroost et al., 2006). Higher scores on this task indicate better cognitive function. This scale can be found in the Appendix.

**Mini Mental State Examination.** The Mini Mental State Examination (MMSE) is one of the most widely used tools for assessing cognitive impairment in older adults (Tombaugh & McIntyre, 1992), as well as in people with Parkinson’s disease (Hoops et al., 2009). MMSE scores (a higher score indicates less impairment) have been shown to correlate with the Wechsler Adult Intelligence Scale (Folstein, Folstein, & McHugh, 1975). This test can be found in the Appendix.

**Montreal Cognitive Assessment.** The Montreal Cognitive Assessment (MoCA) was created as a more sensitive measure for detecting Mild Cognitive Impairment (MCI) and Alzheimer’s disease (AD), in light of findings that the widely used MMSE (Tombaugh & McIntyre, 1992) often did not detect such impairments (Nasreddine et al., 2005). Nasreddine and colleagues compared scores on the MoCA and MMSE to outcomes of a full neuropsychological evaluation in people with AD, MCI, and a comparison group of healthy older adults, and found the MoCA to be more sensitive in detecting MCI and AD than the MMSE. Similar methods were used with groups of people who had PD or MCI, and the MoCA was again found to be a more sensitive screening tool than the MMSE for MCI, and for dementia in people with Parkinson’s disease (Hoops et al., 2009). The MoCA can be found in the Appendix.

**Geriatric Depression Scale.** The Geriatric Depression Scale (GDS) is a self-report measure that examines depression levels in older adults. It contains questions such as, “Are you basically satisfied with your life,” and, “Do you often feel helpless,” to which people give ‘yes’ or ‘no’
answers, with a higher score indicating higher levels of depression. The GDS has been shown to be valid and reliable, and is able to accurately discriminate between non-depressed, mildly depressed, and severely depressed individuals (Yesavage et al., 1983). The present studies used a short 15 item version of the GDS which has been shown to be a reliable measure of depression, also able to differentiate between non-depressed, mildly, moderate, and severely depressed older adults (Almeida & Almeida, 1999). This scale can be found in the Appendix.

**WAIS-III Forward and Backward Digit Span.** The Wechsler Adult Intelligence Scale-III (WAIS-III) Forward and Backward Digit Span tasks were administered (Wechsler, 1997). Forward Digit Span (DSF) is a measure of short-term memory, while Backward Digit Span (DSB) is a measure of working memory.

**North American Adult Reading Test-35.** The North American Adult Reading Test (NAART) is a measure of verbal intelligence, which consists of both long and short versions, with the NAART-35 being a 35 item shortened version (Uttl, 2002). This test consists of irregularly pronounced English words, and is indicative of reading abilities. The NAART-35 has been shown to be highly correlated with scores on the Wechsler Adult Intelligence Scale-R Vocabulary test, which is a widely used measure of verbal IQ, as well as to have the same level of validity as the full length NAART. Uttl determined normative scores for various age groups across the lifespan, taking into consideration levels of education fewer or more than 16 years. We used these norms as a guideline for removing participants who scored well below the suggested cutoff at a particular age and education level. The NAART is scored in reverse, such that the score indicates the number of items that are incorrect. This test can be found in the Appendix.
Perceived Stress Scale. The Perceived Stress Scale (PSS) is a self-report measure of the extent to which a person has experienced or perceived stress within the past month (Cohen, Kamarck, & Mermelstein, 1983). The PSS examines how participants felt about or responded to stressful events, with a self-report rating of frequency of events such as, “In the last month, how often have you felt that you were unable to control the important things in your life,” with a higher score indicating higher levels of perceived stress. This scale can be found in the Appendix.

Procedure

Participants arrived at the lab and first provided Informed Consent. They then completed health screening and biographical questionnaires (Appendix), as well as the short form of the Geriatric Depression Scale (Brink et al., 1982). They were then read instructions for and completed three sessions of the Triplets Learning Task. They were told that they would see four open circles, and that two circles would first fill in red, and then one of the circles would fill in green, and that they were to respond to the location of the green target using the corresponding keypress. There were 10 blocks in a session, and participants were told they could take short breaks between blocks, but asked to not leave the room, and then were allowed longer breaks between each of the sessions, where they were allowed to get up and walk around. Participants took an average break of 90 seconds between blocks (range: 35 – 119 seconds). Following the third session, participants’ explicit knowledge was tested in two ways: first, through the TLT Recognition task, and second, a verbal questionnaire probing strategies and their ability to reproduce the learned triplets (Appendix).

Following the Triplets questionnaire, participants completed the Montreal Cognitive Assessment (Nasreddine et al., 2005), the Grooved Pegboard Test (Lafayette Instruments, Lafayette, IN), the Scales for Outcomes of Parkinson’s disease – Cognition (SCOPA-COG;
Marinus et al., 2003), the Mini Mental State Examination (Folstein et al., 1975), the Wechsler Adult Intelligence Scale-III Forward Digit Span (Wechsler, 1997), the North American Adult Reading Test-35 (Uttl, 2002), and the Perceived Stress Scale (Cohen et al., 1983), in that order. Control participants were not tested on the SCOPA-COG, as it is a test specific to cognition in people with Parkinson’s disease. The entire day of test lasted approximately two and a half hours.

Finally, participants were debriefed and compensated for their time.
CHAPTER 3: STUDY 1 RESULTS AND DISCUSSION

In this study, we examined differences in implicit sequence learning in the Triplets Learning Task between a group of people with Parkinson’s disease and a matched group of healthy Controls. We compared learning through accuracy Triplet type effects (accuracy to High probability triplets minus Low probability triplets), reaction time Triplet type effects (reaction time to Low probability triplets minus High probability triplets), and associative learning measures, and we also examined group differences in intraindividual variability.

One of the PD participants chose to not complete the third session of the TLT, and one participant responded incorrectly (used one hand on the keyboard) in the first session of the TLT, so these data were excluded from the below analyses, but the remaining two session from these participants are included.

**Participant characteristics**

A comparison of Group characteristics and differences can be seen in Table 2. Groups did not differ in Age or Education, two characteristics on which they were matched, or on self-reported health or stress, Forward Digit Span or the NAART-35. Groups showed significant differences in Backward Digit Span, though their ranges of scores were similar (Controls: 5 – 13, PD, 3 – 11). They also differed in Geriatric Depression Scores, with people in the PD group having higher depression scores, though all but two participants in each group had scores below 5, which is a suggested score for non-depressed individuals to seek treatment (Brink et al., 1982). There were also significant group differences in the MoCA and MMSE, though both groups scored well above the cutoff for each of these, suggesting that these differences are likely not indicative of differences in cognitive functioning across groups.
TLT Recognition task

To assess potential awareness of the frequency of occurrence of the triplets during training, we first examined ratings on the TLT Recognition task that participants had completed at the end of training. For each participant, we calculated the proportion of triplets within each Triplet type that were rated as having “occurred more often.” A 2 x 2 mixed-design (Group: PD vs. Controls) x 2 (Triplet type: High probability vs. Low probability) ANOVA, with group varying between-subjects and triplet type within-subjects, yielded no significant effects ($p$’s > .10); participants did not rate High probability triplets as having occurred more frequently than Low probability triplets, and there was no difference in ratings between the two groups (Figure 2). These results suggest that participants were not aware of the triplet frequencies seen during training, and thus any learning that occurred was largely implicit. The post-Triplets verbal questionnaires also gave no indication that participants had any awareness of the pattern they had learned\(^2\).

TLT Accuracy

Accuracy on the TLT task was analyzed to examine if people responded more accurately to High than to Low probabilities, which would indicate learning about the Triplet type frequencies. Accuracy was measured by determining the proportion of trials that were correct for each epoch for each participant, and these data are displayed in Figure 3. A 2 (Group) x 6 (Epoch) x 2 (Triplet type) mixed-design ANOVA on accuracy showed no significant main effects or interactions, $p$’s > .10, such that there was no learning seen through the accuracy measure in either group, nor were there group differences in sequence learning or overall

\(^2\) One PD participant made numerous guesses about the relationship between the red and green cues in the pattern. Two guesses made followed the pattern (1r3 and 3r2), but this person also guessed incorrectly for each of these predictive cues (1r2 and 1r4), suggesting a lack of explicit knowledge. Additionally, this participant rated High probability triplets as having occurred with a .56 probability and Low probability triplets with a .63 probability, giving additional support to our conclusion that this person was not actually explicitly aware.
accuracy. The lack of learning seen in this measure is likely due to a ceiling effect, such that participants in both groups were highly accurate (PD: $M = 0.963$, $SD = 0.025$, Controls: $M = 0.964$, $SD = 0.023$).

**TLT Mean of Median Reaction Time**

We next examined Triplet type learning using a reaction time measure. Because accuracy did not differ significantly between groups, we could analyze reaction time without being concerned about a speed-accuracy tradeoff in one group but not the other. For each participant and each triplet type, we calculated median RTs separately for each Triplet type in each block of 50 trials, and then averaged these medians within an epoch to calculate the mean of median reaction time for each epoch. These data are displayed in Figure 4. A 2 x 6 x 2 mixed-design ANOVA yielded significant main effects of Epoch ($F(5,265) = 23.46, p < .001$) and a marginal Group x Epoch interaction ($F(5,265) = 2.14, p = .061$), such that reaction time decreased with training, and more so for the Control than the PD group.

Most important, there was also a significant effect of Triplet type ($F(1,53) = 117.11, p < .001$), with RT being faster to High than to Low probability triplets, as well as a significant interaction of Group x Triplet type ($F(1,53) = 4.56, p = .037$), such that there was a larger Triplet type effect for the Control group ($M = 21.65$, $SD = 14.66$) than for the PD group ($M = 17.22$, $SD = 12.81$). There were no other significant main effects or interactions, $p > .10$. This ANOVA indicated that, as predicted, the Control group learned more about Triplet type probabilities than the PD group.

To determine whether each group showed learning, we ran follow-up 6 (Epoch) x 2 (Triplet type) ANOVAs on each group separately. Both groups revealed significant main effects of Epoch (Controls: $F(5,145) = 28.10, p < .001$, PD: $F(5,120) = 4.22, p = .002$) and Triplet type
(Controls: $F(1,29) = 64.05, p < .001$, PD: $F(1,24) = 74.17, p < .001$), and neither group had a significant Epoch x Triplet type interaction ($p$’s > .10). Thus, people in the PD group had significant skill (Epoch main effect) and Triplet type learning, showing that they were able to learn in the TLT, though, as was seen in the overall ANOVA on RT, and consistent with our predictions, they learned less about triplet frequencies than the Control group. The fact that the PD group did not have significantly slower RTs than Controls (Group x Epoch was marginal, Group main effect was > .10), suggested that the smaller Triplet type effect seen in the PD group was not due to motor deficits, which would be manifest in slower response times.

**TLT Associative Learning scores**

Learning was next assessed using Associative Learning (AL) scores, which take into account individual differences in response time and variability and have been used in previous studies with the TLT (Gamble, Howard Jr., & Howard, 2014; Howard Jr. et al., 2008; Simon, Howard Jr. et al., 2011). Associative learning scores were calculated for each participant in each epoch by correlating the frequency with which each of the 64 triplets occurred with that individual’s median response time to that triplet. If the person learned, this would yield a negative correlation, such that triplets that occurred more frequently were responded to more quickly. To make learning scores more intuitive, we multiplied them by -1, so that a higher AL score was indicative of more learning. Figure 5 displays these data.

A 2 (Group) x 6 (Epoch) mixed-design ANOVA on the AL scores yielded only a significant main effect of Group ($F(1,53) = 7.08, p = .010$; as predicted, the Control group had higher AL scores than the PD group. This group difference did not change significantly with training in that the Group x Epoch interaction was not significant. However, based on our *a priori* hypothesis that people with PD would show less learning than the Control group at the end
of training, we ran unpaired t-tests for AL scores at each epoch to compare the two groups. There were no differences between groups in Epochs 1-5 ($p$’s > .10), but there was a marginal difference in Epoch 6, $t(54) = 1.88$, $p = .066$. This pattern suggested that the group difference in AL score was being carried by the last epoch, providing tentative support for our prediction that group differences would increase as training continued and the caudate became more important to support learning (Simon et al., 2012)

**TLT Half Block comparison**

As mentioned in the Introduction, a recent paper from Nemeth and colleagues (2013) compared learning in the ASRT in a healthy group of older adults to a group with Mild Cognitive Impairment (MCI) who are known to have hippocampal deficits. They compared learning in the 1st Half of ASRT Blocks (first 40 of 80 trials/block) to learning in the 2nd Half of Blocks (second 40 of 80 trials/block), with the hypothesis that the 1st Half of Blocks involve recall and reactivation of the learned sequence, processes that rely on the hippocampus, while the 2nd Half of Blocks reflect habitual forms of behavior and stimulus-response learning, processes that involve the striatum. Their results supported their theory, finding impaired learning in the 1st Half of Blocks in people with MCI compared to healthy controls, but equal amounts of learning between the two groups in the 2nd Half of Blocks. We reasoned that if their interpretation of this difference was correct, then using this same analysis with the TLT, we should see the opposite pattern in people with Parkinson’s disease. That is, Parkinson’s participants’ learning should not differ from Controls in the 1st Half of Blocks, but should be poorer than Controls in the 2nd Half of Blocks.

Figure 6 shows the mean of median response times broken down by Triplet type and Half Block within each epoch, with the Control group on the left and the PD group on the right. A 2
(Group) x 2 (Half Block) x 6 (Epoch) x 2 (Triplet type) ANOVA on these RTs showed significant main effects of Epoch \((F(5,265) = 23.02, p < .001)\) and Triplet type \((F(1,53) = 166.11, p < .001)\), and a significant Group x Triplet type interaction \((F(1,53) = 4.71, p = .034)\), in keeping with the ANOVA on triplet type RTs reported above.

Of more relevance here, however, are effects involving Half Block. There was a main effect of Half Block \((F(1,53) = 52.93, p < .001)\), and a significant interaction of Half Block x Epoch \((F(5,265) = 9.20, p < .001)\). Post hoc paired t-tests showed that there was a marginal difference between Half Block RTs in Epoch 1 \((t(1.75(55), p = .087))\), with RT being faster in the 2\(^{nd}\) Half Block, and significant differences in Epochs 2 – 6 \((p’s < .001)\), with RT in the 1\(^{st}\) Halves of Blocks being faster than in the 2\(^{nd}\) Halves of Blocks. Thus, RTs were initially faster in the 2\(^{nd}\) Halves of Blocks, but reversed in Epoch 2 and remained significantly slower in the 2\(^{nd}\) Halves of Blocks through the rest of training. There was also a Group x Half Block \((F(1,53) = 8.31, p = .006)\) interaction, such that there was a smaller increase in overall RT between the 1\(^{st}\) and the 2\(^{nd}\) Halves of Blocks for the Control group \((M = 6.74, SD = 14.08)\) than the PD group \((M = 19.68, SD = 19.91)\).

