The Impact of Parkinson's Disease on Mammalian Adult Neurogenesis

Joseph Bastasic

Thesis submitted to the University of Ottawa
in partial fulfillment of the requirements for the
M.Sc. program in Neuroscience

Department of Cellular and Molecular Medicine
Faculty of Medicine
University of Ottawa

© Joseph Bastasic, Ottawa, Canada, 2019

ABSTRACT

Parkinson's disease (PD) has been reported to negatively affect adult neurogenesis. Mitochondrial dysfunction associated with PD may be involved, given that recent studies have identified mitochondria to be central regulators of neural stem cell (NSC) fate decisions. For this thesis, we sought to characterize adult neurogenesis in PINK1 and parkin knockout (KO) mouse models of PD. Immunohistochemical staining of subventricular zone (SVZ) and subgranular zone (SGZ) tissue sections from 6 month old mice was performed in order to identify and quantify changes in specific cell populations involved with adult neurogenesis. The loss of PINK1 or parkin was found to cause aberrant changes in adult neurogenesis, particularly in the SGZ. Going forward, it would be interesting to determine if the observed changes in adult neurogenesis were the result of mitochondrial dysfunction.

TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

LIST OF ABBREVIATIONS

^{14}C	Carbon-14
^{3}H-thymidine	Tritiated thymidine
6-OHDA	6-hydroxydopamine
aa	Amino acid
Acetyl-CoA	Acetyl coenzyme A
AIF	Apoptosis-inducing factor
α-KG	α- ketoglutarate
ANOVA	Analysis of variance
ATP	Adenosine triphosphate
ATP13A2	ATPase Type 13A2
BLBP	Brain lipid-binding protein
BNIP3	BCL2/adenovirus E1B 19 kDa protein-interacting protein 3
BrdU	5-bromo-2'-deoxyuridine
Ca^{2+}	Calcium ion
CBIA	Cell Biology and Image Acquisition Core
CCCP	Carbonyl cyanide m-chlorophenyl hydrazone
CD133	Prominin-1
CIHR	Canadian Institutes of Health Research
CNS	Central Nervous System
DAPI	4',6-diamidino-2-phenylindole
DAT	Dopamine transporter
Dcx	Doublecortin
DG	Dente gyrus
DJ-1	Daisuke-Junko-1
Dlx2	Distal-less homeobox 2
DNA	Deoxyribonucleic acid
Drp1	Dynamin-related protein 1
$\Delta\Psi m$	Mitochondrial membrane potential
ECAR	Extracellular acidification rate
EdU	5-ethynyl-2'-deoxyuridine
EGFR	Epidermal growth factor receptor
ETC	Electron transport chain
FKBP8	FK506-binding protein 8
FUNDC1	FUN14 domain containing 1
GAD65	Glutamate decarboxylase enzyme 65
GAPDH	Glyceraldehyde 3-phosphate dehydrogenase
GFAP	Glial fibrillary acidic protein
Gp78	Glycoprotein 78
GTP	Guanosine-5'-triphosphate
HPRT	Hypoxanthine phosphoribosyltransferase
IBR	In-between RING

IF1	Inhibitory factor 1
IMM	Inner mitochondrial membrane
KO	Knockout
Lhx5	LIM (Lin11, Isl1, and Mec-3) homeobox 5
LRRK2	Leucine-rich repeat kinase 2
MAD	Mitochondria-associated degradation
MAO-B	Monoamine oxidase B
Mash1	Mammalian achaete scute homolog-1
MCM2	Minichromosome maintenance complex component 2
Mdivi-1	Mitochondrial Division Inhibitor 1
MDV	Mitochondria-derived vesicle
MFN1	Mitofusin 1
MFN2	Mitofusin 2
Miro	Mitochondrial Rho
MitAP	Mitochondrial antigen presentation
mito-QC	mCherry-GFP-FIS1 (mitochondrial fission 1 protein)$^{101-152}$ transgenic reporter
MPC	Mitochondrial pyruvate carrier
MPP	Mitochondrial processing peptidase
MPP^+	1-methyl-4-phenylpyridinium
MPTP	1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine
mRNA	Messenger RNA
MSc	Master of Science
mtDNA	Mitochondrial DNA
mt-Keima	Mitochondria-matrix targeted Keima
MTS	Mitochondria targeting sequence
MUL1	Mitochondrial E3 ubiquitin protein ligase 1
NAD	Nicotinamide adenine dinucleotide
NAD^+	Oxidized nicotinamide adenine dinucleotide
Neo	Neomycin
NeuN	Neuronal nuclei
NIX	NIP3-like protein X
Nkx2.1	NK2 homeobox 1
NR	Nicotinamide riboside
NRF2	Nuclear factor erythroid 2–related factor
NSC	Neural Stem Cell
OB	Olfactory Bulb
OCR	Oxygen consumption rate
OMM	Outer mitochondrial membrane
OMS	Outer mitochondrial membrane localization signal
Opa1	Optic atrophy protein 1
$Opa1^{tg}$ mice	Opa1 overexpressing transgenic mice
OXPHOS	Oxidative phosphorylation
PARIS	Parkin interacting substrate

PARL	PINK1/PGAM5 (phosphoglycerate mutase family member 5)-associated rhomboid-like protease (formally known as presenilin-associated rhomboid-like protease)
PBS	Phosphate-buffered saline
PCR	Polymerase chain reaction
PD	Parkinson's disease
PDK	Pyruvate dehydrogenase kinase
PFA	Paraformaldehyde
PGC-1α	Peroxisome proliferator-activated receptor (PPAR)-γ coactivator-1α
PGK	Phosphoglycerate kinase
PGK-HPRT	Human hypoxanthine phosphoribosyltransferase gene construct coupled with a phosphoglycerate kinase promoter element
PGK-Neo	Phosphoglycerate kinase-neomycin
PINK1	Phosphatase and tensin homolog (PTEN)-induced putative kinase 1
PNS	Peripheral Nervous System
POLG	DNA polymerase γ
Prox1	Prospero homeobox protein 1
REP	Repressor of park
RGL	Radial glial-like
RING	Really Interesting New Gene
RING0	Really Interesting New Gene 0
RING1	Really Interesting New Gene 1
RING2	Really Interesting New Gene 2
RMS	Rostral migratory stream
RNA	Ribonucleic acid
RNAseq	RNA sequencing
ROS	Reactive oxygen species
SD	Standard deviation
Ser65	Serine at amino acid position 65
SGZ	Subgranular zone
SIAH1	Seven in absentia homolog 1
Sim1	Single-minded homolog 1
SN	Substantia nigra
SNpc	Substantia nigra pars compacta
SOD1	Superoxide dismutase 1
SOD2	Superoxide dismutase 2
SODPAR	Compound parkin KO and SOD2 hemizygous KO
Sox2	SRY (sex determining region Y)-box 2
SVZ	Subventricular zone
TBK1	TANK [TRAF (tumor necrosis factor receptor-associated factor) family member-associated NF-κB (nuclear factor κ-light-chain-enhancer of activated B cells) activator]-binding kinase 1
Tbr2	T-box brain protein 2
TFAM	Mitochondrial transcription factor A

TIM23	Translocase of the inner membrane 23
TMD	Transmembrane domain
TOM	Translocase of the outer membrane
Tom20	Translocase of the outer membrane subunit 20
TRAK	Trafficking kinesin protein
Type I cell	SGZ neural stem cell
Type II cell	SGZ intermediate progenitor cell
Type III cell	SGZ neuroblast
Type A cell	SVZ neuroblast
Type B cell	SVZ niche specific astrocyte
Type B1 cell	SVZ neural stem cell
Type B2 cell	Non-neurogenic SVZ niche specific astrocyte
Type C cell	SVZ transit amplifying progenitors
Type E cell	Ependymal cell
Type E1 cell	Multiciliated ependymal cell
Type E2 cell	Bi-ciliated ependymal cell
Ub	Ubiquitin
Ubl	Ubiquitin-like
UCP2	Uncoupling protein 2
WT	Wild-type
ZNF746	Zinc finger protein 746

ACKNOWLEDGEMENTS

There are many people who have fostered the incredibly rich and collaborative learning environment in which I completed this project. I am forever grateful to those who made this experience so rewarding.

First and foremost, I would like to thank my supervisor, Dr. Ruth Slack, who expertly guided me through my MSc thesis project. Without her unwavering patience, support, and encouragement, this paper would not be possible.

I would also like to extend my deepest appreciation to the members of the Slack lab. I am grateful for the assistance and dedication of past and present lab managers, Jason MacLaurin and Smitha Paul, respectively. I owe nearly all of my practical lab knowledge to Bensun Cambell Fong. I appreciate the countless hours he spent teaching me invaluable skills that will further my career as a scientist. Dr. Mireille Khacho deserves special acknowledgement for her help in setting up the conceptual framework of this project. My research is largely based off her work on mitochondrial regulation of neural stem cell (NSC) fate decisions. I wish her all the best with her new lab. I offer special thanks to Richard Harris for his assistance in the strategic planning of mouse breeding and experiments. I wish to sincerely thank Mohamed Ariff Iqbal for all his help with the technical aspects of experimental planning. I am very grateful for the assistance that Edward Yakubovich provided with all of the technical issues regarding immunohistochemistry and image processing. Finally, I would like to thank Maria Bilen for her prompt support throughout the writing of this dissertation. I have thoroughly enjoyed working with all of the wonderful members of the Slack Lab.