Most important, as predicted, there was a significant three-way interaction of Group x Half Block x Triplet type \((F(1,53) = 8.60, p = .005)\). This was qualified by a greater increase in RT in the PD group than Controls from the 1\(^{st}\) Half Block to the 2\(^{nd}\) Half Block for High probability triplets (PD: \(M\) difference = 22.46, \(SD = 19.98\), Control: \(M\) difference = 6.61, \(SD = 15.44\), \(t(55) = -3.37, p = .001\)), but not for Low probability triplets (PD: \(M\) difference = 14.20, \(SD = 18.22\), Control: \(M\) difference = 11.09, \(SD = 18.70\), \(t(55) = -0.64, p > .10\)). As can be seen more clearly in Figures 7 and 8, the result of this differential slowing for High probability triplets in the 2\(^{nd}\) Halves Blocks is that people in the PD group had a smaller Triplet type effect than
Controls in the 2nd Halves of Blocks (PD: $M = 15.61$, $SD = 13.46$, Control: $M = 28.05$, $SD = 21.71$, $t(55) = 2.56$, $p = .013$), but not the 1st Halves of Blocks (PD: $M = 23.87$, $SD = 10.66$, Control: $M = 23.56$, $SD = 13.40$, $t(55) = -.096$, $p = .924$). A comparison between learning in the 1st Half Block and the 2nd Half Block yielded a significant difference between Halves of Blocks in the PD group (1st Half Block: $M = 23.87$, $SD = 10.66$, 2nd Half Block: $M = 15.61$, $SD = 13.46$, $t(26) = 2.66$, $p = .013$), while there was not a significant difference between Halves of Blocks in the Control group (1st Half Block: $M = 25.56$, $SD = 13.40$, 2nd Half Block: $M = 28.05$, $SD = 21.71$, $t(29) = -1.33$, $p = .195$). Thus, people with PD not only showed significantly less learning in the 2nd Halves of Blocks than in the 1st, but the PD group also showed learning equal to the Controls in the 1st Halves of Blocks and significantly less than Controls in the 2nd Halves of Blocks.

These data supported the hypothesis from Nemeth et al. (2013), that the learning revealed in the 1st Halves of Blocks relies upon the hippocampus, while that revealed in the 2nd Halves of Blocks relies upon the striatum. That is, comparing across studies, we see a double dissociation between MCI and PD participants; compared to controls, MCI participants are impaired in the 1st Halves of Blocks, while Parkinson’s participants are impaired on the 2nd Halves.

One possible explanation for the learning difference in the 2nd Halves of Blocks in our PD participants is that they were more fatigued in the 2nd Halves than the Controls. The ANOVA described above did show a significant interaction of Group x Half Block, such that people with PD were overall slower than Controls in the 2nd Halves of Blocks (PD: $M = 546.23$, $SD = 80.91$, Controls: $M = 502.90$, $SD = 82.00$, $t(55) = -2.00$, $p = .050$), but not in the 1st Halves of Blocks (PD: $M = 536.50$, $SD = 79.46$, Controls: $M = 496.16$, $SD = 85.96$, $t(55) = -1.38$, $p = .173$). However, as reported above and can be seen in Figure 6, both groups slowed equally from
the 1st to 2nd block halves for the Low Probability triplets, It is only on the High Probability triplets where the PD group is slowing more. This is not a pattern one would expect if the group difference in 2nd half block Triplet type effects was just due to a general fatigue effect.

In addition, repeated measures ANCOVAs were run analyzing the overall Triplet type effect by Half Block, covarying first overall mean of median reaction time, and second, the mean of median reaction time only in the 2nd Halves of Blocks. Both analyses yielded a significant interaction of Group x Half Block (p’s < .05), suggesting that differences in Triplet type effects between the 1st and 2nd Halves of Blocks were not due to response times, either overall or only in the 2nd Halves of Blocks.

**Comparison of Intraindividual Variability Between Groups**

Intraindividual variability has been shown to be an indicator of neurobiological disturbance (Wegesin & Stern, 2004), and because the literature has shown that people with PD have more variability in their response times than healthy older adults (Burton, Strauss, Hultsch, Moll, & Hunter, 2006; Camicioli, Wieler, de Frias, & Martin, 2008), we examined whether we could replicate this group difference in the current study.

We used two measures to compare intraindividual variability between groups, *individual standard deviation*, and the *coefficient of variation*. As is done in the literature, we removed any trials that were more than three standard deviations from the individual’s mean, and included both accurate and inaccurate trials when conducting the following analyses (Burton et al., 2006).

**Individual standard deviation.** Individual standard deviation measures an individual’s spread of performance (variability) around their own mean performance (Stuss, Murphy, Binns, & Alexander, 2003); for this measure, we calculated the standard deviation for each individual’s reaction times within each epoch; data are shown in Figure 9. A 2 (Group) x 6 (Epoch) ANOVA
on these standard deviations revealed significant main effects of Group ($F(1,53) = 3.89, p = .054$) and Epoch ($F(5,265) = 13.21, p < .001$), and a significant interaction of Group x Epoch ($F(5,265) = 4.79, p < .001$). Individuals in the PD group had greater individual standard deviation than those in the Control group, and this group difference increased with training, with variability decreasing across epochs for the Control group (significant main effect of Epoch, $F(5,145) = 19.49, p < .001$) but not the PD group ($F(5,120) = 1.57, p > .10$). The Group difference was not significant in Epoch 1 ($p > .10$), but was marginal in Epoch 2 ($t(54) = -1.74, p = .088$), and significant from Epoch 3 ($t(55) = -2.21, p = .031$) through the rest of training ($p$’s < .028), qualifying the Group x Epoch interaction.

**Coefficient of variation.** Unlike the individual standard deviation, the coefficient of variation takes into consideration differences in mean performance, by dividing an individual’s standard deviation by their mean (Wegesin & Stern, 2004). Data can be seen in Figure 10. A 2 x 6 ANOVA revealed a marginal main effect of Group ($F(1,53) = 3.574.02, p = .064$), a significant main effect of Epoch ($F(5,265) = 4.01, p = .002$), and a significant interaction of Group x Epoch ($F(5,265) = 2.84, p = .016$). Just as with individual standard deviation, the coefficient of variation showed higher variability in the PD group than the Control group, and this difference increased with training, with variability decreasing across epochs for the Control group (significant main effect of Epoch, $F(5,145) = 6.54, p < .001$), but not the PD group, ($F(5,120) = 1.20, p > .10$). The Group difference was not significant in Epochs 1 or 2 ($p$’s > .10), but was marginal in Epoch 3 ($t(55) = -1.86, p = .068$), and was significant from Epoch 4 ($t(55) = -2.53, p = .014$) through the end of training ($p$’s < .021), qualifying the Group x Epoch interaction.

Together, these intraindividual variability analyses confirmed that people in the PD group had more variable response times than the Controls, and that the groups’ variability changes
differently with practice. In order to determine whether or not group difference in mean reaction time affected these results, we ran ANCOVAS for the two intraindividual variability measures covarying mean of median reaction time, and found that the Group x Epoch interactions were still significant for both analyses ($p$’s < .01). Thus, the PD group was more variable than the Control group by both measures of intraindividual variability, even when reaction time was controlled. To our knowledge, this was the first study to measure intraindividual variability in a sequence learning task in a group of people with Parkinson’s disease. Results from choice and simple reaction time tasks, measured both overall (Camicioli et al., 2008) and over time (Burton et al., 2006), showed increased intraindividual variability in people with PD, so we added to the literature by showing that this variability was greater in people with PD than controls even in a learning task.

**Study 1 Conclusions**

We found that people with PD were able to learn sequences in the TLT implicitly, but that they learned less than the Control group as seen through both the reaction time Triplet type effect and the AL score measures. Even though we did not have the predicted interaction of Group x Epoch x Triplet type with our reaction time, or the Group x Epoch interaction with our AL scores, to further examine our hypothesis of greater group differences in learning at the end of training, t-tests indicated that there was a marginal group difference in Epoch 6, suggesting that the group difference in learning was larger toward the end of training when learning has been shown to rely on the caudate (Simon et al., 2012).

Our Half Block analysis provided stronger support for our general hypothesis that declines in the striatum in people with PD would affect learning at specific periods of training. Our findings converge with those of Nemeth et al. (2013) to better understand the neural bases of
implicit sequence learning; findings that learning is impaired in the 1st Halves of Blocks in people with MCI (Nemeth et al.) and in the 2nd Halves of Blocks in PD (present study) converge to provide support for the theory that the hippocampus underlies learning in the 1st Halves of Blocks and the striatum underlies it in the 2nd Half Block.

Intraindividual variability analyses supported previous research to show that people with PD had more variable response times than healthy controls, indicating that not only is implicit sequence learning worse in people with PD, but overall TLT performance variability was different between the two groups, and these differences could not be explained solely by difference in overall response time. As mentioned above, to our knowledge, this is the first evidence showing more intraindividual variability in people with PD in a sequence learning task compared to healthy controls.
CHAPTER 4: STUDY 2 RESULTS AND DISCUSSION

In this study, we examined whether disease-related characteristics or neuropsychological scores relate to TLT learning in people with Parkinson’s disease. All of these correlations for the PD group can be seen in Table 3, with only those relevant to our questions and hypotheses described here. For all learning comparisons, we examined 1) overall RT Triplet type effect, 2) overall AL score, 3) 1st Half Block RT Triplet type effect, and 4) 2nd Half Block Triplet type effect. Table 4 is an equivalent correlation matrix for Control participants, but given our focus on PD in this study, these will only be referred to as relevant for interpreting the correlations in the PD group.

**Disease severity and learning**

We first examined the relationship among the disease characteristics related to severity and motor impairment, Hoehn and Yahr stage, UPDRS scores, Disease duration, Levodopa and Levodopa Equivalence. As can be seen in Table 3, all of these disease characteristics positively and significantly relate to one another (p’s < .020), with the exception of UPDRS motor scores only relating to Hoehn and Yahr stage (other p’s > .10). This is likely due to the fact that these two scales are based on motor symptoms, while Disease duration and medication levels are likely related to other disease-related deficits, including cognitive ones.

We next examined correlations of Hoehn and Yahr stage and UPDRS scores with our learning measures, and results showed that there were no significant correlations between motor scores and implicit sequence learning (p’s > .10). This lack of a relationship may be due to the fact that these ratings are largely based on more gross motor movements (See the UPDRS motor section (III) in the Appendix) rather than finer motor movements that are required during
sequence learning; thus, these scores may not be indicative of how well a person with PD can perform on, and therefore learn in, the Triplet Learning Task.

In examining other disease-related characteristics, we found significant correlations only with the 2\textsuperscript{nd} Half Block Triplet type effect, with no significant correlations seen with the 1\textsuperscript{st} Half Block ($p$'s $> .10$). Learning in the 2\textsuperscript{nd} Half Block Triplet type effect was positively related to Disease duration ($r = .362$, $p = .046$; Figure 11) and Levodopa ($r = .343$, $p = .068$; Figure 12).

As can be seen in Table 3, there was also a significant correlation between the 2\textsuperscript{nd} Half Block and Age ($r = .427$, $p = .020$; Figure 13). These positive relationships with learning are counterintuitive, so to try to better understand them, we emphasize the relationship between Disease duration and the 2\textsuperscript{nd} Half Block Triplet type effect. The positive relationship with Disease duration indicates that people who have lived with PD longer are better able to pick up probabilistic regularities within the environment. We suggest that this increased ability is indicative of compensatory mechanisms, either at the level of the brain or through behavior. Medication is likely also playing a role in this compensation, as Levodopa was marginally related to learning and significantly related to Age and Disease duration. Additionally, though Levodopa Equivalence was not related to the 2\textsuperscript{nd} Half Block Triplet type effect ($r = 0.174$, $p = .370$), it was significantly related to Age ($r = .455$, $p = .012$) and Disease duration ($r = .667$, $p < .001$). However, it is difficult to draw too strong of implications about the mechanisms causing the positive relationships between these variables and learning, as all of these variables of individual characteristics are positively related to one another (see Table 3; Age x Disease duration: $r = .320$, $p = .091$, Figure 14; Age x Levodopa: $r = .585$, $p < .001$, Figure 15; Disease duration x Levodopa: $r = .569$, $p = .001$, Figure 16), and are likely too highly intertwined that one variable cannot be explained without considering the others.
The correlation between Age and the 2nd Block Half Triplet type effect was not significant in the Control group (Table 4), suggesting that the relationship between Age and learning in the PD group is not reflecting age per se, but is likely the effect of other factors related to Age in the PD group, such as the length a person has had Parkinson’s disease.

**SCOPA-COG and Learning**

Scores on the SCOPA-COG overall, as well as the Memory and Learning section of the Scale have been shown to relate to learning in the SRT (Marinus et al., 2003), so we examined correlations between these scores and our four learning measures of interest. We found that only the Overall reaction time Triplet type effect was marginally related to the overall SCOPA-COG score ($r = .335, p = .075$; Figure 17) and was significantly related to the SCOPA-COG Learning and Memory section ($r = 4.05, p = .028$; Figure 18), while there was no correlation with the other three learning measures ($p’s > .10$). It is interesting that this cognitive measure correlates with Overall Triplet type learning and not learning in the 2nd Half Block, which we found to correlate with measures of disease severity. It is possible that the SCOPA-COG is measuring more global cognition, rather than being more specific to striatal deficits, which we think are being tapped by disease severity measures, and thus SCOPA-COG scores are correlated with a more global measure of overall sequence learning.

**Intraindividual variability and disease related characteristics**

Increased intraindividual variability has been shown to be associated with healthy aging as well as neurological disorders (de Frias et al., 2007), and it has been suggested that the nature of a neurological insult may further affect variability (Burton et al., 2006). Therefore, we examined if intraindividual variability was related to disease-related characteristics, whether motor or disease severity. Individual standard deviation was significantly related to Hoehn and...
Yahr stage ($r = .368, p = .049$; Figure 19), Disease duration ($r = .366, p = .050$; Figure 20), Levodopa ($r = .509, p = .004$; Figure 21), and Levodopa equivalence ($r = .390, p = .036$; Figure 22). The Coefficient of variation was not significantly related any of these variables, ($p$’s > .10).

Therefore, using the Individual standard deviation measure of intraindividual variability, higher levels of disease severity (higher Hoehn and Yahr stages, longer Disease duration, and higher levels of medication - Levodopa and Levodopa equivalence) were positively associated with more intraindividual variability. This suggests that people who have had PD for a longer time, or are on more medication, likely have larger fluctuations in their dopamine levels, both within a day and perhaps across weeks or months, such that the brain is not able to adapt to consistent levels of dopamine. Therefore, tasks that rely on the striatum or regions that are innervated by the striatum, such as the motor cortex, will likely produce more variable performance and behavior due to larger fluctuations in levels of dopamine.

**Study 2 Conclusions**

In this study, we found that motor deficits in people with PD (as assessed by Hoehn and Yahr stage and UPDRS scores) were not related to learning in the Triplets Learning Task. We found that other disease-related characteristics which are more indicative of disease severity, including Disease duration and Levodopa, were related to TLT learning, significantly and marginally, respectively. It is particularly important that these correlations were found only with the Triplet type effect in the 2nd Halves of Blocks and not the 1st Halves. That is, Levodopa levels were positively related to how much learning was seen in a portion of learning that is thought to rely on the striatum (Nemeth et al., 2013). However, because both measures of medication, Disease duration and Age are all positively related to one another, it is difficult to make hypotheses about how any of them individually affected learning.
Summary

This research examined implicit sequence learning in a group of medicated people with mild to moderate idiopathic Parkinson’s disease and a group of age- and education-matched Controls. This work contributes four novel findings to the literature on sequence learning in people with Parkinson’s disease. First, we found that people with PD were able to implicitly learn a probabilistic sequence, but they showed less learning in the Triplets Learning Task than healthy Controls. Second, the PD group showed less learning than the Controls in the 2nd Half of Blocks, but not in the 1st Half. Third, the 2nd, but not the 1st Half Block Triplet type effect correlated with measures of disease severity in Parkinson’s disease. Fourth, the PD group had more intraindividual variability than Controls, and this difference increased with training.

In the following, I first consider some of the aspects of the study design that are relevant to all of our major findings, and why we chose this design. Then, I discuss our four main findings listed above, and their implications. I then consider some of the limitations of these studies and how these limitations could be overcome in future studies. I end by discussing some of the main implications and what we have learned from our findings.

Study design

**Triplets Learning Task.** Sequence learning has been examined in people with PD in a number of studies, with many of them using the SRT to examine this type of learning, but we think that the TLT overcame many of the limitations that are a result of studying learning in PD in the SRT. First, the SRT has a large motor sequencing component, with participants responding to the location of all of the stimuli, which may make it a more difficult task for people with PD than for healthy older adults. Responding to the location of each dot also creates the potential for
people to learn a motor sequence in addition to the perceptual sequence. Compared to studies using the SRT, there was no potential for motor sequence learning in the TLT and a reduced motor component, as participants only responded to the location of the target and not to all stimuli, giving us a measure of more perceptual sequence learning. Second, because of the repeating nature of the pattern in the SRT, people often become explicitly aware, and explicit awareness has been shown to hurt learning in healthy older adults (Howard & Howard Jr., 2001). We can be sure that learning in the TLT is implicit due to the probabilistic nature of the pattern; this task has a second-order regularity, such that predictive events are one event removed from the subsequent target. Sensitive post-training measures also probe awareness to ensure that there is no explicit knowledge of the learned sequence. Third, compared to the SRT, which typically only measures learning at one point in time, the TLT allows us to measure learning continuously throughout training. This enables us to not only examine how learning changes over time, but how different neural deficits (e.g., dopamine declines in PD) affect learning at different points in training, such as early versus late learning, or learning in the 1\textsuperscript{st} versus 2\textsuperscript{nd} Halves of Blocks.

**Medication status.** All of the PD participants in our study were receiving anti-Parkinsonian medication. We chose to examine learning in medicated participants for three reasons. First, this was the first time that learning was examined in the TLT in people with Parkinson’s disease. Thus, we first wanted to determine if people were able to show any learning in the task. Because people with PD have dopaminergic deficits in the striatum (Kish et al., 1988), we reasoned that the disease would affect learning in the TLT, as learning is related to dopamine (Simon, Stollstorff et al., 2011) and relies on the caudate (Simon et al., 2012), so that dopamine-replacement medication might help to moderate dopamine deficits, and thus improve their ability to show learning in the Triplets Learning Task.
Second, we recruited participants who were patients at MedStar Georgetown University Hospital, the leading location for treatment of Parkinson’s disease locally, which thus attracts patients from all surrounding areas. Because we hoped to recruit a relatively large sample of participants, many of whom would have to travel from outside of Washington, DC, we wanted participation to be as little of a burden as possible. Testing PD participants OFF of medication requires at least a 12 hour withdrawal, so as a first study with this group of participants, we wanted to make participation easy by not asking people to change their normal medication routine.

Finally, assuming high adherence to their prescribed medication, people with PD are constantly medicated, so therefore, we wanted to examine how learning of regularities occurs in people with PD in the state in which they live on a daily basis. Though people with Parkinson’s disease often report low adherence to their medication, better adherence is usually seen in people with higher education and more knowledge about the disease (Daley, Myint, Gray, & O’Leary Deane, 2012), characteristics seen in our PD participants. While having all of our participants tested in a medicated state left us to speculate about the implications of some of our findings, such as effects of the disease versus effects of medication (this will be examined in detail, later), in our first study examining implicit sequence learning in PD, we were more interested in the basic questions of whether or not learning could occur in people with PD as they live, and if so, how this learning compared to healthy matched controls and related to disease-related characteristics.

**Findings and Implications**

**Group differences in sequence learning.** People with PD were able to learn in the TLT, but compared to Controls, they showed significantly less learning of Triplet type probabilities, as
seen through both mean of median reaction time Triplet type effects (significant Group x Triplet type interaction, \( p = .037 \)) and Associative Learning scores (main effect of Group, \( p = .010 \)).

These group differences were seen in reaction time measures even though accuracy was the same for the two groups, eliminating the possibility of a speed-accuracy tradeoff that can often be a problem in comparing groups that differ in overall response time.

Additionally, the main effect of Group in the AL measure was carried by Epoch 6 at the end of training; Epoch 6 was the only individual epoch in which the group difference in sequence learning approached significance (\( p = .066 \)). Although, the predicted Group x Epoch interaction was not significant, this pattern of results provides some support for the prediction that we would see greater group differences toward the end of training due to the reliance on the caudate for learning (Bennett et al., 2011; Simon et al., 2012). We know of only one study that examined learning in the SRT at various points in time. People with PD were tested ON and OFF medication in an explicit version of the SRT, and results showed impaired learning in early training in PD ON, suggesting a selective impairment of early learning as a result of medication (Kwak, Muller, Bohnen, Dayalu, & Seidler, 2010). This study, however, was based on findings that different regions of the striatum are involved in different learning phases during an explicit sequence learning task, as opposed to the hippocampus and caudate being involved in different learning phases in the TLT, so this was a very different task compared to the implicit TLT used in the present studies.

We can be sure that these group differences were not due to possible explicit learning occurring in one group and not the other, as our post-training recognition measures, both computer-based and verbal, showed no awareness of the learned sequence in any of our participants. In probabilistic classification tasks, which involve both the hippocampus and the
striatum for learning (Poldrack & Rodriguez, 2004), and have been widely studied in people with Parkinson’s disease, explicit instructions are given to direct learning, and trial-by-trial feedback is usually explicit (de Wit et al., 2010; Foerde et al., 2013; Foerde & Shohamy, 2011). Thus, dopamine is driving learning in a way that is well understood, with phasic bursts in response to positive feedback, and dips in response to negative feedback (Frank, 2005). While we know that dopamine is involved in learning in the TLT (Simon, Stollstorff et al., 2011), this learning is implicit and does not involve explicit feedback (e.g., correct or incorrect following each response) or instructions to direct responding, making our investigation very different from those in probabilistic classification tasks.

We do not think that the learning differences we saw between our two groups were due to slower reaction time in our PD group, as an ANCOVA covarying overall RT still yielded significant group differences in learning. Therefore, the group differences likely reflected a sequence learning deficit in people with PD, and were not merely an artifact of a general deficit in response time.

While accuracy and reaction time have been examined in other sequence learning tasks, this was the first study to examine Associative Learning scores in a group of people with Parkinson’s disease. To our knowledge, this was also the first study to find impaired implicit sequence learning in a sequence learning task with a reduced motor component in a group of medicated Parkinson’s disease participants. Our results showed that even when people with PD were receiving dopaminergic medication to help moderate dopamine denervation in the striatum, they were still less sensitive to regularities in the environment than healthy Controls. However, because PD participants were all on anti-Parkinsonian medication, we cannot know for sure whether learning impairments occurred as a result of the disease, as a result of medication, or if
the medication helped to reduce negative effects of the disease. Additionally, because Age, Disease duration and Levodopa were all correlated with one another, it was difficult for us to disentangle the effects of each of these factors and examine their individual effects on learning. Based on the findings of other studies that have examined sequence learning in people with PD, however, we think that the disease was the cause of sequence learning deficits, and that medication improved learning. The effects of different levels of medication on TLT learning in the PD group as well as studies that offer further support for this hypothesis will be discussed, below.

**Group differences in the Half Block comparison.** As predicted, there were significant Group differences in the Triplet type effect in the 2nd Half Block (significant Group x Block Half interaction, $p = .006$), but not in the 1st Half Block, where learning in the 2nd Half has been suggested to rely on the striatum (Nemeth et al., 2013). The PD group revealed significantly less learning in the 2nd Half Block than in the 1st Half Block, and showed less learning than Controls in the 2nd Halves of Blocks, but equal amounts in the 1st Halves. Nemeth et al. hypothesized that the 1st Halves of Blocks involve recall and reactivation of previously learned sequences, tasks that rely on the hippocampus, while the 2nd Halves of Blocks involve automation and proceduralization of the sequence, which relies in the striatum. They showed impaired learning in the 1st Half Block in people with MCI, supporting their hypothesis that learning in the 1st Halves of Blocks relied on the hippocampus, and further suggested that learning in the 2nd Halves of Blocks relied on the striatum. Our data converged with theirs to show a double dissociation supporting this hypothesis, such that learning was impaired in the 2nd Halves of Blocks in people with PD who have striatal deficits.
Examining how RT changed from the 1\textsuperscript{st} to the 2\textsuperscript{nd} Half Block in each group helps to clarify the nature of the Group difference in Triplet type effect. In the PD group, reaction time to High Probability triplets increased more from the 1\textsuperscript{st} to the 2\textsuperscript{nd} Half Blocks than did RT to Low Probability triplets, a difference not seen in Controls. That is, reaction time seemed to become less sensitive to High Probability triplets in the 2\textsuperscript{nd} Halves of Blocks in people with Parkinson’s disease. It is possible that people with PD may never have relied on the striatum, or at least not fully, for learning in the Triplets Learning Task.

The hippocampus has been shown to compensate for striatal declines in PD (Dagher et al., 2001; Moody et al., 2004), so it is possible that the hippocampus was additionally recruited for learning in the TLT. Despite hippocampal deficits that have been shown in PD (Bouchard et al., 2008; Camicioli et al., 2003), the hippocampus may still be helpful to task performance in medicated PD participants. That is, dopaminergic medication operates in a tonic, rather than a phasic manner (Mercuri & Bernardi, 2005), and it has been suggested that the hippocampus responds better to tonic levels of dopamine than to phasic bursts of dopamine (which occur in striatal-based learning; Foerde & Shohamy, 2011). Thus, dopaminergic medication may offer a boost to hippocampal-based learning to a greater degree than striatal-based learning.

We do not think that fatigue in the PD group was responsible for the learning difference in the 2\textsuperscript{nd} Halves of Blocks, because the increase in RT from the 1\textsuperscript{st} to the 2\textsuperscript{nd} Halves of Blocks was only greater in PD compared to Controls for High Probability triplets, and not Low. If this learning difference were due to fatigue, we would expect RT to increase for both Triplet types from the 1\textsuperscript{st} to the 2\textsuperscript{nd} Half Blocks. Additionally, when we ran ANCOVAs covarying out overall RT and RT in the 2\textsuperscript{nd} Half Blocks, we still saw group differences in the 2\textsuperscript{nd} Half Block Triplet type effect, suggesting that this difference was due to learning and not fatigue or RT differences.
Correlations with learning in the PD group. Triplet type learning in the 2\textsuperscript{nd} Half Block, but not the 1\textsuperscript{st} Half Block, was positively related to Age ($r = .427$, $p = .020$), Disease duration ($r = .372$, $p = .046$) and Levodopa ($r = .343$, $p = .068$). This finding with Levodopa supported our hypothesis that people with PD who were on higher levels of Levodopa would show more learning in the TLT, though because this finding was only marginal, we cannot draw a strong conclusion. This relationship only occurred in the 2\textsuperscript{nd} Halves of Blocks, however, which suggests that this effect was specific to a portion of training that has been hypothesized to involve the striatum (Nemeth et al., 2013). However, because Age, Disease duration and Levodopa, as well as Levodopa Equivalence (though it is not related to the 2\textsuperscript{nd} Half Block Triplet type effect) are all correlated, it is difficult to disentangle their effects, and it is likely that they all had some effect on learning. This is especially true because Age is one of the primary risk factors of PD (Collier, Kanaan, & Kordower, 2011), and because the amount of Levodopa prescribed often increases with Disease duration (Mercuri & Bernardi, 2005).

There were no significant correlations of disease-related characteristics with AL scores or Triplet type effects, either Overall or in the 1\textsuperscript{st} Half of Blocks. Therefore, there seems to be something very specific about the neural underpinnings of the 2\textsuperscript{nd} Half Blocks that is particularly sensitive to the effects of PD and/or PD medication. There is no reason to expect that Age and Disease duration should correlate positively with learning without the influence of Levodopa. Age was not correlated with learning in Controls ($r = .082$, $p = .668$), and because dopamine levels should decrease with increased Age and Disease duration in PD, it seems more intuitive that these two variables should be negatively correlated with learning. We think that Disease duration is the best explanation for the positive relationships seen between the 2\textsuperscript{nd} Half Block Triplet type effect and Age and Levodopa. People who were living with PD for longer were also
older (Age x Disease duration: $r = .320, p = .091$), on more Levodopa (Levodopa x Disease duration: $r = .676, p < .001$), and on higher levels of daily anti-Parkinsonian medication (Levodopa Equivalence x Disease duration: $r = .667, p < .001$), so they were, in general, experiencing a different internal environment than their younger and less affected counterparts.