I would like to sincerely thank my thesis advisory committee members Dr. David Park and Dr. Jing Wang for their contributions to this project. I am grateful for their superb guidance throughout this process.

I would also like to thank Dr. Chloë van Oostende-Triplet and Skye McBride from the Cell Biology and Image Acquisition Core (CBIA) for all of their assistance. Their expertise on microscopy proved to be invaluable for this project.

I would like to acknowledge the labs that provided the mice used in this work. Dr. David Park's lab provided me with the PINK1 KO mice. Dr. Michael Schlossmacher's lab provided me with the Parkin KO, SOD2 heterozygous KO, and SODPAR mice.

I would also like to acknowledge the financial support from the Canadian Institutes of Health Research (CIHR).

Finally, I would like to thank my family and friends for their enduring love and support. I am deeply appreciative of your positive outlook, words of encouragement, and ongoing interest in my thesis work.

INTRODUCTION

1.1. Neurogenesis in the Adult Mammalian Brain

1.1.1. The Early History of Adult Neurogenesis

A long held belief in the field of neuroscience was the idea that neurogenesis only occurred during embryonic and early postnatal stages of development (Gross, 2000). This view started to change in the 1960's, when ^{3}H-thymidine labelling experiments identified the presence of newborn neurons in the hippocampus and olfactory bulbs (OBs) of adult rats (Altman, 1969; Altman and Das, 1965). Studies in several different animal models and humans were able to explain these findings with the discovery of two distinct neurogenic niches in the adult mammalian brain: (1) the subgranular zone (SGZ) of the hippocampal dente gyrus (DG) and (2) the subventricular zone (SVZ) lining the lateral ventricles (Bond et al., 2015; Eriksson et al., 1998; Gage et al., 1995; Lois and Alvarez-Buylla, 1993, 1994; Lois et al., 1996; Palmer et al., 1997). In both of these niches, neural precursor cells, which refer to both neural stem cells (NSCs) and progenitor cells, are able to undergo differentiation and maturation to become mature, post-mitotic neurons. The discovery of continued neurogenesis in the adult mammalian brain offers several promising avenues of research, particularly with respect to neural regenerative medicine (Bond et al., 2015).

For the purposes of this dissertation, the fundamentals of adult neurogenesis in the SVZ and SGZ will be described with respect to rodent animal models. Evidence of adult neurogenesis and its significance in humans will also be discussed.

1.1.2. Adult Neurogenesis in the SVZ

The adult SVZ neurogenic niche is defined by the presence of four major cell populations: type E, B, C, and A cells (Doetsch et al., 1997). Type E cells (ependymal cells) are multiciliated (Type E1) or bi-ciliated (Type E2) cells lining the lateral ventricles that are responsible for regulating cerebrospinal fluid circulation with their cilia (Mirzadeh et al., 2008; Sawamoto et al., 2006). Type B cells are niche-specific astrocytes that can be divided into two subgroups based on their localization in the niche. Type B1 cells span the entire SVZ by making contact with both ependymal cells and SVZ blood vessels, while type B2 cells are only found surrounding type A cells (Doetsch et al., 1997; García-Verdugo et al., 1998). Type B1 cells are considered to be the NSCs of this niche and can exist in a quiescent or activated state (Capilla-Gonzalez et al., 2014; Codega et al., 2014; Doetsch et al., 1999a, 1999b; Mirzadeh et al., 2008; Obernier and Alvarez-Buylla, 2019). Activated type B1 cells undergo differentiation to give rise to type C cells (transit-amplifying progenitors), which in turn become type A cells (neuroblasts) (Doetsch et al., 1997). In rodents, these newly generated neuroblasts migrate along the rostral migratory stream (RMS) in order to reach the OBs to become mature granule or periglomerular cells (Doetsch and Alvarez-Buylla, 1996). These newly integrated neurons are currently thought to be involved with short-term olfactory memory (Breton-Provencher et al., 2009).

1.1.3. Adult Neurogenesis in the SGZ

Similar to the SVZ, adult neurogenesis in the SGZ is based on the step-wise differentiation of astrocyte-like NSCs into mature neurons (Seri et al., 2001). The current model of neurogenesis in the adult SGZ argues that the NSCs (Type I cells) of this niche are comprised of a mixed population of SRY (sex determining region Y)-box 2 (Sox2) positive cells that exhibit either radial

glial-like (RGL) or horizontal morphologies (Suh et al., 2007). The RGL cell subpopulation is relatively quiescent, while the horizontal cell subpopulation is comprised of both quiescent and activated cells (Lugert et al., 2010; Shin et al., 2015). Once activated, NSCs will differentiate into intermediate progenitor cells (Type II cells), that later give rise to neuroblasts (Type III cells) (Kronenberg et al., 2003; Seri et al., 2001). Newly generated neuroblasts will undergo radial migration in the granule cell layer and eventually become mature granule cells (Kempermann et al., 2004). While the exact function of these newborn neurons has not been fully elucidated, adult neurogenesis in the SGZ has been shown to affect spatial-temporal memories and pattern-separation ability (Clelland et al., 2009; Ko et al., 2009; Nakashiba et al., 2012; Saxe et al., 2006; Tronel et al., 2012). Figure 1 provides a summary of adult neurogenesis in the SVZ and SGZ.

1.1.4. Adult Neurogenesis in Humans

Evidence of adult neurogenesis in the human SVZ and SGZ was first obtained from a study analyzing BrdU (5-bromo-2'-deoxyuridine) incorporation in post-mortem brains from cancer patients (Eriksson et al., 1998). Subsequent studies based on immunohistochemistry and the study of cell cultures derived from cells obtained from the human SVZ and SGZ provided additional evidence supporting the existence of human adult neurogenesis (Curtis et al., 2007; Göritz and Frisén, 2012; Johansson et al., 1999; Knoth et al., 2010; Sanai et al., 2004, 2011; Wang et al., 2011a). One interesting finding was that neurogenesis in the human SVZ was found to only meaningfully contribute to the production of new OB neurons until about 18 months of age, as opposed to the lifelong production of OB neurons from neurogenesis in the SVZ of rodents (Sanai et al., 2011). In order to investigate the relevance of adult neurogenesis in humans, researchers

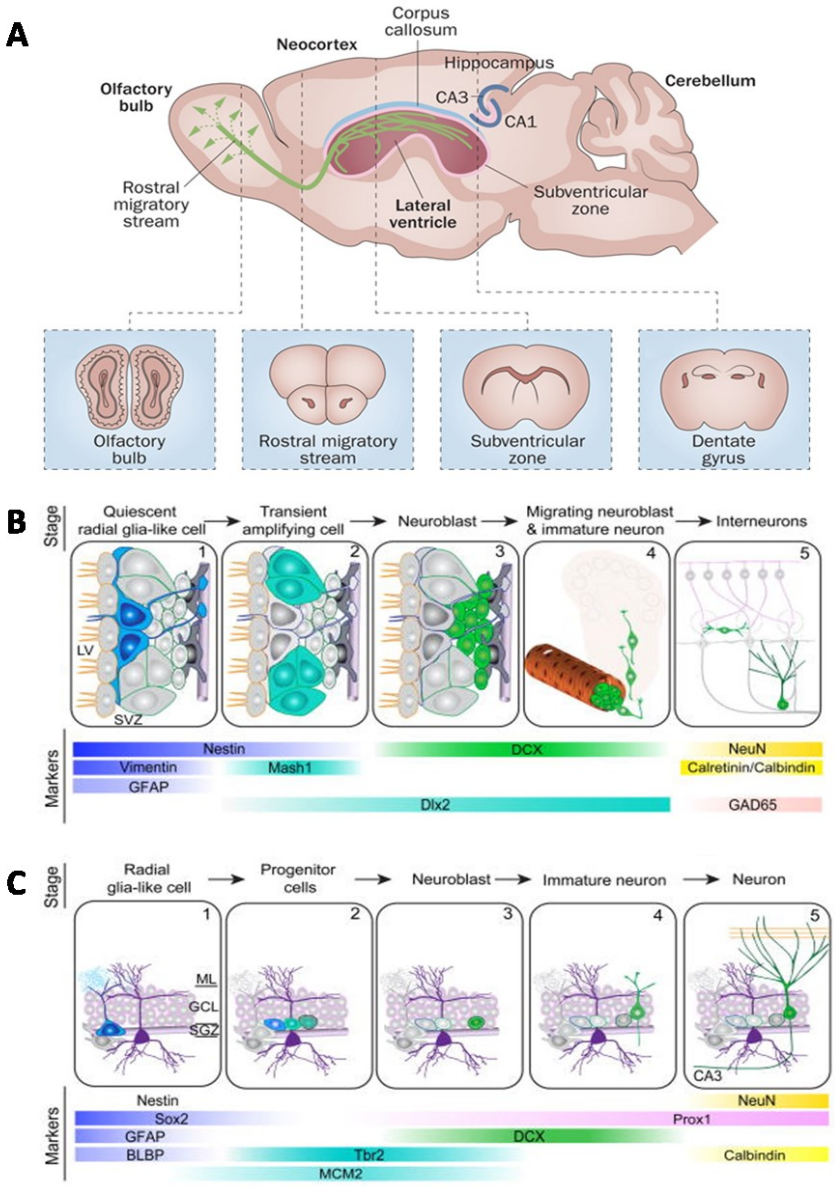

Figure 1: Overview of adult neurogenesis in the rodent brain. (A) Diagram of the adult rodent brain from a sagittal viewpoint. Zoomed-in images represent coronal sections of key areas involved with adult neurogenesis in the subventricular zone (SVZ) and the subgranular zone (SGZ). (B) Diagram of the progression of SVZ adult neurogenesis. (C) Diagram of the progression of SGZ adult neurogenesis. Adult neurogenesis in the SVZ (B) and SGZ (C) can be studied using different combinations of biomarkers to identify specific cell populations in both neurogenic niches. BLBP, Brain lipid-binding protein; DCX, doublecortin; Dlx2, Distal-less homeobox 2; GAD65, Glutamate decarboxylase enzyme 65; GFAP, glial fibrillary acidic protein; Mash1, mammalian achaete scute homolog-1; MCM2, Minichromosome maintenance complex component 2; NeuN, neuronal nuclei; Prox1, Prospero homeobox protein 1; Sox2, SRY (sex determining region Y)-box 2; Tbr2, T-box brain protein 2. Adapted from (Ming and Song, 2011; Ziegler et al., 2015).

decided to assess neuronal turnover by using carbon dating (comparing ^{14}C levels in genomic DNA to atmospheric ^{14}C levels over time) to retrospectively determine when particular cell populations were born (Bergmann et al., 2012; Ernst et al., 2014; Spalding et al., 2013). By using this carbon dating based retrospective birth-dating approach, it was discovered that approximately 35% of neurons in the human hippocampus (corresponding to the dente gyrus) are able to undergo neuronal exchange at a relatively consistent rate of approximately 1.75%/year (Spalding et al., 2013). Adult neurogenesis in the human SVZ was also found to contribute to the production of new neurons in the striatum (with approximately 25% of neurons in the striatum able to undergo neuronal exchange at a relatively consistent rate of 2.7%/year) and not in the OBs (Bergmann et al., 2012; Ernst et al., 2014). The presence of significant neuronal turnover in the adult human brain (Ernst et al., 2014; Spalding et al., 2013) supports a potential functional role for adult neurogenesis in humans (Bergmann et al., 2015; Kempermann et al., 2018).

The purpose of studying adult neurogenesis in rodents is to gain a better understanding of endogenous NSCs, which can then be applied for the human brain. At a fundamental level, the process of adult neurogenesis is relatively conserved amongst species (Ernst and Frisén, 2015). The study of adult neurogenesis in humans is difficult due to several technical issues, such as post-mortem tissue sample handling (Boekhoorn et al., 2006) and a lack of reliable cell markers for the quantitative study of human adult neurogenesis (Mathews et al., 2017). In fact, two recent studies published in high impact journals obtained opposite conclusions on the longevity and significance of adult neurogenesis in the human SGZ (Boldrini et al., 2018; Sorrells et al., 2018) as a result of differences between their methodologies (Lee and Thuret, 2018). Future studies will need to focus on resolving technical issues involved with studying adult neurogenesis in humans, in order to

obtain meaningful results that can contribute to the development of novel therapeutic strategies for the brain (Kempermann et al., 2018).

1.1.5. NSC Fate Decisions

A key area of research in the field of adult neurogenesis is the study of cell fate decisions for NSCs. In the SVZ and SGZ, the differentiation and maturation of NSCs is a tightly regulated process since a balance must be achieved between NSC self-renewal and differentiation. NSCs must undergo differentiation to generate new mature neurons, but NSCs must also undergo self-renewal to prevent the depletion of the NSC pool. This conundrum leads to a simplistic, but nevertheless critical question on what drives NSCs towards self-renewal or differentiation (Ito and Suda, 2014; Shohayeb et al., 2018).

Regulation of NSC fate decisions is achieved through a combination of external and internal signaling mechanisms that alter the expression of genes involved with quiescence, self-renewal, and neuronal differentiation (Faigle and Song, 2013; Shohayeb et al., 2018). NSCs within the brain exist in defined neurogenic niches that provide these cells with a unique microenvironment (Conover and Notti, 2008). These neurogenic niches are comprised of several types of cells (Doetsch et al., 1997; Mirzadeh et al., 2008; Seri et al., 2004) and exhibit extensive vascularization (Shen et al., 2008; Sun et al., 2015; Tavazoie et al., 2008). NSCs receive external signalling cues from their niche, which are known to affect cell fate decisions. These external signals are comprised of morphogens (Choe et al., 2016), growth factors (Oliveira et al., 2013), neurotrophic factors (Vilar and Mira, 2016), and neurotransmitters (Berg et al., 2013). Internal signaling mechanisms that alter NSC fate decisions involve transcription factors and epigenetic modifications (Hsieh, 2012; Hsieh and Zhao, 2016; Semerci and Maletic-Savatic, 2016; Sun et al., 2011). For the purposes of this dissertation, only specific external and internal signalling pathways

involved with the regulation of NSC fate decisions through mitochondrial function will be discussed in detail.

1.2. Importance of Mitochondria in NSC Fate Decisions

1.2.1. Mitochondrial Metabolism Regulates Stem Cell Fate Decisions

Stem cells tend to rely on glycolysis and have globular mitochondria with poorly developed cristae structure, while differentiated cells tend to rely on oxidative phosphorylation (OXPHOS) and have elongated mitochondria with highly organized cristae structure (Chen et al., 2008; Cho et al., 2006; Chung et al., 2007; St. John et al., 2005; Khacho et al., 2016; Kondoh et al., 2007; Piccoli et al., 2005; Varum et al., 2011). As a result, stem cells generate less cellular ATP compared to their differentiated counterparts (Cho et al., 2006; Folmes et al., 2011; Prigione et al., 2010; Varum et al., 2011). Based on these observations, it was initially thought that stem cells had immature and dysfunctional mitochondria, which suggested that the metabolic shift from glycolysis to OXPHOS during differentiation was simply a consequence of specialized cells requiring more cellular energy than stem cells (Cho et al., 2006; Chung et al., 2010; Facucho-Oliveira et al., 2007; St. John et al., 2005; Prigione et al., 2010; Yanes et al., 2010). However, several studies have provided compelling evidence arguing that cellular metabolism plays an activate role in regulating cell fate decisions (Folmes et al., 2011; Khacho et al., 2016; Takubo et al., 2013). Inhibiting glycolysis or enhancing OXPHOS has been demonstrated to promote differentiation, while inhibiting OXPHOS or enhancing glycolysis has been demonstrated to promote pluripotency (Chen et al., 2008; Folmes et al., 2011; Khacho et al., 2016; Mandal et al., 2011; Panopoulos et al., 2012; Pereira et al., 2013; Varum et al., 2011; Yanes et al., 2010; Zhu et al., 2010).

Interestingly, cellular metabolism plays an ATP-independent role in regulating cell fate decisions. As previously mentioned, mitochondria in stem cells were once considered to be immature and defective due to their low bioenergetic output. However, it has been reported that stem cells can be forced into using OXPHOS, suggesting that these cells can generate ATP through the electron transport chain (ETC) (Folmes et al., 2011; Sánchez-Aragó et al., 2013; Zhang et al., 2011, 2012). Several OXPHOS suppression pathways have been identified in stem cells, including the upregulation of uncoupling protein 2 (UCP2), pyruvate dehydrogenase kinase (PDK), and the ATPase inhibitory factor 1 (IF1) (Sánchez-Aragó et al., 2013; Takubo et al., 2013; Zhang et al., 2011). Another example involves the downregulation of the mitochondrial pyruvate carrier (MPC) in order to physically prevent pyruvate from entering the mitochondria (Schell et al., 2017). Taken together, these findings suggest that stem cells need to actively maintain a glycolytic state in order to preserve their pluripotency (Khacho and Slack, 2017).

ATP-independent metabolic reprograming is achieved through the activity of specific metabolic intermediates and by-products known to affect cell fate decisions (Zhang et al., 2018). Reactive oxygen species (ROS), which were once considered to be a toxic by-product of OXPHOS, have been shown to act as signaling molecules involved with molecular pathways related to stem cell differentiation (Le Belle et al., 2011; Khacho et al., 2016; Lyublinskaya et al., 2015; Owusu-Ansah and Banerjee, 2009; Paul et al., 2014; Tormos et al., 2011). Metabolite intermediates such as α- ketoglutarate (α-KG), acetyl coenzyme A (acetyl-CoA), and nicotinamide adenine dinucleotide (NAD) can influence cell fate decisions through epigenetic modifications (Imai and Guarente, 2014; Kaelin et al., 2013; Ryall et al., 2015; Wellen et al., 2009). α-KG promotes DNA and histone demethylation (Kaelin et al., 2013), while acetyl-CoA and oxidized

NAD (NAD^+) facilitate histone acetylation and deacetylation, respectively (Imai and Guarente, 2014; Ryall et al., 2015; Wellen et al., 2009).