Because we know that the brain maintains plasticity and is able to adapt in PD (Sehm et al., 2014), it is likely that as disease duration increases, the brain adapts to its own declines, forming new connections and recruiting different regions for activation, similar to what is seen in healthy aging as different brain regions decline in a healthy age-related pattern (Cabeza, 2002; Park & Reuter-Lorenz, 2009). People who have lived with PD longer may have also adapted their own external compensatory mechanisms. For example, if they are experiencing difficulties with language that they had previously not experienced, such as problems with complex sentence comprehension (Grossman et al., 2001), they may read more often or more challenging material to better exercise their exposure to and abilities to understand complex sentences. By engaging in this sort of scaffolding behavior that may specifically focus on their deficits, they may improve, or at least slow, some declines.

One study examined SRT learning in people with PD who used their most affected hand to perform the task, and found that Levodopa Equivalence was negatively related to learning (Stephan et al., 2011). A closer examination of their data show that two participants have a Levodopa Equivalence level that is more than two times higher than that of the other participants, suggesting that those two individuals may have been driving this correlation. Additionally, Hoehn and Yahr stage and UPDRS scores in their participants were higher than the participants in our studies (Hoehn and Yahr: Stephan $M = 2.2$, our $M = 1.5$; UPDRS: Stephan $M = 24.0$, our $M = 8.6$), suggesting that their participants had greater disease severity than our
participants. Therefore, while our results are in contrast to those of Stephan et al., our participants differed enough from theirs that we do not think the studies are comparable.

**Group differences in Intraindividual variability.** People with PD had more intraindividual variability than the Control group (main effect of Group; Individual standard deviation: $p = .054$, Coefficient of variation: $p = .064$), and this variability increased with training (Group x Epoch interaction; Individual standard deviation: $p < .001$, Coefficient of variation: $p = .016$). Additionally, when we covaried out overall mean of median reaction time, we still found that intraindividual variability was greater in the PD than the Control group, suggesting that increased variability was not simply a factor of higher response time in the PD group. To our knowledge, this was the first time that intraindividual variability has been examined in an implicit sequence learning task in people with PD, and therefore the first to show that variability increased as training and learning continued.

Increased intraindividual variability is thought to be an indicator of increased neural noise or dysfunction (de Frias et al., 2007), so it is possible that the increase we saw in variability in our PD group was due to increased variability in their dopamine levels due to their dopaminergic medication (Levodopa or Levodopa Equivalence), which may have fluctuated over the testing session. That is, Levodopa has a half-life of 60-90 minutes (Olanow et al., 2011), so dopamine levels in our PD participants very likely changed over the course of their training, which would have introduced additional neural noise, and may have manifest as increased intraindividual variability. This hypothesis is supported by the positive correlations between variability and Levodopa (Individual standard deviation: $r = .509$, Coefficient of variance: $r = .335$) and Levodopa Equivalence (Individual standard deviation: $r = .390$, Coefficient of variance: $r = .180$, n.s.).
Limitations and Future directions

Medication vs. disease effects. In the present studies, all of the Parkinson’s disease participants were medicated, so it is difficult to disentangle whether learning differences in the 2nd Half Block Triplet type effect were due to effects of the disease or medication. Therefore, we can only speculate on how Levodopa affected learning, particularly due to the fact that the relationship was marginal, and that we did not see correlations with Levodopa Equivalence and learning in the 2nd Half Blocks. The best way to examine individual effects of Levodopa on learning would be to measure the extent of dopamine denervation in each person using positron emission tomography. Because this is a difficult and expensive process, a more reasonable future direction would be to examine learning under different conditions of dopaminergic medication. Many studies test participants in a medication OFF state in order to examine how the disease affects learning compared to how medication affects learning, but as previously mentioned, testing participants during a withdrawal from medication can present complications and would likely limit participation in a study. Therefore, the best next step would be to administer a fixed level of Levodopa to all participants, and to then examine how this medication affects learning in participants while taking into account their normal levels of medication. Again, this assumes medication levels as a proxy for dopamine denervation, but would allow the investigation of how a fixed level of dopaminergic medication differentially affects participants who are prescribed varying levels of medication. This methodology of giving fixed levels of Levodopa is used by Kwak and colleagues (Kwak et al., 2013; Kwak et al., 2010, 2012).

Limited range of disease severity. Because this was the first study to examine learning in the TLT in a group of people with PD, we recruited participants who were less affected by the disease, and thus had a smaller range of disease-related characteristics, leaving less within-group
variability to examine differences. While we chose to only test participants in Hoehn and Yahr stages 3 and below, our sample ultimately had only three participants in Stage 2.5, with the remaining participants in Stages 1 - 2. As motor impairment becomes more severe in higher disease stages, it becomes more difficult for people with PD to participate in a task that requires relatively fine motor movements, but future testing should attempt to test more people in Stage 2.5, or people in Stage 3 whose motor symptoms are well enough controlled that they are able to participate. This increased individual variability among participants would provide additional variability to determine if, and if so how, greater disease severity affects learning in the TLT.

**Effects of medication fluctuations.** We do not know where participants were in their medication cycles when they were tested. People with PD are generally able to recognize when they are at peak or waning stages of their medication, and often begin to exhibit more motor symptoms when they are close to the time of a next medication dose. By not knowing how long before testing participants took their medication, or how “on” or “off” they were feeling, we do not know whether our participants were tested at more or less optimal doses of medication, or if their medicated state changed throughout training. Therefore, future testing should attempt to test participants at similar times in their medication cycles, as well as ask participants throughout their testing session how “on” they feel in terms of the efficacy of their medication.

**Grooved Pegboard effects.** We predicted that the Grooved Pegboard would provide us with a proxy for dopamine denervation in the striatum, as has previously been shown (Bohnen et al., 2007). In contrast to our predictions, we did not find correlations in our PD group with Pegboard reaction time to any of our learning measures, and only found marginal correlations with Levodopa and Age. In Controls, however, Pegboard RT was significant correlated with Age and Overall Associative Learning score, such that people who had faster RTs (should correspond to
less dopamine denervation) were younger, and had more learning as seen through the AL measure. Parkinson’s disease participants who were tested by Bohnen and colleagues were either drug naïve or had withheld their medication before test when they showed a correlation between Pegboard RT and dopamine denervation, so it is possible that dopaminergic medication in our PD participants masked a relationship between dopamine denervation and the fine motor skills that are measured by the Grooved Pegboard.

Implications

These studies offered an opportunity to further understand the neural bases of implicit sequence learning and how these differ by analyses. By examining learning in people with Parkinson’s disease, a group with known dopamine declines and striatal deficits, in analyses where people with PD showed worse learning than healthy controls, we hypothesize that this is indicative of those points in training relying on the striatum. Specifically, finding that people with PD showed less learning in the 2nd Half of Blocks, which converged with the findings of impaired learning in the 1st Half of Blocks in a group with MTL deficits (Nemeth et al., 2013), we can make a stronger hypotheses about the two different brain regions that underlie these points in learning. That is, we can see the different effects of these two neural systems within a training block in a sequence learning task. Thus, this analysis can be used in sequence learning tasks to examine clinical populations who have neural insults, such that deficits in performance in the 1st vs. 2nd Half Blocks may help to elucidate hippocampal from striatal deficits.

The present studies found that people with PD have implicit sequence learning deficits which cannot be attributed to group differences in explicit learning or deficits in motor sequencing. However, correlations with learning and medication suggest that Levodopa may moderate this deficit and improve the ability to learn regularities. Implicit learning also seems to
only be impaired in PD participants during training phases that rely on the striatum, such that the hippocampus is involved in intact learning (1st Half Blocks and early training), and may even help with learning throughout, compensating for striatal deficits. Although the present studies focused on implicit learning, this compensation hypothesis suggests that explicit strategies in some forms of rehabilitation may help improve performance in tasks that rely on the striatum. In that light, people can use top-down scaffolding to improve performance, but there also may be scaffolding that occurs in the brain of people with PD as the disease progresses and compensation becomes a standard process. That is, as the disease progresses, due to maintained plasticity, the brain of a person with PD may adapt to its own changes, so that as the efficacy of the striatum declines, other brain regions may play a larger role in successful performance of tasks that primarily rely upon the striatum in healthy adults.

As alluded to above, measuring levels of dopamine denervation is expensive, which can only be done through positron emission tomography or single-photon emission computed tomography (SPECT) imaging. Therefore, for the most part, doctors prescribe dopaminergic medication levels to people with PD based solely on their observable symptoms, and as treatment progresses, to an extent on trial-and-error. Because we know that learning in the TLT relates to dopamine availability (Simon, Stollstorff et al., 2011), and know which brain regions underlie learning in particular points in training (Bennett et al., 2011; Nemeth et al., 2013; Simon et al., 2012), it is possible that the TLT could be used as a tool to determine levels of dopamine denervation in people with Parkinson’s disease. That is, we know that denervation moves from the putamen to the caudate (Kish et al., 1988), so it follows that more denervation would generally suggest that dopamine has begun to decline in the caudate, as well. With a larger sample, the relationship between TLT learning and disease severity could be better understood,
such that it could eventually be determined if learning differences in some parts of the TLT may be a diagnostic tool for dopamine denervation moving from the putamen to the caudate.

This set of studies helped to both better understand how implicit sequence learning occurs in a group of medicated Parkinson’s disease participants, and using their known striatal deficits, gain a better understanding of the neural bases that underlie certain portions of training in a sequence learning task. An important conclusion is that, despite extensive dopamine declines in people with PD, they are able to compensate for these deficits, through either medication or perhaps through the recruitment of other brain regions, in order to still show successful performance and learning in a task that largely relies on regions where they have extensive deficits.


Owen, A. M., James, M., Leigh, P. N., Summers, B. A., Marsden, C. D., Quinn, N. P., et al.  


**Table 1.** Individual PD participant characteristics

<table>
<thead>
<tr>
<th>Participant</th>
<th>Age</th>
<th>Gender</th>
<th>Years of Ed</th>
<th>Hoehn &amp; Yahr</th>
<th>UPDRS Motor section</th>
<th>SCOPA-COG (Learn &amp; Mem)</th>
<th>Disease Duration</th>
<th>Levodopa</th>
<th>Levodopa equivalence</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>75</td>
<td>female</td>
<td>16</td>
<td>2.5</td>
<td>9</td>
<td>35 (17)</td>
<td>14</td>
<td>300</td>
<td>1664</td>
</tr>
<tr>
<td>2</td>
<td>62</td>
<td>male</td>
<td>18</td>
<td>2</td>
<td>5</td>
<td>28 (10)</td>
<td>17</td>
<td>250</td>
<td>1464</td>
</tr>
<tr>
<td>3</td>
<td>52</td>
<td>male</td>
<td>16</td>
<td>1</td>
<td>7</td>
<td>37 (17)</td>
<td>2</td>
<td>0</td>
<td>250</td>
</tr>
<tr>
<td>4</td>
<td>68</td>
<td>male</td>
<td>19</td>
<td>1</td>
<td>8</td>
<td>30 (11)</td>
<td>10</td>
<td>100</td>
<td>400</td>
</tr>
<tr>
<td>5</td>
<td>65</td>
<td>male</td>
<td>22</td>
<td>1</td>
<td>0</td>
<td>31 (10)</td>
<td>1</td>
<td>0</td>
<td>100</td>
</tr>
<tr>
<td>6</td>
<td>70</td>
<td>female</td>
<td>14</td>
<td>1</td>
<td>3</td>
<td>28 (9)</td>
<td>12</td>
<td>150</td>
<td>600</td>
</tr>
<tr>
<td>7</td>
<td>61</td>
<td>female</td>
<td>18</td>
<td>1</td>
<td>11.5</td>
<td>31 (13)</td>
<td>3</td>
<td>0</td>
<td>550</td>
</tr>
<tr>
<td>8</td>
<td>69</td>
<td>female</td>
<td>21</td>
<td>2</td>
<td>1.5</td>
<td>31 (13)</td>
<td>6</td>
<td>200</td>
<td>1060</td>
</tr>
<tr>
<td>9</td>
<td>66</td>
<td>female</td>
<td>21</td>
<td>1</td>
<td>12</td>
<td>35 (14)</td>
<td>3</td>
<td>0</td>
<td>120</td>
</tr>
<tr>
<td>10</td>
<td>74</td>
<td>male</td>
<td>19</td>
<td>2</td>
<td>9.5</td>
<td>31 (12)</td>
<td>3</td>
<td>100</td>
<td>400</td>
</tr>
<tr>
<td>11</td>
<td>63</td>
<td>female</td>
<td>16</td>
<td>2</td>
<td>2</td>
<td>34 (14)</td>
<td>7</td>
<td>200</td>
<td>850</td>
</tr>
<tr>
<td>12</td>
<td>65</td>
<td>female</td>
<td>23</td>
<td>2.5</td>
<td>29</td>
<td>35 (14)</td>
<td>11</td>
<td>150</td>
<td>400</td>
</tr>
<tr>
<td>13</td>
<td>72</td>
<td>female</td>
<td>16</td>
<td>2.5</td>
<td>18</td>
<td>31 (12)</td>
<td>12</td>
<td>200</td>
<td>1680</td>
</tr>
<tr>
<td>14</td>
<td>62</td>
<td>male</td>
<td>16</td>
<td>1</td>
<td>5</td>
<td>30 (10)</td>
<td>2</td>
<td>0</td>
<td>300</td>
</tr>
<tr>
<td>15</td>
<td>66</td>
<td>male</td>
<td>21</td>
<td>1</td>
<td>8</td>
<td>29 (12)</td>
<td>7</td>
<td>100</td>
<td>400</td>
</tr>
<tr>
<td>16</td>
<td>66</td>
<td>male</td>
<td>21</td>
<td>2</td>
<td>10.5</td>
<td>30 (11)</td>
<td>7</td>
<td>100</td>
<td>775</td>
</tr>
<tr>
<td><strong>17</strong></td>
<td><strong>58</strong></td>
<td>female</td>
<td><strong>16</strong></td>
<td><strong>2</strong></td>
<td><strong>3</strong></td>
<td><strong>24 (9)</strong></td>
<td><strong>5</strong></td>
<td><strong>100</strong></td>
<td><strong>650</strong></td>
</tr>
<tr>
<td>18</td>
<td>58</td>
<td>male</td>
<td>21</td>
<td>2</td>
<td>15.5</td>
<td>29 (9)</td>
<td>2</td>
<td>0</td>
<td>400</td>
</tr>
<tr>
<td>19</td>
<td>57</td>
<td>male</td>
<td>18</td>
<td>1</td>
<td>14</td>
<td>34 (14)</td>
<td>6</td>
<td>0</td>
<td>180</td>
</tr>
<tr>
<td>20</td>
<td>52</td>
<td>male</td>
<td>16</td>
<td>1</td>
<td>12</td>
<td>32 (13)</td>
<td>5</td>
<td>0</td>
<td>400</td>
</tr>
<tr>
<td>21</td>
<td>69</td>
<td>male</td>
<td>12</td>
<td>2</td>
<td>17</td>
<td>31 (11)</td>
<td>4</td>
<td>100</td>
<td>300</td>
</tr>
<tr>
<td>22</td>
<td>62</td>
<td>female</td>
<td>12</td>
<td>1</td>
<td>3</td>
<td>27 (10)</td>
<td>6</td>
<td>100</td>
<td>700</td>
</tr>
<tr>
<td>23</td>
<td>69</td>
<td>female</td>
<td>18</td>
<td>1</td>
<td>4</td>
<td>34 (13)</td>
<td>5</td>
<td>100</td>
<td>700</td>
</tr>
<tr>
<td>24</td>
<td>59</td>
<td>male</td>
<td>18</td>
<td>1</td>
<td>11</td>
<td>32 (15)</td>
<td>1</td>
<td>0</td>
<td>250</td>
</tr>
<tr>
<td>25</td>
<td>64</td>
<td>male</td>
<td>21</td>
<td>1</td>
<td>2</td>
<td>31 (11)</td>
<td>8</td>
<td>100</td>
<td>875</td>
</tr>
<tr>
<td>26</td>
<td>71</td>
<td>female</td>
<td>18</td>
<td>2</td>
<td>5.5</td>
<td>29 (10)</td>
<td>4</td>
<td>200</td>
<td>1164</td>
</tr>
<tr>
<td>27</td>
<td>66</td>
<td>male</td>
<td>16</td>
<td>1</td>
<td>0</td>
<td>36 (16)</td>
<td>4</td>
<td>100</td>
<td>700</td>
</tr>
<tr>
<td>28</td>
<td>65</td>
<td>male</td>
<td>18</td>
<td>2</td>
<td>16</td>
<td>25 (8)</td>
<td>18</td>
<td>100</td>
<td>1000</td>
</tr>
<tr>
<td>29</td>
<td>66</td>
<td>male</td>
<td>18</td>
<td>1</td>
<td>6</td>
<td>30 (10)</td>
<td>6</td>
<td>100</td>
<td>600</td>
</tr>
</tbody>
</table>

Average: 64.55  17.90  1.5  8.55  31.03 (12)  6.59  98.28  652.83

SD: 5.77  2.78  0.58  6.53  3.12 (2.43)  4.55  83.97  440.09

*Participants in **bold** are those excluded from Study 1.*
**Table 2.** Parkinson's disease and healthy control participant characteristics for Study 1. Standard deviation in parentheses.

<table>
<thead>
<tr>
<th></th>
<th>Parkinson's disease participants</th>
<th>Control Participants</th>
<th>t</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>n</td>
<td>27</td>
<td>30</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Years of Education</td>
<td>18.11 (2.75)</td>
<td>16.83 (3.52)</td>
<td>-1.51</td>
<td>0.136</td>
</tr>
<tr>
<td>Reported Overall health (out of 5)</td>
<td>3.93 (0.82)</td>
<td>4.27 (0.87)</td>
<td>1.45</td>
<td>0.153</td>
</tr>
<tr>
<td>Geriatric Depression Score</td>
<td>2.00 (2.15)</td>
<td>0.90 (1.56)</td>
<td>-2.23</td>
<td>0.03</td>
</tr>
<tr>
<td>Self-reported Stress</td>
<td>10.93 (5.67)</td>
<td>9.93 (4.76)</td>
<td>-0.71</td>
<td>0.479</td>
</tr>
<tr>
<td>Montreal Cognitive Assessment</td>
<td>27.11 (1.72)</td>
<td>27.97 (1.50)</td>
<td>2.01</td>
<td>0.05</td>
</tr>
<tr>
<td>Mini Mental State Examination</td>
<td>28.59 (0.69)</td>
<td>29.17 (0.83)</td>
<td>2.81</td>
<td>0.007</td>
</tr>
<tr>
<td>SCOPA-COG overall</td>
<td>31.41 (2.85)</td>
<td>n/a</td>
<td></td>
<td></td>
</tr>
<tr>
<td>SCOPA-COG Learning &amp; Memory</td>
<td>12.22 (2.38)</td>
<td>n/a</td>
<td></td>
<td></td>
</tr>
<tr>
<td>WAIS-III Digit Span Forward</td>
<td>10.70 (2.20)</td>
<td>10.23 (2.03)</td>
<td>-0.84</td>
<td>0.405</td>
</tr>
<tr>
<td>WAIS-III Digit Span Backward</td>
<td>7.00 (1.88)</td>
<td>8.36 (2.06)</td>
<td>2.49</td>
<td>0.016</td>
</tr>
<tr>
<td>North American Adult Reading Test-35</td>
<td>9.70 (5.50)</td>
<td>8.23 (5.71)</td>
<td>-0.99</td>
<td>0.328</td>
</tr>
</tbody>
</table>
### Table 3 (part 1). Correlations Matrix for Parkinson’s disease participants

<table>
<thead>
<tr>
<th></th>
<th>H &amp; Y stage</th>
<th>Disease duration</th>
<th>L-Dopa</th>
<th>L-Dopa EQ</th>
<th>UPDRS</th>
<th>Pegboard</th>
<th>MMRT</th>
<th>ACC tt effect</th>
<th>B Digit Span</th>
<th>F Digit Span</th>
<th>Age</th>
<th>Education</th>
<th>MoCA</th>
<th>MMSE</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>H &amp; Y stage</strong></td>
<td>1.00</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Disease duration</strong></td>
<td></td>
<td>1.00</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>L-Dopa</strong></td>
<td></td>
<td></td>
<td>.676*</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>L-Dopa EQ</strong></td>
<td></td>
<td>.846*</td>
<td></td>
<td>-0.099</td>
<td>0.447*</td>
<td>.414*</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>UPDRS</strong></td>
<td>1.00</td>
<td>-0.136</td>
<td>-.248</td>
<td></td>
<td>-.367*</td>
<td>.404*</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Pegboard</strong></td>
<td>1.00</td>
<td></td>
<td>.232</td>
<td>.009</td>
<td>.231</td>
<td>.030</td>
<td>.349†</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>MMRT</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>ACC tt effect</strong></td>
<td>1.00</td>
<td></td>
<td>.108</td>
<td>.055</td>
<td>.045</td>
<td>.071</td>
<td>.213</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>B Digit Span</strong></td>
<td>1.00</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>F Digit Span</strong></td>
<td>1.00</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Age</strong></td>
<td>1.00</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Education</strong></td>
<td>1.00</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>0.196</td>
<td>-0.136</td>
<td></td>
</tr>
<tr>
<td><strong>MoCA</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>1.00</td>
<td>0.110</td>
<td></td>
</tr>
<tr>
<td><strong>MMSE</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>1.00</td>
<td></td>
</tr>
<tr>
<td><strong>NAART</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Stress</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>SCOPA-Cog</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Scopa-Cog Learning &amp; Memory</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*Significant correlations are in **bold**.
Table 3 (part 2). Correlations Matrix for Parkinson’s disease participants

<table>
<thead>
<tr>
<th></th>
<th>NAART</th>
<th>Stress</th>
<th>SCOPA-Cog</th>
<th>SCOPA-Cog Learning &amp; Memory</th>
<th>Overall tt effect</th>
<th>Overall AL score</th>
<th>1st Half Block tt effect</th>
<th>2nd Half Block tt effect</th>
<th>Overall ISD</th>
<th>Overall CoV</th>
</tr>
</thead>
<tbody>
<tr>
<td>H &amp; Y</td>
<td>0.042</td>
<td>0.062</td>
<td>-0.067</td>
<td>0.042</td>
<td>0.022</td>
<td>-0.081</td>
<td>-0.065</td>
<td>-0.066</td>
<td>.368*</td>
<td>.231</td>
</tr>
<tr>
<td>Disease duration</td>
<td>0.163</td>
<td>-0.019</td>
<td>-0.288</td>
<td>-0.208</td>
<td>0.218</td>
<td>-0.128</td>
<td>-0.066</td>
<td>0.372*</td>
<td>.366*</td>
<td>.215</td>
</tr>
<tr>
<td>L-Dopa</td>
<td>0.156</td>
<td>0.047</td>
<td>-0.095</td>
<td>0.009</td>
<td>0.272</td>
<td>-0.023</td>
<td>-0.014</td>
<td>.343†</td>
<td>.509*</td>
<td>.335†</td>
</tr>
<tr>
<td>L-Dopa EQ</td>
<td>0.136</td>
<td>0.292</td>
<td>-0.176</td>
<td>-0.035</td>
<td>0.266</td>
<td>-0.103</td>
<td>-0.039</td>
<td>0.174</td>
<td>.390*</td>
<td>.180</td>
</tr>
<tr>
<td>UPDRS</td>
<td>-0.100</td>
<td>-0.038</td>
<td>0.149</td>
<td>0.114</td>
<td>-0.077</td>
<td>-0.024</td>
<td>0.169</td>
<td>-0.161</td>
<td>.186</td>
<td>.240</td>
</tr>
<tr>
<td>Pegboard</td>
<td>0.150</td>
<td>-0.325†</td>
<td>0.185</td>
<td>0.189</td>
<td>0.298</td>
<td>-0.032</td>
<td>0.134</td>
<td>0.248</td>
<td>.302</td>
<td>.225</td>
</tr>
<tr>
<td>MMRT</td>
<td>-0.054</td>
<td>-0.030</td>
<td>-0.313†</td>
<td>-0.309</td>
<td>0.014</td>
<td>-0.489*</td>
<td>0.268</td>
<td>0.157</td>
<td>.763*</td>
<td>.150</td>
</tr>
<tr>
<td>ACC tt effect</td>
<td>-0.056</td>
<td>-0.313†</td>
<td>0.126</td>
<td>0.075</td>
<td>.469*</td>
<td>0.326†</td>
<td>0.151</td>
<td>.373*</td>
<td>-1.52</td>
<td>-1.28</td>
</tr>
<tr>
<td>B Digit Span</td>
<td>0.258</td>
<td>-0.060</td>
<td>0.518*</td>
<td>0.489*</td>
<td>0.088</td>
<td>0.08</td>
<td>0.098</td>
<td>-0.017</td>
<td>-0.259</td>
<td>-0.182</td>
</tr>
<tr>
<td>F Digit Span</td>
<td>0.282</td>
<td>-0.079</td>
<td>0.249</td>
<td>0.132</td>
<td>-0.080</td>
<td>0.087</td>
<td>0.133</td>
<td>-0.271</td>
<td>-0.243</td>
<td>-0.023</td>
</tr>
<tr>
<td>Age</td>
<td><strong>0.446</strong></td>
<td>0.100</td>
<td>-0.025</td>
<td>-0.081</td>
<td>0.176</td>
<td>0.063</td>
<td>-0.053</td>
<td><strong>0.427</strong></td>
<td><strong>0.495</strong></td>
<td><strong>0.331†</strong></td>
</tr>
<tr>
<td>Education</td>
<td>0.141</td>
<td>-0.007</td>
<td>0.124</td>
<td>0.005</td>
<td>0.082</td>
<td>0.059</td>
<td>-0.093</td>
<td>-0.030</td>
<td>-1.42</td>
<td>.132</td>
</tr>
<tr>
<td>MoCA</td>
<td>0.195</td>
<td>0.122</td>
<td><strong>0.493</strong></td>
<td><strong>0.440</strong></td>
<td>0.205</td>
<td>0.09</td>
<td>-0.070</td>
<td>0.072</td>
<td>-0.088</td>
<td>.179</td>
</tr>
<tr>
<td>MMSE</td>
<td>0.220</td>
<td>-0.331†</td>
<td>0.191</td>
<td>0.107</td>
<td>0.062</td>
<td>-0.037</td>
<td>0.062</td>
<td>0.059</td>
<td>.013</td>
<td>-0.063</td>
</tr>
<tr>
<td>NAART</td>
<td>1.00</td>
<td>0.124</td>
<td>0.204</td>
<td>0.098</td>
<td>0.011</td>
<td>0.195</td>
<td>-0.120</td>
<td>0.271</td>
<td>-0.025</td>
<td>.041</td>
</tr>
<tr>
<td>Stress</td>
<td>1.00</td>
<td>-0.069</td>
<td>-0.147</td>
<td>0.093</td>
<td>0.154</td>
<td>0.164</td>
<td>-0.028</td>
<td>.066</td>
<td>.168</td>
<td></td>
</tr>
<tr>
<td>SCOPA-Cog</td>
<td>1.00</td>
<td><strong>0.888</strong></td>
<td>0.335†</td>
<td>0.262</td>
<td>0.258</td>
<td>0.257</td>
<td>.006</td>
<td>.282</td>
<td></td>
<td></td>
</tr>
<tr>
<td>SCOPA-Cog Learning &amp; Memory</td>
<td>1.00</td>
<td>.405*</td>
<td>0.258</td>
<td>0.162</td>
<td>0.193</td>
<td>-0.044</td>
<td>.186</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Overall tt effect</td>
<td>1.00</td>
<td><strong>.454</strong></td>
<td><strong>.546</strong></td>
<td><strong>.569</strong></td>
<td>.149</td>
<td>.165</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Overall AL score</td>
<td>1.00</td>
<td>.202</td>
<td>.382*</td>
<td>-2.66</td>
<td>.055</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1st Half Block tt effect</td>
<td>1.00</td>
<td>.164</td>
<td>.401*</td>
<td>.322†</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2nd Half Block tt effect</td>
<td>1.00</td>
<td>.267</td>
<td>.216</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Overall ISD</td>
<td>1.00</td>
<td>.747*</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Overall CoV</td>
<td>1.00</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*Significant correlations are in **bold**.
**Table 4. Correlations Matrix for Control participants**