1.2.2. Mitochondrial Dynamics and Cellular Metabolism

Mitochondrial dynamics describe changes in mitochondrial morphology as a result of fusion and fission events. Mitochondrial fusion is achieved by mitofusin 1 (MFN1), mitofusin 2 (MFN2), and optic atrophy protein 1 (Opa1). MFN1 and MFN2 are involved with outer mitochondrial membrane (OMM) fusion, while Opa1 is involved with inner mitochondrial membrane (IMM) fusion and cristae remodelling (Chen et al., 2003; Cipolat et al., 2004; Meeusen et al., 2006; Song et al., 2009). Mitochondrial fission is mediated by dynamin-related protein 1 (Drp1) and other fission factors (Losón et al., 2013; Smirnova et al., 2001). Exposing cells to various stress conditions has been shown to cause changes in mitochondrial morphology (Benard and Rossignol, 2008; Gomes et al., 2011; Tondera et al., 2009). These changes are intended to promote cell survival by not only preventing cell-death by apoptosis (Cipolat et al., 2006; Frank et al., 2001; Germain et al., 2005; Montessuit et al., 2010), but by also ensuring sufficient ATP production (Cogliati et al., 2013; Gomes et al., 2011; Khacho et al., 2014; Patten et al., 2014; Rambold et al., 2011). Mitochondrial fusion enhances OXPHOS efficiency by promoting the formation of respiratory chain supercomplexes, while mitochondrial fission impairs OXPHOS efficiency by disrupting the formation of respiratory chain supercomplexes (Cogliati et al., 2013). These findings suggest a bidirectional relationship between mitochondrial dynamics and metabolism.

1.2.3. Mitochondrial Regulation of NSC Fate Decisions

Given the central role of mitochondria in regulating stem cell fate decisions, we wanted to gain a better understanding of the mitochondrial regulation of NSC fate decisions (Khacho et al., 2016). Characterization of NSCs, progenitor cells, and neurons in the embryonic mouse brain revealed that NSCs and neurons have elongated mitochondria, while progenitor cells have fragmented mitochondria. This finding suggested a potential role for mitochondrial dynamics in neuronal differentiation. Acute genetic modifications of mitochondrial morphology were found to alter NSC fate decisions, independently of ATP production and mitochondrial integrity. Enhancing mitochondrial fusion was found to promote NSC self-renewal, while enhancing mitochondrial fission was found to promote NSC differentiation. These observations were discovered to be the result of an NRF2 (nuclear factor erythroid 2–related factor 2) dependent retrograde gene expression pathway mediated by ROS signaling (Figure 2). According to this pathway, increased mitochondrial fragmentation promotes NSC differentiation by causing increased ROS production, which leads to the NRF2-dependent upregulation of the notch-dependent self-renewal inhibitor botch (Chi et al., 2012) and several pro-neuronal transcription factors. This study demonstrated that mitochondrial morphology itself could affect NSC fate decisions (Khacho et al., 2016).

A different team of researchers decided to study mitochondrial regulation of adult hippocampal neurogenesis (Beckervordersandforth et al., 2017). Mitochondria were demonstrated to undergo a characteristic change in both morphology and bioenergetics during NSC differentiation in the adult hippocampus. Adult NSCs have highly fragmented mitochondria and rely primarily on glycolysis. During neuronal differentiation, mitochondria become increasingly elongated and shift towards OXPHOS. Genetic and transgenic approaches to disrupt OXPHOS in NSCs revealed that the shift towards OXPHOS occurs as early as the transition from activated

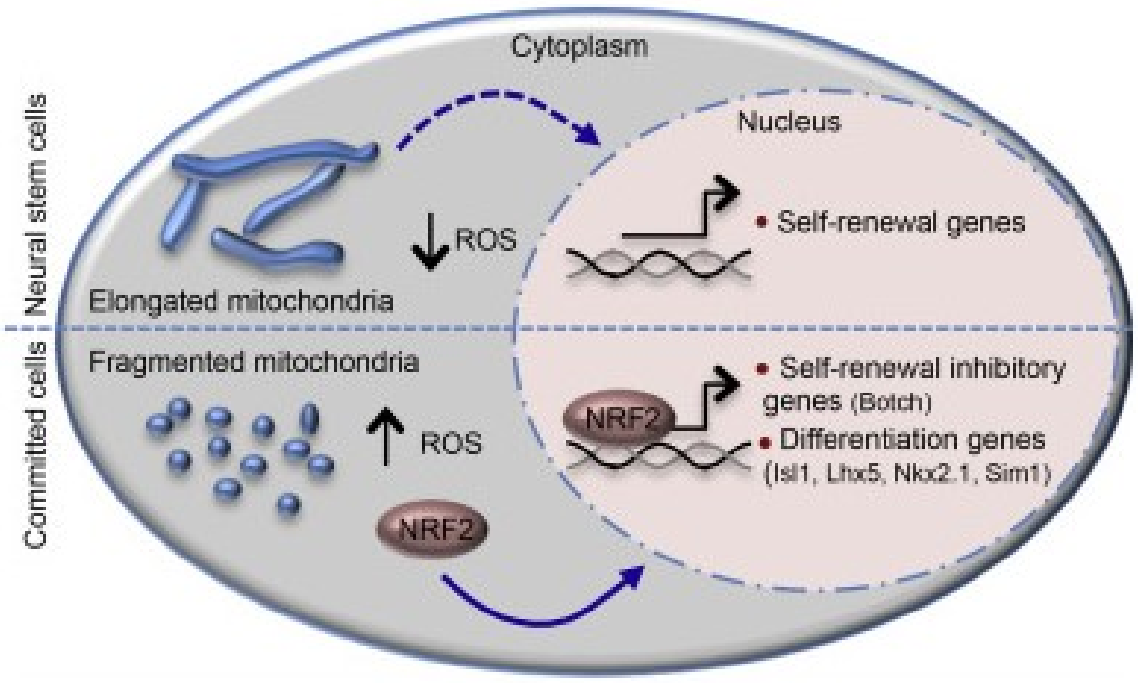

Figure 2. Mitochondrial dynamics is a regulator of NSC fate decisions. This diagram illustrates how mitochondrial dynamics can regulate neural stem cell (NSC) fate decisions through a nuclear factor erythroid 2–related factor 2 (NRF2)-dependent retrograde gene expression pathway mediated by reactive oxygen species (ROS) signalling. Mitochondrial fusion promotes NSC self-renewal by causing a reduction in ROS production. This results in NRF2 remaining inactive, which allows for the continued expression of genes related to self-renewal. Mitochondrial fission promotes NSC differentiation by causing an increase in ROS production. This results in the activation of NRF2, which can then upregulate the expression of genes that promote neuronal commitment and inhibit self-renewal. Lhx5, LIM (Lin11, Isl1, and Mec-3) homeobox 5; Nkx2.1, NK2 homeobox 1; Sim1, Single-minded homolog 1. Adapted from (Khacho et al., 2016).

NSCs to progenitor cells and is required for the proliferation and differentiation of progenitors. The conditional knockout (KO) of the mitochondrial transcription factor A (TFAM) in mice did not affect the NSC population in the adult SGZ, but did cause a decrease in the total number of T-box brain protein 2 (Tbr2)-positive progenitors and doublecortin (Dcx)-positive neuroblasts and immature neurons. The conditional KO of TFAM was also found to impair the dendritic maturation of neurons, demonstrating a role for mitochondria in regulating neuronal maturation. Interestingly, similar results were obtained when comparing adult hippocampal neurogenesis between young and aged mice (Beckervordersandforth et al., 2017). Lastly, the researchers demonstrated that piracetam, a pharmacological agent known to improve mitochondrial function (Costa et al., 2013; Keil et al., 2006; Leuner et al., 2010; Waegemans et al., 2002), could ameliorate many of the observed age-dependent defects in adult NSC differentiation and maturation, provided that these cells had intact ETC components. Compared to the study by Khacho et al. (2016), this study focused entirely on the importance of mitochondria in regulating NSC fate decisions and neuronal maturation in the adult brain (Beckervordersandforth et al., 2017).

Given the importance of mitochondria in regulating NSC fate decisions (Beckervordersandforth et al., 2017; Khacho et al., 2016), we sought to study the long-term impact of mitochondrial dysfunction on neuronal development (Khacho et al., 2017). This was achieved by using conditional apoptosis-inducing factor (AIF) KO mouse models to study the effects of mitochondrial dysfunction on neurogenesis during embryonic and adult stages of development (Cheung et al., 2006; Germain et al., 2013; Khacho et al., 2017; Klein et al., 2002; Vahsen et al., 2004). The loss of AIF during embryogenesis was found to cause decreased NSC self-renewal, increased proliferation of neural progenitor cells, and impaired neuronal commitment and survival. A prolonged loss of AIF in the dorsal telencephalon was found to cause cognitive defects due to a

complete loss of adult neurogenesis in the SGZ. These results suggested that mitochondrial dysfunction could lead to impaired neurogenesis by causing a decrease in NSC self-renewal coupled with a defect in neuronal commitment and survival (Khacho et al., 2017). The findings from this study could provide useful insights into understanding aging and neurodegenerative diseases; two conditions associated with mitochondrial dysfunction and defects in adult neurogenesis (Beal, 2005; Khacho et al., 2017; Seib and Martin-Villalba, 2014; Winner and Winkler, 2015).