<table>
<thead>
<tr>
<th></th>
<th>Pegboard</th>
<th>MMRT</th>
<th>ACC tt effect</th>
<th>B Digit Span</th>
<th>F Digit Span</th>
<th>Age</th>
<th>Education</th>
<th>MoCA</th>
<th>MMSE</th>
<th>NAART</th>
<th>Stress</th>
<th>Overall tt effect</th>
<th>Overall AL score</th>
<th>Overall 1st Half Block tt effect</th>
<th>Overall 2nd Half Block tt effect</th>
<th>Overall ISD</th>
<th>Overall CoV</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pegboard</td>
<td>1.00</td>
<td>.326†</td>
<td>.145</td>
<td>.070</td>
<td>.010</td>
<td>.572*</td>
<td>.040</td>
<td>-0.118</td>
<td>-0.422*</td>
<td>-0.19</td>
<td>.204</td>
<td>-0.131</td>
<td>-0.416*</td>
<td>-0.118</td>
<td>0.217</td>
<td>.877*</td>
<td>.533*</td>
</tr>
<tr>
<td>MMRT</td>
<td>1.00</td>
<td>.328†</td>
<td>.010</td>
<td>.015</td>
<td>-0.117</td>
<td>.379*</td>
<td>-0.323†</td>
<td>-0.251</td>
<td>.434*</td>
<td>-0.084</td>
<td>-0.455*</td>
<td>0.217</td>
<td>0.217</td>
<td>-0.877*</td>
<td>-0.222</td>
<td>.024</td>
<td></td>
</tr>
<tr>
<td>ACC tt effect</td>
<td></td>
<td></td>
<td>1.00</td>
<td>.259</td>
<td>-.044</td>
<td>.078</td>
<td>-0.070</td>
<td>-0.117</td>
<td>.074</td>
<td>-0.175</td>
<td>.548*</td>
<td>.615*</td>
<td>.540*</td>
<td>0.303</td>
<td>0.068</td>
<td>-0.123</td>
<td></td>
</tr>
<tr>
<td>B Digit Span</td>
<td>1.00</td>
<td>.266</td>
<td>.164</td>
<td>.290</td>
<td>.169</td>
<td>.045</td>
<td>.790*</td>
<td>.068</td>
<td>0.137</td>
<td>0.124</td>
<td>0.227</td>
<td>0.117</td>
<td>0.092</td>
<td>.488*</td>
<td>.348†</td>
<td>.348†</td>
<td></td>
</tr>
<tr>
<td>F Digit Span</td>
<td></td>
<td></td>
<td>1.00</td>
<td>-0.142</td>
<td>-0.020</td>
<td>-0.065</td>
<td>.356*</td>
<td>-0.075</td>
<td>-0.359*</td>
<td>-0.242</td>
<td>-0.289</td>
<td>-0.221</td>
<td>-0.021</td>
<td>-0.215</td>
<td>-0.257</td>
<td>.257</td>
<td></td>
</tr>
<tr>
<td>Age</td>
<td></td>
<td></td>
<td></td>
<td>1.00</td>
<td>0.091</td>
<td>-0.002</td>
<td>-0.532*</td>
<td>0.021</td>
<td>0.096</td>
<td>0.013</td>
<td>0.019</td>
<td>0.001</td>
<td>0.082</td>
<td>.488*</td>
<td>.348†</td>
<td>.348†</td>
<td></td>
</tr>
<tr>
<td>Education</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>1.00</td>
<td>.391*</td>
<td>.362*</td>
<td></td>
<td>.438*</td>
<td></td>
<td>.318†</td>
<td>-0.174</td>
<td>-0.174</td>
<td>-0.081</td>
<td>-0.275</td>
<td>-0.215</td>
<td></td>
</tr>
<tr>
<td>MoCA</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>1.00</td>
<td>.364*</td>
<td>.435*</td>
<td>.143</td>
<td>-0.082</td>
<td>0.035</td>
<td>-0.146</td>
<td>-0.204</td>
<td>-0.410*</td>
<td>-0.356†</td>
<td></td>
<td></td>
</tr>
<tr>
<td>MMSE</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NAART</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Stress</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Overall tt effect</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Overall AL score</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>1.00</td>
<td>.660*</td>
<td>.409*</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Overall 1st Half Block tt effect</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Overall 2nd Half Block tt effect</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Overall ISD</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Overall CoV</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*Significant correlations are in **bold.***
Figure 1. The Triplets Learning Task. The first triplet indicates a High Probability triplet (e.g., 3r1) and the second triplet indicates a Low Probability triplet (e.g., 3r2).
**Figure 2.** TLT Recognition task rating of the frequency of triplets’ occurrence. Bars indicate standard error.

### ANOVA for Recognition task

<table>
<thead>
<tr>
<th></th>
<th>DF</th>
<th>Sum of Squares</th>
<th>Mean Square</th>
<th>F-Value</th>
<th>P-Value</th>
<th>Lambda</th>
<th>Power</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group</td>
<td>1</td>
<td>.003</td>
<td>.003</td>
<td>.036</td>
<td>.8509</td>
<td>.036</td>
<td>.054</td>
</tr>
<tr>
<td>Subject (Group)</td>
<td>55</td>
<td>4.900</td>
<td>.089</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Triplet type</td>
<td>1</td>
<td>.012</td>
<td>.012</td>
<td>1.996</td>
<td>.1633</td>
<td>1.996</td>
<td>.269</td>
</tr>
<tr>
<td>Triplet type * Group</td>
<td>1</td>
<td>.014</td>
<td>.014</td>
<td>2.270</td>
<td>.1376</td>
<td>2.270</td>
<td>.300</td>
</tr>
<tr>
<td>Triplet type * Subject (Group)</td>
<td>55</td>
<td>.342</td>
<td>.006</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Inclusion criteria: Subjects removed for comparison from 30 PD vs. 26 Control subjects.
Figure 3. TLT accuracy. Bars indicate standard error.

Accuracy by Group ANOVA

ANOVA Table for Mean ACC w s509 fixed w Reps/Trills
Inclusion criteria: Subjects removed for comparison from 30 PD vs. 26 Control subjects.svd

<table>
<thead>
<tr>
<th></th>
<th>DF</th>
<th>Sum of Squares</th>
<th>Mean Square</th>
<th>F-Value</th>
<th>P-Value</th>
<th>Lambda</th>
<th>Power</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group</td>
<td></td>
<td>2.111E-4</td>
<td>2.111E-4</td>
<td>.029</td>
<td>.8652</td>
<td>.029</td>
<td>.053</td>
</tr>
<tr>
<td>Subject(Group)</td>
<td>53</td>
<td></td>
<td>.384</td>
<td>.007</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Triplet type</td>
<td>1</td>
<td>.001</td>
<td>.001</td>
<td>1.156</td>
<td>.2871</td>
<td>1.156</td>
<td>.174</td>
</tr>
<tr>
<td>Triplet type * Group</td>
<td>1</td>
<td>.001</td>
<td>1.590</td>
<td>.2128</td>
<td>1.590</td>
<td>.223</td>
<td></td>
</tr>
<tr>
<td>Triplet type * Subject(Group)</td>
<td>53</td>
<td>.044</td>
<td>.001</td>
<td>1.156</td>
<td>.2871</td>
<td>1.156</td>
<td>.174</td>
</tr>
<tr>
<td>Epoch</td>
<td>5</td>
<td>.004</td>
<td>.001</td>
<td>1.643</td>
<td>.1488</td>
<td>8.217</td>
<td>.561</td>
</tr>
<tr>
<td>Epoch * Group</td>
<td>265</td>
<td>.003</td>
<td>.001</td>
<td>1.253</td>
<td>.2849</td>
<td>6.263</td>
<td>.434</td>
</tr>
<tr>
<td>Epoch * Subject(Group)</td>
<td>265</td>
<td>.123</td>
<td>4.656E-4</td>
<td>.3006</td>
<td>6.094</td>
<td>.423</td>
<td></td>
</tr>
<tr>
<td>Triplet type * Epoch</td>
<td>5</td>
<td>.003</td>
<td>.001</td>
<td>1.219</td>
<td>.3006</td>
<td>6.094</td>
<td>.423</td>
</tr>
<tr>
<td>Triplet type * Epoch * Group</td>
<td>265</td>
<td>.123</td>
<td>4.656E-4</td>
<td>.3006</td>
<td>6.094</td>
<td>.423</td>
<td></td>
</tr>
<tr>
<td>Triplet type * Epoch * Subject(Group)</td>
<td>265</td>
<td>.123</td>
<td>4.656E-4</td>
<td>.3006</td>
<td>6.094</td>
<td>.423</td>
<td></td>
</tr>
</tbody>
</table>
**Figure 4.** TLT Mean of median reaction time. Bars indicate standard error.

### ANOVA Table for MMRT w.o s509 e1/e2 w Reps/Trills

Inclusion criteria: Subjects removed for comparison from 30 PD vs. 26 Control subjects.svd

<table>
<thead>
<tr>
<th>Factor</th>
<th>DF</th>
<th>Sum of Squares</th>
<th>Mean Square</th>
<th>F-Value</th>
<th>P-Value</th>
<th>Lambda</th>
<th>Power</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group</td>
<td>1</td>
<td>170286.463</td>
<td>170286.463</td>
<td>2.431</td>
<td>.1249</td>
<td>2.431</td>
<td>.318</td>
</tr>
<tr>
<td>Subject(Subject)</td>
<td>53</td>
<td>3713017.917</td>
<td>70056.942</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Triplet type</td>
<td>1</td>
<td>53162.607</td>
<td>53162.607</td>
<td>117.108</td>
<td>&lt;.0001</td>
<td>117.108</td>
<td>1.000</td>
</tr>
<tr>
<td>Triplet type * Group</td>
<td>1</td>
<td>2070.562</td>
<td>2070.562</td>
<td>4.561</td>
<td>.0373</td>
<td>4.561</td>
<td>.545</td>
</tr>
<tr>
<td>Triplet type * Subject(Subject)</td>
<td>53</td>
<td>24060.056</td>
<td>453.963</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Epoch</td>
<td>5</td>
<td>198658.058</td>
<td>39731.612</td>
<td>23.459</td>
<td>&lt;.0001</td>
<td>117.293</td>
<td>1.000</td>
</tr>
<tr>
<td>Epoch * Group</td>
<td>5</td>
<td>18111.304</td>
<td>3622.261</td>
<td>2.139</td>
<td>.0612</td>
<td>10.693</td>
<td>.699</td>
</tr>
<tr>
<td>Epoch * Subject(Subject)</td>
<td>265</td>
<td>448827.667</td>
<td>1693.689</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Triplet type * Epoch</td>
<td>5</td>
<td>787.617</td>
<td>157.523</td>
<td>.783</td>
<td>.5627</td>
<td>3.915</td>
<td>.275</td>
</tr>
<tr>
<td>Triplet type * Epoch * Group</td>
<td>5</td>
<td>137.107</td>
<td>27.421</td>
<td>.136</td>
<td>.9838</td>
<td>1.000</td>
<td>.081</td>
</tr>
</tbody>
</table>

105
Figure 5. TLT Associative Learning (AL) scores. Bars indicate standard error.

AL scores by Group ANOVA

<table>
<thead>
<tr>
<th></th>
<th>DF</th>
<th>Sum of Squares</th>
<th>Mean Square</th>
<th>F-Value</th>
<th>P-Value</th>
<th>Lambda Power</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group</td>
<td>1</td>
<td>.113</td>
<td>.113</td>
<td>7.082</td>
<td>.0103</td>
<td>.751</td>
</tr>
<tr>
<td>Subject(Subject)</td>
<td>53</td>
<td>.846</td>
<td>.016</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Epoch</td>
<td>5</td>
<td>.037</td>
<td>.007</td>
<td>.808</td>
<td>.5446</td>
<td>.283</td>
</tr>
<tr>
<td>Epoch * Group</td>
<td>5</td>
<td>.027</td>
<td>.005</td>
<td>.591</td>
<td>.7067</td>
<td>.212</td>
</tr>
<tr>
<td>Epoch * Subject(Subject)</td>
<td>265</td>
<td>2.406</td>
<td>.099</td>
<td>4.042</td>
<td>.283</td>
<td></td>
</tr>
</tbody>
</table>
Figure 6. Half Block MMRT across epochs. Control group on the left, PD group on the right. Bars indicate standard error.

<table>
<thead>
<tr>
<th>Source of Variance</th>
<th>DF</th>
<th>Sum of Squares</th>
<th>Mean Square</th>
<th>F-Value</th>
<th>P-Value</th>
<th>Lambda</th>
<th>Power</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group</td>
<td>1</td>
<td>335916.296</td>
<td>335916.296</td>
<td>2.366</td>
<td>.1298</td>
<td>2.366</td>
<td>.310</td>
</tr>
<tr>
<td>Subject (Group)</td>
<td>54</td>
<td>7665940.265</td>
<td>141961.857</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Triplet type</td>
<td>1</td>
<td>165160.566</td>
<td>165160.566</td>
<td>173.567</td>
<td>&lt;.0001</td>
<td>173.567</td>
<td>1.000</td>
</tr>
<tr>
<td>Triplet type * Group</td>
<td>1</td>
<td>4007.448</td>
<td>4007.448</td>
<td>4.211</td>
<td>.0450</td>
<td>4.211</td>
<td>.510</td>
</tr>
<tr>
<td>Triplet type * Subject (Group)</td>
<td>54</td>
<td>51384.716</td>
<td>951.569</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Block half</td>
<td>1</td>
<td>70958.918</td>
<td>70958.918</td>
<td>53.935</td>
<td>&lt;.0001</td>
<td>53.935</td>
<td>1.000</td>
</tr>
<tr>
<td>Block half * Group</td>
<td>1</td>
<td>10841.307</td>
<td>10841.307</td>
<td>8.240</td>
<td>.0058</td>
<td>8.240</td>
<td>.818</td>
</tr>
<tr>
<td>Block half * Subject (Group)</td>
<td>54</td>
<td>71044.678</td>
<td>1315.642</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Epoch</td>
<td>5</td>
<td>585159.521</td>
<td>117031.904</td>
<td>19.994</td>
<td>&lt;.0001</td>
<td>99.969</td>
<td>1.000</td>
</tr>
<tr>
<td>Epoch * Group</td>
<td>5</td>
<td>16103.510</td>
<td>3220.702</td>
<td>.550</td>
<td>.7381</td>
<td>.749</td>
<td>.130</td>
</tr>
<tr>
<td>Epoch * Subject (Group)</td>
<td>270</td>
<td>1580419.774</td>
<td>5853.407</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Triplet type * Block half</td>
<td>1</td>
<td>352.616</td>
<td>352.616</td>
<td>.749</td>
<td>.3905</td>
<td>.749</td>
<td>.130</td>
</tr>
<tr>
<td>Triplet type * Block half * Group</td>
<td>1</td>
<td>3799.664</td>
<td>3799.664</td>
<td>8.075</td>
<td>.0063</td>
<td>8.075</td>
<td>.810</td>
</tr>
<tr>
<td>Triplet type * Block half * Subject (Group)</td>
<td>54</td>
<td>25408.031</td>
<td>470.519</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Triplet type * Epoch</td>
<td>5</td>
<td>2108.139</td>
<td>421.628</td>
<td>.837</td>
<td>.5245</td>
<td>4.184</td>
<td>.293</td>
</tr>
<tr>
<td>Triplet type * Epoch * Group</td>
<td>5</td>
<td>2079.358</td>
<td>415.872</td>
<td>.825</td>
<td>.5325</td>
<td>4.127</td>
<td>.289</td>
</tr>
<tr>
<td>Triplet type * Epoch * Subject (Group)</td>
<td>270</td>
<td>136045.991</td>
<td>503.874</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Block half * Epoch</td>
<td>5</td>
<td>31540.126</td>
<td>6308.025</td>
<td>8.217</td>
<td>&lt;.0001</td>
<td>41.086</td>
<td>1.000</td>
</tr>
<tr>
<td>Block half * Epoch * Group</td>
<td>5</td>
<td>1282.296</td>
<td>256.459</td>
<td>.334</td>
<td>.8921</td>
<td>1.670</td>
<td>.34</td>
</tr>
<tr>
<td>Block half * Epoch * Subject (Group)</td>
<td>270</td>
<td>207266.164</td>
<td>767.652</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Triplet type * Block half * Epoch</td>
<td>5</td>
<td>1416.704</td>
<td>283.341</td>
<td>.540</td>
<td>.7461</td>
<td>2.699</td>
<td>.196</td>
</tr>
<tr>
<td>Triplet type * Block half * Epoch * Group</td>
<td>5</td>
<td>1464.958</td>
<td>292.992</td>
<td>.558</td>
<td>.7320</td>
<td>2.791</td>
<td>.201</td>
</tr>
<tr>
<td>Triplet type * Block half * Epoch * Subject (Group)</td>
<td>270</td>
<td>141730.450</td>
<td>524.928</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Figure 7. Half Block Triplet type effect (RT to Low Probability triplets – RT to High Probability triplets) across epochs. Bars indicate standard error.