1.3. Overview of PD

1.3.1. Pathology and Clinical Aspects of PD

Parkinson's disease (PD) is a neurodegenerative disorder defined by the progressive loss of dopaminergic neurons within the substantia nigra pars compacta (SNpc). The loss of these neurons causes dopamine deficiency in the striatum, resulting in impaired motor activity. Typical motor symptoms of PD include bradykinesia, rigidity, postural instability, and resting tremors (Kalia and Lang, 2015; Magrinelli et al., 2016). PD also affects other neuronal populations in the central and peripheral nervous systems (CNS and PNS), which leads to the development of non-motor symptoms such as hyposmia, sleep disturbances, constipation, mood changes, and cognitive decline (Braak and Braak, 2000; Braak et al., 2004; Giguère et al., 2018; Schapira et al., 2017; Winkler et al., 2011). Interestingly, these non-motor symptoms have been reported in patients to occur well before the onset of any motor deficits (Winkler et al., 2011). In the majority of PD cases, protein aggregates comprised primarily of alpha-synuclein can be found in the soma (Lewy bodies) and processes (Lewy neurites) of afflicted neurons (Kim et al., 2014; Shults, 2006; Spillantini et al., 1997). There is currently no cure for PD and current treatment options, such as

levodopa therapy and deep brain stimulation, focus on symptomatic relief (Anderson et al., 2017; Schapira et al., 2009).

1.3.2. Sporadic vs. Familial PD

PD can be broadly classified as being either sporadic or familial, depending on the nature of the disease. Sporadic PD accounts for ~90% of all cases of PD and is the result of a combination of genetic and environmental risk factors. Familial PD accounts for the remaining ~10% of PD cases and is caused by mutations in specific genes that follow Mendelian inheritance (Exner et al., 2012). Autosomal dominant PD has been reported as a result of gain-of-function mutations in genes encoding proteins like alpha-synuclein (Chartier-Harlin et al., 2004; Polymeropoulos et al., 1997; Singleton et al., 2003) or leucine-rich repeat kinase 2 (LRRK2) (Funayama et al., 2002; Paisán-Ruíz et al., 2004; Zimprich et al., 2004). Autosomal recessive PD has been reported as a result of loss-of-function mutations in genes encoding proteins like parkin (Kitada et al., 1998), phosphatase and tensin homolog (PTEN)-induced putative kinase 1 (PINK1) (Valente et al., 2004), Daisuke-Junko-1 (DJ-1) (Bonifati et al., 2003), or ATPase Type 13A2 (ATP13A2) (Ramirez et al., 2006). Interestingly, all of these proteins have been shown to be involved in one or more aspects of maintaining proper mitochondrial function (Exner et al., 2012).

1.3.3. PD is Associated with Mitochondrial Dysfunction

The underlying cause of neurodegeneration in PD remains unknown (Giguère et al., 2018). However, numerous studies have shown mitochondrial dysfunction to be associated with PD (Giguère et al., 2018; Park et al., 2018; Pickrell and Youle, 2015). Evidence supporting this relationship was first obtained from observations of human patients that had developed Parkinsonism (displayed motor symptoms typical of PD) and degeneration of dopaminergic

neurons in the SN after accidently taking 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) (Davis et al., 1979; Langston et al., 1983). MPTP is oxidized by monoamine oxidase B (MAO-B) into 1-methyl-4-phenylpyridinium (MPP^+), which is able to selectively enter dopaminergic neurons via the dopamine transporter (DAT) and inhibit the activity of complex I of the ETC (Javitch et al., 1985; Nicklas et al., 1985; Ramsay et al., 1986; Vyas et al., 1986). Signs of mitochondrial dysfunction were then found in patients with sporadic PD, such as complex I deficiency (Parker et al., 1989, 2008; Schapira et al., 1989, 1990) and increased mitochondrial DNA (mtDNA) mutations in dopaminergic neurons of the SN (Bender et al., 2006; Kraytsberg et al., 2006). The discovery of familial forms of PD, involving genes related to proper mitochondrial maintenance, has provided even more evidence supporting a link between mitochondrial dysfunction and PD (Exner et al., 2012). Whether mitochondrial dysfunction is causative or simply a symptom of PD is still under investigation (Giguère et al., 2018; Park et al., 2018; Pickrell and Youle, 2015).

1.4. Involvement of Parkin and PINK1 in PD

1.4.1. The Parkin and PINK1 Proteins

The first gene discovered to play a role in autosomal recessive PD was *PARK2*, which encodes the E3 ubiquitin (Ub) ligase parkin (Kitada et al., 1998; Matsumine et al., 1997). This 465 aa (amino acid) protein is composed of an ubiquitin-like (Ubl) domain, three really interesting new gene (RING) domains (RING0, RING1, and RING2), an in-between RING (IBR) domain, and a repressor of parkin (REP) region (Gladkova et al., 2018; Trempe and Fon, 2013; Trempe et al., 2013; Wauer and Komander, 2013). As an E3 ubiquitin enzyme, parkin is responsible for facilitating the transfer of Ub from E2 conjugating enzymes to other proteins (Hristova et al., 2009;

Shimura et al., 2000; Trempe and Fon, 2013). Parkin is found in the cytosol in an auto-inhibited state, which is thought to prevent auto-ubiquitination and its subsequent degradation (Gladkova et al., 2018; Kumar et al., 2015; Rakovic et al., 2013; Sauvé et al., 2018; Trempe and Fon, 2013; Wauer and Komander, 2013).

The second gene discovered to play a role in autosomal recessive PD was *PARK6*, which encodes the serine/threonine kinase PINK1 (Unoki and Nakamura, 2001; Valente et al., 2004). This 581 aa protein is composed of a mitochondria targeting sequence (MTS), an outer mitochondrial membrane localization signal (OMS), a transmembrane domain (TMD), and a kinase domain (Okatsu et al., 2015; Silvestri et al., 2005; Sim et al., 2012; Valente et al., 2004; Zhou et al., 2008). Through its kinase activity, PINK1 can activate parkin (Kane et al., 2014; Kazlauskaite et al., 2014; Kondapalli et al., 2012; Koyano et al., 2014; Shiba-Fukushima et al., 2012). Figure 3 provides an overview of the domain structures of human parkin and PINK1.

1.4.2. Parkin and PINK1 KO Animal Models

After discovering the involvement of parkin and PINK1 in familial PD, various animal models were developed in order to study the roles of these proteins in the cell (Pickrell and Youle, 2015). Parkin and PINK1 KO *Drosophila* models demonstrated reduced lifespans, male sterility, flight muscle degradation, loss of specific subsets of dopaminergic neurons, locomotor defects, and aberrant changes in mitochondrial integrity (Clark et al., 2006; Greene et al., 2003; Park et al., 2006; Pesah et al., 2004; Whitworth et al., 2005; Yang et al., 2006). The first parkin and PINK1 KO mouse models failed to recapitulate key aspects of PD pathology, such as a significant loss of dopaminergic neurons in the SN and gross motor impairments (Akundi et al., 2011; von Coelln et al., 2004; Goldberg et al., 2003; Itier et al., 2003; Kitada et al., 2007; Perez and Palmiter, 2005).

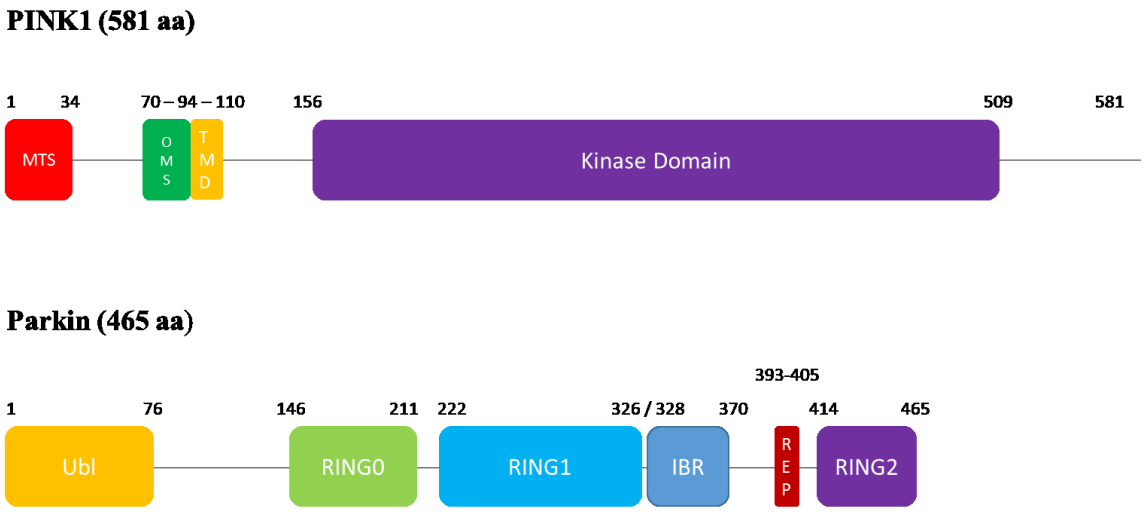

Figure 3: Domain structure diagrams of human PINK1 and parkin. PINK1 (581 aa) is a serine/threonine kinase and parkin (465 aa) is an E3 ubiquitin ligase (Gladkova et al., 2018; Sekine and Youle, 2018). Numbers refer to specific aa locations. aa, amino acid(s); IBR, in-between RING; MTS, mitochondria targeting sequence; OMS, outer mitochondrial membrane localization signal; PINK1, phosphatase and tensin homolog (PTEN)-induced putative kinase 1; REP, repressor of parkin; RING, really interesting new gene; RING0, really interesting new gene 0; RING1, really interesting new gene 1; RING2, really interesting new gene 2; TMD, transmembrane domain; Ubl, ubiquitin-like.

Even aged parkin, PINK1, and DJ-1 triple KO mice showed no signs of neurodegeneration in the SN (Kitada et al., 2009). However, signs of altered dopamine metabolism in the striatum and mitochondrial dysfunction have been observed in certain parkin and PINK1 KO mouse models (Gandhi et al., 2009; Gautier et al., 2008; Goldberg et al., 2003; Itier et al., 2003; Palacino et al., 2004; Periquet et al., 2005). PD mouse models based on the inducible KO of PD associated genes (Lee et al., 2017; Shin et al., 2011) or combining germline KO models with an additional cellular stressor (Pickrell et al., 2015), have reported dopaminergic neurodegeneration.

1.4.3. Parkin and PINK1 Maintain Mitochondrial Integrity

Complementation studies in parkin and PINK1 KO *Drosophila* suggested that these proteins were involved in a common pathway responsible for maintaining proper mitochondrial function and morphology, with PINK1 acting upstream of parkin (Clark et al., 2006; Greene et al., 2003; Park et al., 2006; Yang et al., 2006). A common pathway involving both proteins was soon discovered by a study demonstrating that treatment of cultured cells with carbonyl cyanide m-chlorophenyl hydrazone (CCCP) resulted in parkin translocation to depolarized mitochondria, which leads to the selective removal of these impaired mitochondria through a process of autophagy known as mitophagy (Narendra et al., 2008). PINK1 was later found to be necessary for the activation and translocation of parkin in this process (Matsuda et al., 2010; Narendra et al., 2010; Vives-Bauza et al., 2010). Parkin and PINK1 have also been discovered to be involved with several other pathways related to processes such as mitochondrial quality control (Braschi et al., 2010; McLelland et al., 2014; Soubannier et al., 2012; Vincow et al., 2013; Xu et al., 2011), dynamics (Park et al., 2018), transport (Liu et al., 2012b; Wang et al., 2011c), biogenesis (Lee et al., 2017; Shin et al., 2011), and mRNA processing on the OMM (Gehrke et al., 2015).

1.5. Cellular Functions of PINK1 and Parkin

1.5.1. PINK1 and Parkin Regulation of Mitochondrial Quality Control

PINK1 and parkin regulate mitochondrial quality control through mitochondria-associated degradation (MAD), mitochondria-derived vesicles (MDVs), and mitophagy (Truban et al., 2017). During MAD, polyubiquitination of OMM proteins selectively targets these proteins for degradation by the proteasome (Karbowski and Youle, 2011; Taylor and Rutter, 2011). Examples of PINK1/parkin-dependent MAD include the degradation of mitofusins during PINK1/parkin-dependent mitophagy (Tanaka et al., 2010; Ziviani et al., 2010) and the mitochondrial Rho (Miro) GTPase during cessation of mitochondrial transportation (Liu et al., 2012b; Wang et al., 2011c). PINK1 and parkin can also facilitate the formation of MDVs that can transport oxidized proteins towards lysosomes for degradation (McLelland et al., 2014; Soubannier et al., 2012). Interestingly, PINK1 and parkin were also discovered to inhibit a novel MDV-mediated mitochondrial antigen presentation (MitAP) pathway, which raises questions regarding the roles of PINK1 and parkin in regulating innate immunity (Matheoud et al., 2016). Lastly, PINK1/parkin-dependent mitophagy involves the removal of entire defective mitochondria through autophagy (Matsuda et al., 2010; Narendra et al., 2008, 2010; Vives-Bauza et al., 2010). Given the interest in PINK1/parkin-dependent mitophagy with respect to PD pathology (Pickrell and Youle, 2015), this process will be described below in greater detail. Figure 4 provides an overview of PINK1/parkin-dependent regulation of mitochondrial quality control.

Under basal conditions, PINK1 is rapidly degraded shortly after being translated (Matsuda et al., 2010; Narendra et al., 2010). This process begins with PINK1 entering the mitochondria through the translocase of the outer membrane (TOM) and translocase of the inner membrane 23

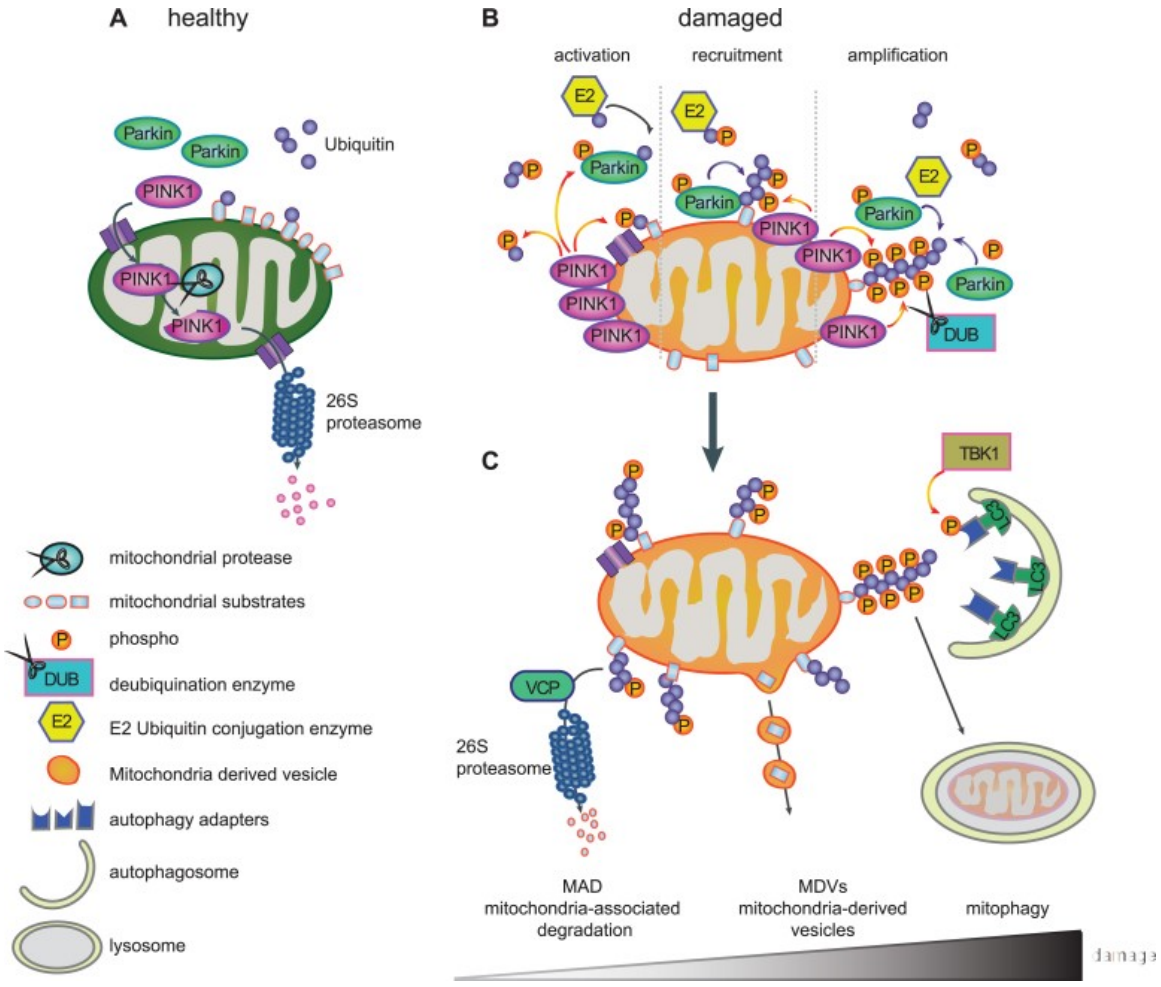

Figure 4. Overview of PINK1/parkin-dependent regulation of mitochondrial quality control. (A) Under basal conditions, PINK1 is continuously recruited to the mitochondria to undergo degradation, while parkin remains in an auto-inhibited state in the cytosol. (B) Damage to the mitochondria causes PINK1 to stabilize on the outer mitochondrial membrane (OMM). PINK1 recruits and fully activates parkin by phosphorylating parkin and ubiquitin (Ub). By working in concert, PINK1 and parkin are able to facilitate the rapid polyubiquitination of OMM proteins. (C) Depending on the severity of the mitochondrial damage, mitochondrial homeostasis can be maintained through several different PINK1/parkin-dependent pathways. Individual polyubiquitinated OMM proteins can be selectively degraded through mitochondria-associated degradation (MAD). Localized mitochondrial damage could lead to the degradation of a collection of oxidized proteins through mitochondria-derived vesicles (MDVs). Lastly, OMM polyubiquitin chains can recruit autophagosome components to facilitate the degradation of the entire mitochondria through the process of mitophagy. TBK1, TANK [TRAF (tumor necrosis factor receptor-associated factor) family member-associated NF-κB (nuclear factor κ-light-chain-enhancer of activated B cells) activator]-binding kinase 1. Adapted from (Truban et al., 2017).

(TIM23) complexes (Beilina et al., 2005; Lin and Kang, 2008; Okatsu et al., 2015; Silvestri et al., 2005; Takatori et al., 2008; Valente et al., 2004). During mitochondrial entry, PINK1 is cleaved by the mitochondrial processing peptidase (MPP) in the matrix and the PINK1/PGAM5 (phosphoglycerate mutase family member 5)-associated rhomboid-like protease (PARL) in the IMM (previously named as the presenilin-associated rhomboid-like protease) (Greene et al., 2012; Jin et al., 2010; Meissner et al., 2011; Spinazzi and De Strooper, 2016). The resulting 52 kDa PINK1 fragment is then retro-translocated from the mitochondria back into the cytosol to be ubiquitinated and degraded by the proteasome (Lin and Kang, 2008; Takatori et al., 2008; Yamano and Youle, 2013).