Figure 8. Overall Half Block Triplet type effect (RT to Low Probability triplets – RT to High Probability triplets) by Group. Bars indicate standard error.
**Figure 9.** Individual standard deviation. Bars indicate standard error.

### Individual standard deviation by Group and Epoch ANOVA

<table>
<thead>
<tr>
<th></th>
<th>DF</th>
<th>Sum of Squares</th>
<th>Mean Square</th>
<th>F-Value</th>
<th>P-Value</th>
<th>Lambda</th>
<th>Power</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group</td>
<td>1</td>
<td>21225.345</td>
<td>21225.345</td>
<td>4.131</td>
<td>.0470</td>
<td>4.131</td>
<td>.502</td>
</tr>
<tr>
<td>Subject(Group)</td>
<td>54</td>
<td>277447.329</td>
<td>5137.914</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Epoch</td>
<td>5</td>
<td>15074.682</td>
<td>3014.936</td>
<td>14.135</td>
<td>&lt;.0001</td>
<td>70.675</td>
<td>1.000</td>
</tr>
<tr>
<td>Epoch * Group</td>
<td>5</td>
<td>2566.250</td>
<td>513.250</td>
<td>2.406</td>
<td>.0371</td>
<td>12.031</td>
<td>.761</td>
</tr>
<tr>
<td>Epoch * Subject(Group)</td>
<td>270</td>
<td>57589.826</td>
<td>213.296</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Figure 10. Coefficient of variation. Bars indicate standard error.

Coefficient of variation by Group and Epoch ANOVA

<table>
<thead>
<tr>
<th></th>
<th>DF</th>
<th>Sum of Squares</th>
<th>Mean Square</th>
<th>F-Value</th>
<th>P-Value</th>
<th>Lambda</th>
<th>Power</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group</td>
<td>1</td>
<td>.028</td>
<td>.028</td>
<td>4.022</td>
<td>.0499</td>
<td>4.022</td>
<td>.491</td>
</tr>
<tr>
<td>Subject(Group)</td>
<td>54</td>
<td>.381</td>
<td>.007</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Epoch</td>
<td>5</td>
<td>.010</td>
<td>.002</td>
<td>4.626</td>
<td>.0005</td>
<td>23.130</td>
<td>.980</td>
</tr>
<tr>
<td>Epoch * Group</td>
<td>5</td>
<td>.006</td>
<td>.001</td>
<td>2.670</td>
<td>.0224</td>
<td>13.351</td>
<td>.812</td>
</tr>
<tr>
<td>Epoch * Subject(Group)</td>
<td>270</td>
<td>.112</td>
<td>4.142E-4</td>
<td>4.142E-4</td>
<td>.000005</td>
<td>23.130</td>
<td>.980</td>
</tr>
</tbody>
</table>
Figure 11. Correlation of the 2\textsuperscript{nd} Half Block Triplet type effect and Disease duration.

<table>
<thead>
<tr>
<th>Block half 2 overall tt effect, Disease Duration</th>
</tr>
</thead>
<tbody>
<tr>
<td>Correlation</td>
</tr>
<tr>
<td>.396</td>
</tr>
</tbody>
</table>

29 observations were used in this computation.
Figure 12. Correlation of the Triplet type effect in the 2\textsuperscript{nd} Half Block and Levodopa.

<table>
<thead>
<tr>
<th>Levodopa, Second Block half Overall MMRT tt effect</th>
<th>Correlation</th>
<th>P-Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>29 observations were used in this computation.</td>
<td>.343</td>
<td>.0680</td>
</tr>
</tbody>
</table>
Figure 13. Correlation of the Triplet type effect in the 2\textsuperscript{nd} Half Block and Age.

<table>
<thead>
<tr>
<th>Correlation</th>
<th>P-Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>.434</td>
<td>.0177</td>
</tr>
</tbody>
</table>

29 observations were used in this computation.
Figure 14. Correlation of Age x Disease duration.

<table>
<thead>
<tr>
<th>Correlation</th>
<th>P-Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, Disease Duration</td>
<td>.320</td>
</tr>
</tbody>
</table>

29 observations were used in this computation.
Figure 15. Correlation of Age x Levodopa.

29 observations were used in this computation.
Figure 16. Correlation of Disease duration x Levodopa.

Levodopa, Disease Duration  | Correlation  | P-Value
<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>.676</td>
<td>&lt;.0001</td>
</tr>
</tbody>
</table>

29 observations were used in this computation.
Figure 17. Correlation of the overall Triplet type effect and total SCOPA-COG score.

Overall tt effect w Reps/Trills s509 fixed, SCOPA-COG
29 observations were used in this computation.
Figure 18. Correlation of the overall Triplet type effect and total SCOPA-COG Learning and Memory score.

Overall tt effect w Reps/Trills s509 fixed, SCOPA-COG Mem Learn
29 observations were used in this computation.

<table>
<thead>
<tr>
<th>Correlation</th>
<th>P-Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>.405</td>
<td>.0284</td>
</tr>
</tbody>
</table>
Figure 19. Correlation of Individual standard deviation and Hoehn and Yahr stage.

Overall ISD s509 fixed, H&Y numerical
29 observations were used in this computation.
Figure 20. Correlation of Individual standard deviation and Disease duration.

<table>
<thead>
<tr>
<th>Overall ISD s509 fixed, Disease Duration</th>
<th>Correlation</th>
<th>P-Value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>.366</td>
<td>.0500</td>
</tr>
</tbody>
</table>

29 observations were used in this computation.
Figure 21. Correlation of Individual standard deviation and Levodopa.

Overall ISD $s_{509}$ fixed, Levodopa

<table>
<thead>
<tr>
<th>Correlation</th>
<th>P-Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>.553</td>
<td>.0015</td>
</tr>
</tbody>
</table>

29 observations were used in this computation.
**Figure 22.** Correlation of Individual standard deviation and Levodopa equivalence.

Overall ISD s509 fixed, DA Equivalence

<table>
<thead>
<tr>
<th>Correlation</th>
<th>P-Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>.454</td>
<td>.0126</td>
</tr>
</tbody>
</table>

29 observations were used in this computation.
APPENDIX

Scales for Outcomes in Parkinson’s disease – Cognition

Memory and learning

1. Verbal recall

Ten words are repeatedly shown for at least 4 seconds, get the patient to read them out loud, the time allowed for recall is unlimited. Underline each word that has been named. When words are named that were not shown, no penalty is given. When a false answer is changed (e.g. king into queen), it is correct.

Instruction: "Read the following 10 words aloud and try to remember as many as possible. After reading them all, name as many words as possible, the order of the words is not important".

10 words: Butter arm shore letter queen cabin pole ticket grass engine

(10 correct = 5, 8-9 correct = 4, 6-7 correct = 3, 5 correct = 2, 4 correct = 1, ≤ 3 correct= 0)

score ……../5

2. Digit span backward

Ask the patient to repeat a series of numbers backwards; the numbers are read out separately, 1 second per number; if incorrectly repeated, the alternative in the second column is presented. Continue until both the first and the alternative series are repeated incorrectly. Make sure the time interval between numbers stays the same. Read the numbers calmly and make sure the time between numbers is equal. Record the highest series that is repeated correctly at least once; Give an example: "If I say 2-7-3, than you say (3-7-2)

backwards score:

2-4 5-8 = 1
6-2-9 4-1-5 = 2
3-2-7-9 4-9-6-8 = 3
1-5-2-8-6 6-1-8-4-3 = 4
5-3-9-4-1-8 7-2-4-8-5-6 = 5
8-1-2-9-3-6-5 4-7-3-9-1-2-8 = 6
9-4-3-7-6-2-5-8-7-2-8-1-9-6-5-3 = 7

score ……..../7

3. Indicate cubes

Point to the cubes in the order given below; the patient should copy this; do this slowly; the patient decides for himself with which hand he/she prefers. Indicate the cubes in the order as indicated. Observe carefully if the patient copies the order correctly. When a patient wants to correct a mistake, let him/her do the complete order again. This is not counted as a mistake. However, if the patient forgets the order and would like to see the order a second time, the researcher does not repeat the order again but starts with the next order.

1 2 3 4
Attention

4. Counting backwards (30 to 0)

**Instruction**: "Would you subtract three from 30, and subtract three again from the result and continue till zero".

Mistakes can be: the order, missing or not knowing a number, or not finishing off the series. Record the order of numbers named by the patient. If the patient asks where to start or how much to subtract, the researcher repeats the instructions but counts that as one mistake. If the patient makes a mistake but continues from that point to subtract three, it is only one mistake. If the patient stops the order and starts all over again, it is one mistake.

(0 mistakes = 2, 1 mistake = 1, ≥ 2 mistakes = 0) score ………./2

5. Months backwards

**Instruction**: "Name the months of the year in reverse order, starting with the last month of the year".

Mistakes are: the order, missing or not knowing the next month, or not finishing off the series. Underline the months that are named correctly. When a month is passed over, this is a mistake, even if the patient corrects it later on. If the patient stops the order and starts all over again, it is one mistake. If the patient starts naming the month forward, repeat the instructions and count it as one mistake.


(0 mistakes = 2, 1 mistake = 1, ≥ 2 mistakes = 0) score ………./2
Instructions: “Now it is your turn to make the three movements, fist-stretch-palm, 10 times in a row. You don’t have to count, I will tell you when to stop”.

Note the number of correct trios from a total of 10; Count carefully but not out loud. Every time a patient makes a wrong movement, count it as a mistake, even when the patient corrects it halfway.

(10 correct = 3, 9 correct = 2, 8 correct = 1, ≤ 7 correct = 0)

score ……../3

7. Semantic fluency

Tell the patient to name as many animal as he/she knows in one minute. Note all answers that are given by the patient. No repetition or variations of words, such as lion-lioness, tiger-tigress; categories are allowed, bird and pigeon are both correct. Count the number of animals correctly named. The purpose is that the patient generates the animals actively, therefore no clues are allowed. When the patient asks whether, for instance, naming different types of birds is allowed, this may be confirmed. When the patient almost immediately says he/she does not know any more animals, try to SCOPA-COG stimulate the patient by saying “there is still a lot of time left”, but do not give clues. When the patient starts naming other things than animals, do not correct the patient. Naming other things besides animals is not counted as an additional mistake.

(≥ 25 correct = 6, 20-24 = 5, 15-19 = 4, 10-14 = 3, 5-9 = 2, 1-4 = 1 0=0)

number of animals correct: …….

score ……../6

Write down all animals named:

8. Dice

Use 2 cards, one with YES = EVEN, NO = ODD; one with YES = HIGHER, NO = LOWER. Put the correct card face up next to the explanation of the test and make sure that the other, irrelevant card is out of sight. The first round (situation 1) is not scored, and the patient is corrected if necessary.

**Situation 1: YES = EVEN**

Put the card “YES=EVEN, NO=ODD” on the table and leave it there during the test. **Instruction:** "Say YES for an even number on a dice and NO for an odd number, when you see a picture of a dice with an EVEN number of pips, I would like you to say YES, and NO when the number of pips is ODD".
Show the first two examples (3 even and 3 odd dices) and ask the patient “If you see one of these dice, do you say yes or no?” Tell the patient if the answer is correct or not. If the answer is not correct, explain why. It is important that the patient says YES or NO and not EVEN or ODD. Show the next two examples (with only one dice) and ask the patient “if you see this dice, do you say yes or no?” Tell the patient if the answer is correct or not. If the answer is not correct, explain why.

Then show the patient the following 10 dices. Correct the patient if the answer is wrong.

**Situation 2: YES = HIGHER**

With the card “example 1” (dice with 3 pips) the next condition starts. Put the card “YES=HIGHER, NO=LOWER” on the table and remove the former card.

**Instruction:** “Now, we change the test a little. When you see a picture of a dice that is higher than the dice on the page before, you say YES. When the dice is lower, you say NO”.

Tell the patient you have an example (example 1). “Try to remember this dice” (turn the page) “Is this YES or NO?” Tell the patient whether the answer is correct or not. If the answer is not correct, explain why. Continue with example 2 and say “now remember this dice” (turn the page) “Is this YES or NO?” Tell the patient if the answer is correct or not. If the answer is not correct, explain why.

Then start the test and show all 10 dices one after another. The first response counts and corrections are not allowed. Do NOT correct when a wrong answer is given. If a patient corrects a wrong answer, it is still counted as a mistake. If the patient asks for the instruction, the researcher explains but that is counted as one mistake.