During a cell stress event that causes mitochondrial dysfunction, translocation through the TIM23 complex stops, due to a loss of mitochondrial membrane potential ($\Delta\Psi m$) (Bertolin et al., 2013; Jin et al., 2010). When PINK1 is unable to translocate through the TIM23 complex, PINK1 is retained in the OMM and forms a complex with another PINK1 protein and components of the TOM complex (Bertolin et al., 2013; Lazarou et al., 2012; Okatsu et al., 2012, 2013, 2015). This leads to the activation of PINK1 through auto-phosphorylation (Okatsu et al., 2012, 2013; Rasool et al., 2018). PINK1 is then able to phosphorylate parkin and OMM protein Ub chains at their Ser65 amino acid residues. The phosphorylation of Ub and parkin is necessary for the recruitment of parkin to impaired mitochondria and to fully activate its ligase activity (Kane et al., 2014; Kazlauskaite et al., 2014; Kondapalli et al., 2012; Koyano et al., 2014). Parkin is able to create polyubiquitinated chains that can be phosphorylated by PINK1, thereby creating a feed-forward loop that rapidly causes polyubiquitination of OMM proteins (Ordureau et al., 2014; Sarraf et al., 2013). These polyubiquitinated chains act as receptors for autophagy components that will be

recruited to allow for the selective removal of damaged mitochondria (Heo et al., 2015; Lazarou et al., 2015).

Following the characterization of PINK1/parkin-dependent mitophagy, there was debate on the involvement of mitophagy in regulating mitochondrial homeostasis in neurons (Cummins and Götz, 2018; Mouton-Liger et al., 2017). PINK1/parkin-dependent mitophagy was first characterized in artificial cell-culture models that used immortalized cells that primarily rely on glycolysis for ATP production, protonophores to induce significant mitochondrial depolarization, and the overexpression or even exogenous expression of parkin and PINK1. It was also possible to induce a complete loss of the mitochondrial network under these conditions (Matsuda et al., 2010; Narendra et al., 2008, 2010; Vives-Bauza et al., 2010). Neurons rely on OXPHOS to meet their high bioenergetic demands, meaning that a process that could potentially lead to a loss of the entire mitochondrial network seems impractical and unlikely to occur in neurons (Almeida et al., 2001, 2004; Bolaños, 2016). Results from initial studies on mitophagy in neurons were mixed since there were studies that failed to observe PINK1/parkin-dependent mitophagy in neurons (Van Laar et al., 2011; Rakovic et al., 2013; Sterky et al., 2011), while other studies had reported this process to occur in neurons under specific culture conditions (Ashrafi et al., 2014; Cai et al., 2012). A study using a mitochondrial-targeted form of the pH-sensitive fluorescent probe Keima (mt-Keima) was able to demonstrate basal mitochondrial turnover by PINK1/parkin-dependent mitophagy in neurons (Bingol et al., 2014). Transgenic reporter systems based on mt-Keima or mito-QC have since been used to provide *in vivo* evidence of mitophagy in various cell types, including neurons (Cornelissen et al., 2018; Mcwilliams et al., 2018; McWilliams et al., 2016; Sun et al., 2017; Williams et al., 2017).

Questions regarding the relevance of PINK1 and parkin in mediating mitophagy in neurons have been raised (Cummins and Götz, 2018; Mouton-Liger et al., 2017; von Stockum et al., 2018). In a recent study by McWilliams et al. (2018), basal mitophagy in PINK1 KO mice was reported to be unaffected in almost all analyzed tissues, including those from the brain. PINK1 and/or parkin independent pathways of mitophagy have been discovered and could potentially explain these findings (Cummins and Götz, 2018; Mcwilliams et al., 2018; von Stockum et al., 2018). E3 Ub ligases other than parkin, such as glycoprotein 78 (Gp78), mitochondrial E3 ubiquitin protein ligase 1 (MUL1), and seven in absentia homolog 1 (SIAH1), have been found to facilitate ubiquitination of OMM proteins and promote mitophagy (Fu et al., 2013; Li et al., 2015; Szargel et al., 2016). Autophagy receptors in the OMM such as NIX (NIP3-like protein X), BNIP3 (BCL2/adenovirus E1B 19 kDa protein-interacting protein 3), FUNDC1 (FUN14 domain containing 1), and FKBP8 (FK506-binding protein 8) can recruit autophagosome components to the mitochondria, independent of OMM protein ubiquitination by an E3 Ub ligase (Bhujabal et al., 2017; Georgakopoulos et al., 2017; Liu et al., 2012a; Novak et al., 2010; Rikka et al., 2011). The presence of cardiolipin in the OMM has been shown to directly recruit autophagosome components to mitochondria (Chu et al., 2013). Iron and glyceraldehyde 3-phosphate dehydrogenase (GAPDH)-mediated pathways of mitophagy have also been suggested (Allen et al., 2013; Hwang et al., 2015; Yogalingam et al., 2013). Therefore, future studies will need to investigate the potential context-dependent roles of PINK1 and parkin in regulating neuronal mitophagy (Mcwilliams et al., 2018).

1.5.2. PINK1 and Parkin Regulation of Mitochondrial Dynamics

Aberrant changes in mitochondrial morphology have been observed in various parkin and PINK1 KO models (Lim et al., 2012). Initial studies in parkin and PINK1 KO *Drosophila* models revealed an increase in mitochondrial fusion (Clark et al., 2006; Greene et al., 2003; Park et al., 2006; Yang et al., 2006). Mitochondrial morphology was found to be relatively unchanged in corresponding early mouse models, except for an increase in mitochondrial elongation in the striatum of PINK1 KO mice (Gautier et al., 2008; Palacino et al., 2004). Interestingly, a number of parkin and PINK1 KO mammalian cell models have shown that a loss of either protein results in an increase in mitochondrial fragmentation (Cui et al., 2010; Dagda et al., 2009; Exner et al., 2007; Lutz et al., 2009). In the study by Lutz et al. (2009), loss of parkin or PINK1 in cultured cells from *Drosophila* was found to cause a transient increase in mitochondrial fragmentation, followed by a sustained increase in mitochondrial fusion. These observations suggest that various factors including the choice of species, time of analysis, presence of compensatory mechanisms, and the method of altering parkin/PINK1 gene expression can impact the study of parkin/PINK1-dependent regulation of mitochondrial dynamics (Lim et al., 2012; Lutz et al., 2009). Several mechanistic studies have provided evidence supporting a role for parkin and PINK1 interacting with components of the mitochondrial fission or fusion machinery in order to promote either mitochondrial fragmentation or elongation (Buhlman et al., 2014; Deng et al., 2008; Gegg et al., 2010; Lutz et al., 2009; Poole et al., 2008, 2010; Pryde et al., 2016; Wang et al., 2011b; Yang et al., 2008; Ziviani et al., 2010). It has been hypothesized that the regulation of mitochondrial dynamics by parkin and PINK1 is important in facilitating mitochondrial quality control, through the efficient separation of defective mitochondria from their healthy counterparts (Buhlman et al., 2014; Pryde et al., 2016; Wang et al., 2011b; Ziviani et al., 2010).

1.5.3. PINK1 and Parkin Regulation of Mitochondrial Transportation

Mitochondria can be transported along microtubules within the cell by utilizing a protein complex consisting of Miro, a trafficking kinesin protein (TRAK)/Milton motor-adaptor protein, and a kinesin or dynein motor protein (Brickley and Stephenson, 2011; Glater et al., 2006; Guo et al., 2005; Stowers et al., 2002; van Spronsen et al., 2013). The binding of Ca^{2+} to Miro inhibits mitochondrial motility, which suggests a possible role for mitochondrial transportation in maintaining Ca^{2+} homeostasis by promoting the localization of mitochondria to areas with high Ca^{2+} concentrations (MacAskill et al., 2009; Saotome et al., 2008; Wang and Schwarz, 2009). PINK1 and parkin have both been implicated in regulating mitochondrial transportation by inhibiting mitochondrial motility through the degradation of Miro (Liu et al., 2012b; Wang et al., 2011c; Weihofen et al., 2009). A loss of PINK1 or parkin can prove detrimental to maintaining mitochondrial homeostasis, since the rapid cessation of mitochondrial transportation is necessary to ensure the efficient removal of defective mitochondria by mitophagy (Hsieh et al., 2016; Liu et al., 2012b).

1.5.4. PINK1 and Parkin Regulation of Mitochondrial Biogenesis

Mitochondrial biogenesis requires the precise coordinated transcription of mitochondrial related genes from both the nuclear and mitochondrial genomes (Jornayvaz and Shulman, 2010). Peroxisome proliferator-activated receptor (PPAR)-γ coactivator-1α (PGC-1α) is essential for proper mitochondrial biogenesis since PGC-1α activates nuclear respiratory factors 1 and 2, which are responsible for the activation of TFAM (Andersson and Scarpulla, 2001; Lin et al., 2002; Meirhaeghe et al., 2003; Puigserver et al., 1998; Virbasius and Scarpulla, 1994; Wu et al., 1999). Parkin was discovered to be involved with regulating mitochondrial biogenesis through the

ubiquitination of the parkin interacting substrate [(PARIS; also known as zinc finger protein 746 (ZNF746)], a transcriptional repressor of PGC-1α that undergoes degradation following ubiquitination (Shin et al., 2011). PINK1 was later revealed to be involved with this pathway by phosphorylating PARIS in order to facilitate parkin-dependent ubiquitination (Lee et al., 2017). The conditional KO of parkin or PINK1 in mice was noted to result in PARIS-dependent dopaminergic neurodegeneration (Lee et al., 2017; Shin et al., 2011).