(10 correct = 3, 9 correct = 2, 8 correct =1, ≤ 7 correct = 0)

**Visuo-spatial functions**

9. **Assembling patterns**

The patient is shown 5 incomplete patterns and has to choose 2 or 3 shapes out of 4 to 6 possible alternatives in order to complete the pattern. First practice 2 figures.

Show the patient example A and give the instruction to choose the shapes that form the pattern. Tell the patient if the answer is correct or not. If the answer is not correct, explain why and give the correct solution. Repeat this with example B. Then show the 5 patterns. Do not tell the patient whether the answer is correct or not. There is no time limit. If the patient corrects a wrong answer, this is not counted as a mistake.

a. b. c. d. e.

**score ……. /5**
Memory

10. Delayed recall

**Instruction**: "Can you name as many as possible of the 10 words that you learned during the first test?"

Underline each word that has been named. When words are named that were not shown, no penalty is given. When a false answer is changed (e.g. king into queen), it is correct.

10 words: butter arm shore letter queen cabin pole ticket grass engine

(10 correct = 5, 8-9 correct = 4, 6-7 correct = 3, 5 correct = 2, 4 correct = 1, ≤ 3 correct= 0)

correct words: …. /10

correct words: … /5

Total COG score: … /43

© This questionnaire is made available free of charge, with the permission of the authors, to all those undertaking non-profit and profit making research. Future users may be requested to share data for psychometric purposes. Use of this questionnaire in studies should be communicated to the developers. No changes may be made to the questionnaire without permission. Please use the following reference in publications: Marinus J, Visser M, Verwey NA, Verhey FRJ, Middelkoop HAM, Stiggelbout AM, van Hilten JJ. Assessment of cognition in Parkinson’s disease. *Neurology* 2003;61:1222-1228. For further information, please contact Dr. J. Marinus, Leiden University Medical Center, Department of Neurology (K5Q), P.O. Box 9600, NL-2300 RC Leiden (email: j.marinus@lumc.nl).
Mini Mental State Examination

MINI MENTAL STATUS EXAMINATION

Patient Name: ___________________________ Date: ___________ Rater: ___________

Orientation

Correct Incorrect

___ ___ What is today’s date?
___ ___ What is the year?
___ ___ What is the month?
___ ___ What day is today?
___ ___ What season is it?
___ ___ What is the name of this place?
___ ___ What floor are we on?
___ ___ What district [town, city] are we in?
___ ___ What area [county, district, area] are we in?
___ ___ What state are we near [in]?

Immediate Recall

Tell the patient “I’m going to name three objects. After I have said them, I want you to repeat them. Remember what they are because I’m going to ask you to name them again in a few minutes.” First repetition determines score (0-3), but repeat all three, up to six trials.

Correct Incorrect

___ ___ Ball
___ ___ Flag
___ ___ Tree

Attention and Calculation

Ask the patient to spell “world” backwards. Indicate the first five letters of the patient’s response on the lines provided.

Correct Incorrect

___ ___ D
___ ___ L
___ ___ R
___ ___ O
___ ___ W

Delayed Recall

Ask the patient to recall the three words you previously asked him/her to remember.

Correct Incorrect

___ ___ Ball
___ ___ Flag
___ ___ Tree

Language

Correct Incorrect

Show the patient a wrist watch and ask him/her to name it.
Show the patient a pencil and ask him/her to name it.

Ask the patient to repeat “No ifs, ands, or buts”.

Give the patient a plain piece of paper and say,
“Take the paper in your right hand,
fold it in half,
and put it on the floor”.

Hold the piece of paper which reads, “close your eyes” so the patient can see it clearly. Ask him/her to read it and do what it says. Score correct only if the patient actually closes his/her eyes.

Give the patient a blank piece of paper and ask him/her to write a sentence. It is to be written spontaneously. It must contain a subject and a verb and be sensible. Correct grammar and punctuation are not necessary.

On the page provided, ask the patient to draw the intersecting pentagons. All 10 angles must be present and two must intersect. Tremor and rotation are ignored.
Montreal Cognitive Assessment

**Montreal Cognitive Assessment (MOCA)**

**Version 7.1 Original Version**

**VISUOSPATIAL / EXECUTIVE**

- **Copy cube**
  - Draw **CLOCK** (Ten past eleven) (3 points)
  - [ ] Contour
  - [ ] Numbers
  - [ ] Hands
  - __5__/5

**NAMING**

- [ ] Rhinoceros
- [ ] Lion
- [ ] Wolf

**MEMORY**

Read list of words, subject must repeat them. Do 2 trials, even if 1st trial is successful.

- 1st trial: [ ]
- 2nd trial: [ ]

**ATTENTION**

Read list of letters. The subject must tap with his hand at each letter A. No points if ≥ 2 errors


Serial 7 subtraction starting at 100:

- [ ] 93
- [ ] 86
- [ ] 79
- [ ] 72
- [ ] 65

[ ] 4 or 5 correct subtractions: 3 pts., 2 or 3 correct: 2 pts., 1 correct: 1 pt., 0 correct: 0 pt

**LANGUAGE**

Repeat: I only know that John is the one to help today.

The cat always hid under the couch when dogs were in the room.

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**

- FACE [ ]
- VELVET [ ]
- CHURCH [ ]
- DAISY [ ]
- RED [ ]

Points for UNCUED recall only [ ]

**Optional**

- Category cue
- Multiple choice cue

**ORIENTATION**

- [ ] Date
- [ ] Month
- [ ] Year
- [ ] Day
- [ ] Place
- [ ] City

**TOTAL** [ ]

129
Geriatric Depression Scale (Short form)

GDS Scoring Guide

Bolded Items Receive a Point

1. Are you basically satisfied with your life? ... YES / NO
2. Have you dropped many of your activities and interests? ... YES / NO
3. Do you feel that your life is empty? ... YES / NO
4. Do you often get bored? ... YES / NO
5. Are you in good spirits most of the time? ... YES / NO
6. Are you afraid that something bad is going to happen to you? ... YES / NO
7. Do you feel happy most of the time? ... YES / NO
8. Do you often feel helpless? ... YES / NO
9. Do you prefer to stay at home, rather than going out and doing new things? ... YES / NO
10. Do you feel you have more problems with memory than most? ... YES / NO
11. Do you think it is wonderful to be alive now? ... YES / NO
12. Do you feel pretty worthless the way you are now? ... YES / NO
13. Do you feel full of energy? ... YES / NO
14. Do you feel that your situation is hopeless? ... YES / NO
15. Do you think that most people are better off than you are? ... YES / NO

Interpreting the GDS Short Form

TOTAL: Please sum all bolded answers (worth one point) for a total score.______________

Scores: 0-5 Normal 5-10 Suggestive of Depressive 11-15 Almost Always Depression

North American Adult Reading Test – 35

1. Debris
2. Simile
3. Subtle
4. Bouquet
5. Colonel
6. Rarefy
7. Gist
8. Corps
9. Hors d’oeuvre
10. Sieve
11. Hiatus
12. Gauche
13. Zealot
14. Paradigm
15. Façade
16. Cellist
17. Indict
18. Détente
19. Impugn
20. Aeon
21. Epitome
22. Reify
23. Indices
24. Assignate
25. Topiary
26. Caveat
27. Leviathan
28. Quadruped
29. Sidereal
30. Abstemious
31. Beatify
32. Gaoled
33. Demesne
34. Syncope
35. Ennui
Perceived Stress Scale

The questions in this scale ask you about your feelings and thoughts during the last month. In each case, you will be asked to indicate by circling how often you felt or thought a certain way.

Name ____________________________ Date __________
Age ______ Gender (Circle): M F Other __________________________

<table>
<thead>
<tr>
<th>Item</th>
<th>Code</th>
<th>0 = Never</th>
<th>1 = Almost Never</th>
<th>2 = Sometimes</th>
<th>3 = Fairly Often</th>
<th>4 = Very Often</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. In the last month, how often have you been upset</td>
<td></td>
<td>0</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>because of something that happened unexpectedly?</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2. In the last month, how often have you felt that you were unable</td>
<td></td>
<td>0</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>to control the important things in your life?</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>3. In the last month, how often have you felt nervous and “stressed”?</td>
<td></td>
<td>0</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>4. In the last month, how often have you felt confident about your</td>
<td></td>
<td>0</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>ability to handle your personal problems?</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>5. In the last month, how often have you felt that things were</td>
<td></td>
<td>0</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>going your way?</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>6. In the last month, how often have you found that you could not</td>
<td></td>
<td>0</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>cope with all the things that you had to do?</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>7. In the last month, how often have you been able</td>
<td></td>
<td>0</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>to control irritations in your life?</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>8. In the last month, how often have you felt that you were on top</td>
<td></td>
<td>0</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>of things?</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>9. In the last month, how often have you been angry because of</td>
<td></td>
<td>0</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>things that were outside of your control?</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>10. In the last month, how often have you felt difficulties were</td>
<td></td>
<td>0</td>
<td>1</td>
<td>2</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>piling up so high that you could not overcome them?</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Please feel free to use the Perceived Stress Scale for your research.

Mind Garden, Inc.
1690 Woodside Road, Suite #202
Redwood City, CA 94061 USA
Phone: (650) 261-3500 Fax: (650) 261-3505
e-mail: mindgarden@msn.com
www.mindgarden.com

References
Health Screening Questionnaire

Date_______ Subject Number____

HEALTH SCREENING QUESTIONNAIRE

(Circle ‘Y’ for yes and ‘N’ for no)

1. Have you ever had a stroke or a transient ischemic attack (TIA)? Y N
2. Do you have Parkinson’s disease? Y N
3. Do you have multiple sclerosis, cerebral palsy, or Huntington’s disease? Y N
4. Have you ever had brain surgery? Y N
5. Do you have diabetes that requires insulin control? Y N
6. Do you have hypertension that is not well controlled? Y N
7. Have you had cancer diagnosed within the past three years (not including skin cancer)? Y N
8. a. Have you ever had a heart attack? Y N
   b. If yes, did your memory, ability to talk, or ability to solve problems change? Y N
9. Are you currently taking medications for mental or emotional problems? Y N
10. Have you ever been unconscious for more than one hour other than during surgery? Y N
11. Have you ever had any permanent decrease in memory or other mental function? Y N
12. Do you have trouble with your vision (reading normal print) even when you have glasses on? Y N
13. Have you ever been resuscitated? Y N
14. Have you had a head injury with loss of consciousness greater than five minutes? Y N
15. Have you ever been diagnosed as learning disabled? Y N
16. Have you ever been diagnosed as having a brain tumor? Y N
17. Do you have difficulty using your hands (i.e. arthritis)? Y N
18. Please rate your overall level of health on a scale from one to five (1 = poor, 5 = excellent overall health) (circle one) 1 2 3 4 5
19. Have you or your parents, siblings, or children ever been diagnosed with (or been suspected of having)

<table>
<thead>
<tr>
<th>Condition</th>
<th>Yourself</th>
<th>Parent/Sibling/Child</th>
</tr>
</thead>
<tbody>
<tr>
<td>Speech or language deficits (Speech Language Impairment)</td>
<td>Y N</td>
<td>Y N</td>
</tr>
<tr>
<td>Learning disability (dyslexia, ADHD)</td>
<td>Y N</td>
<td>Y N</td>
</tr>
<tr>
<td>Seizures</td>
<td>Y N</td>
<td>Y N</td>
</tr>
<tr>
<td>Anxiety</td>
<td>Y N</td>
<td>Y N</td>
</tr>
<tr>
<td>Panic disorder</td>
<td>Y N</td>
<td>Y N</td>
</tr>
<tr>
<td>Schizophrenia</td>
<td>Y N</td>
<td>Y N</td>
</tr>
<tr>
<td>Bipolar (manic depressive disorder)</td>
<td>Y N</td>
<td>Y N</td>
</tr>
<tr>
<td>Major depressive disorder</td>
<td>Y N</td>
<td>Y N</td>
</tr>
<tr>
<td>Obsessive compulsive disorder</td>
<td>Y N</td>
<td>Y N</td>
</tr>
</tbody>
</table>
20. Please list any medications you are taking (name of medication, dosage, and frequency)

21. Please list any vitamins or supplements you may be taking (name, dosage, frequency). For example, ginkgo biloba, vitamin E, chondroitin glucosamine, etc.

22. How many caffeinated beverages do you drink each day?
   ____ 0
   ____ 1-2
   ____ 2 or more

23. Do you drink alcoholic beverages? YES NO
   What do you drink most often? BEER WINE LIQUOR
   If so, how often?
   ____ Daily
   ____ A few times a week
   ____ A few times a month
   ____ Only on special occasions/infrequently
Biographical Questionnaire

Date_________ Subject Number____

1. SEX (circle one)     M     F

2. DATE OF BIRTH: ____________

   (Since the National Institute of Health would like to ensure minority representation, we ask you to indicate your ethnicity and race. However, if you would rather not respond, please leave the following question unanswered.)

3. RACIAL CATEGORIES (check one)
   American Indian / Alaska Native
   Asian
   Native Hawaiian/Other Pacific Islander
   Black or African American
   Caucasian
   More than one race
   Unknown
   Other

4. ETHNIC CATEGORIES (check one)
   Hispanic or Latino
   Not Hispanic or Latino
   Unknown

5. MARITAL STATUS (circle one) Single Married Divorced Widowed

6. OCCUPATION (If retired, please indicate so and specify your primary occupation prior to retirement)_____________________________________________________________________

7. EDUCATION / DEGREE’s COMPLETED
   Did you obtain a HIGH SCHOOL diploma? Yes No Year? _____
   Did you obtain a BACHELOR’s degree? Yes No Year? _____
   If not, please indicate the number of years of undergraduate education completed _____
   Did you obtain a MASTER’s degree? Yes No Year? _____
   Did you obtain a JURIS DOCTORATE degree? Yes No Year? _____
   Did you obtain a DOCTORATE degree? Yes No Year? _____

8. IS ENGLISH YOUR NATIVE LANGUAGE? Yes No

   ARE YOU BILINGUAL OR MULTILINGUAL? Yes No
   If yes, what languages do you speak? ____________________________
   What is your preferred or most fluent language? __________
   What is your primary language? __________

9. ARE YOU RIGHT HANDED? Yes No Ambidextrous
Post-Triplets Questionnaire

1) Did you notice anything about the tasks you have performed?

2) Did you notice any repeating patterns within the tasks?

3) Did you use any particular strategies?

4) There were, in fact, regularities in the tasks you just completed. Knowing this, could you describe any of these regularities?