1.5.5. PINK1 and Parkin Regulation of mRNA Translation at the Mitochondria

The translation of mRNA encoding mitochondrial proteins was originally thought to occur in the cytosol and mitochondrial matrix (Schmidt et al., 2010; Wenz et al., 2015). However, numerous studies have provided evidence demonstrating mitochondrial protein mRNA translation at the OMM (Lesnik et al., 2015). PINK1 and parkin have been implicated in regulating the translation of specific nuclear encoded mRNAs at the OMM, by facilitating the removal of several translational repressors (Gehrke et al., 2015).

1.6. Adult Neurogenesis and PD

Results from animal studies have suggested that the non-motor symptoms of PD could be partially explained by an impairment in adult neurogenesis. Non-motor symptoms of PD such as hyposmia, depression, and cognitive decline have been observed in animal models of dysfunctional adult neurogenesis (Regensburger et al., 2014). Toxin-based PD mouse models using 6-hydroxydopamine (6-OHDA) or MPTP have reported decreased proliferation in the SVZ (Baker et al., 2004; Höglinger et al., 2004; O'Keeffe et al., 2009; Sui et al., 2012; Winner et al., 2006). Interestingly, mice treated with 6-OHDA or MPTP have also been reported to have an

increased number of dopaminergic glomerular neurons in the OBs (Winner et al., 2006; Yamada et al., 2004). Decreased neuronal survival in the SGZ and OBs have been observed in transgenic mouse models overexpressing α-synuclein (Kohl et al., 2012; Marxreiter et al., 2009, 2013a; May et al., 2012; Nuber et al., 2008; Winner et al., 2004, 2008) or LRRK2 (Winner et al., 2011). A loss of PINK1 in mice was shown to impair neuronal differentiation, possibly due to aberrant changes in mitochondrial function (Agnihotri et al., 2017). There is also some evidence in post-mortem studies on human PD patients that supports the notion that PD can lead to impaired adult neurogenesis in the SVZ and SGZ (Höglinger et al., 2004; O'Keeffe et al., 2009). Studying the impact of PD on adult neurogenesis could prove useful in the development of novel therapeutic strategies involving NSCs.

1.7. Rationale for the Hypothesis and Objectives of the Present Study

Based on the literature, mitochondrial function can be considered as a major player in NSC fate decisions and PD pathology (Khacho et al., 2019). This means it would be interesting to study the impact of PD related mitochondrial dysfunction on NSC fate decisions. This type of study would involve the characterization of adult neurogenesis in a suitable model of PD, analysis of mitochondrial dynamics and function in specific cell populations of interest, identification of the molecular pathways behind any observed changes, and a demonstration of a rescue of adult neurogenesis through the restoration of mitochondrial integrity. Given the magnitude of such a project, this thesis will focus on the preliminary characterization of adult neurogenesis in animal models of PD. **I hypothesize that a loss of PINK1 or parkin in mice causes impaired adult neurogenesis.** In order to test this hypothesis, there are two objectives:

Aim I: Characterization of SVZ and SGZ adult neurogenesis in PINK1 KO mice.

Aim II: Characterization of SVZ and SGZ adult neurogenesis in parkin KO mice.

Assessment of adult neurogenesis will be done using immunohistochemistry to compare differences in specific SGZ and SVZ cell populations between PD mouse models and wild-type (WT) controls.

MATERIALS AND METHODS

2.1. Mice

Three transgenic mouse lines were used for this study: PINK1$^{-/-}$, parkin$^{-/-}$, and SOD2$^{+/-}$ (Fournier et al., 2009; Haque et al., 2012; Itier et al., 2003; Kitada et al., 2007; Lebovitz et al., 1996). All mice were examined at 6 months of age. All mice were maintained on a pure C57BL6 genetic background. Littermate controls were used whenever possible (otherwise outsourced C57BL6 mice were used as controls). All experimental protocols involving mice were approved by the University of Ottawa Animal Care Committee.

The PINK1 KO mice were provided by Dr. David Park's lab (University of Ottawa Brain and Mind Research Institute). PINK1 KO was achieved by using a phosphoglycerate kinase-neomycin (PGK-Neo) cassette to cause a deletion of exons 4-7 of PINK1. This disrupts the coding of the PINK1 kinase domain and also introduces a premature stop codon (Kitada et al., 2007). The original PINK1 KO mice had either 129SV/C57BL6 or 129SV/BALBc/C57BL6 mixed genetic backgrounds (Kitada et al., 2007), until being backcrossed onto a pure C57BL6 genetic background (Haque et al., 2012).

The parkin KO mice were provided by Dr. Michael Schlossmacher's lab (Ottawa Hospital Research Institute). Parkin KO was achieved by using a Neo-resistance cassette to cause a deletion of exon 3 of parkin (Itier et al., 2003). The original parkin KO mice had either a pure 129SV or mixed 129SV/C57BL6 genetic backgrounds (Itier et al., 2003), until being backcrossed onto a pure C57BL6 genetic background (Fournier et al., 2009).

The SOD2 (superoxide dismutase 2) hemizygous KO mice were also provided by Dr. Michael Schlossmacher's lab (Ottawa Hospital Research Institute). A complete SOD2 KO is lethal. The SOD2 KO was achieved through the deletion of exons 1 and 2 of SOD2 by using a

human hypoxanthine phosphoribosyltransferase (HPRT) gene construct coupled with a PGK promoter element. The original SOD2 hemizygous mice were developed on a pure 129SV genetic background (Lebovitz et al., 1996). These mice had been backcrossed onto a pure C57BL6 genetic background and can be purchased through The Jackson Laboratory (JAX #002973). Compound parkin KO and SOD2 hemizygous KO mice are referred to as SODPAR mice.

Genotyping was done by PCR using REDExtract-N-Amp™ PCR ReadyMix™ (MilliporeSigma) and following the guidelines from the manufacturer. Primer sequences for genotyping (Table 1) were based off of published protocols (Fournier et al., 2009; Kessova and Cederbaum, 2007; Madeo et al., 2014). Genotyping for SOD2 was done using primers specific for the PGK-HPRT construct, with primers for SOD1 (superoxide dismutase 1) being used as an internal control (Kessova and Cederbaum, 2007). While this would had have technically made it impossible to genotype between a hemizygous or homozygous SOD2 KO, any homozygous SOD2 KO mice would not be viable (Lebovitz et al., 1996).

2.2. Tissue Fixation, Cryoprotection, and Cryosectioning

Adult mice that were sacrificed first received an intraperitoneal injection of 0.65mg/g of a diluted Euthanyl solution (65mg/mL pentobarbital sodium USP), before undergoing a transcardial perfusion with 1X phosphate-buffered saline (PBS) followed by a 4% paraformaldehyde (PFA) solution in 1X PBS (pH=7.4). Perfused brains were stored for 24 hours in 4% PFA to ensure complete fixation. Afterwards, the brains were transferred to a 20% sucrose and 0.01% sodium azide solution in 1X PBS for long-term storage. Fixed brains were partitioned into left and right hemispheres, with one hemisphere returned to storage and the other frozen in isopentane (Thermo Fisher Scientific) at a temperature between -35^{o}C and -40^{o}C. Frozen hemispheres were mounted

Target	Genotyping Primer Sequences
PINK1	WT-Forward: 5'-AGAGGATGCTAGTCCCTGTGAAGGG-3' (PINK1-F) WT-Reverse: 5'-ACACTCAGTCCTTGGGCAATGCTA-3' (PINK1-X) KO-Reverse: 5'-ACCAAAGAAGGGAGCCGGTTG-3' (NeoA)
Parkin	WT-Forward: 5'-TGCTCTGGGGTTCGTC-3' KO-Forward: 5'-TTGTTTTGCCAAGTTCTAAT-3' Common-Reverse: 5'-TCCACTGGCAGAGTAAATGT-3'
SOD1 and PGK-HPRT	SOD1-Forward (IMR0878): 5'-TGAACCAGTTGTGTTGTCAGG-3' SOD1-Reverse (IMR0888): 5'-TCCATCACTGGTCACTAGCC-3' PGK-HPRT-Forward (IMR0781): 5'-TGTTCTCCTCTTCCTCATCTCC-3' PGK-HPRT-Reverse (IMR0782): 5'-ACCCTTTCCAAATCCTCAGC-3'

Table 1. PCR primer sequences used for genotyping. Forward and reverse genotyping primers for specific regions of PINK1 (Madeo et al., 2014), parkin (Fournier et al., 2009), SOD1, and PGK-HPRT (Kessova and Cederbaum, 2007).

with Tissue-Tek® O.C.T. Compound (Sakura Finetek) and sectioned using a cryostat (Leica CM 1850) to obtain 30µm thick serial coronal sections of the SVZ and SGZ. Nine wells of serial coronal sections were obtained for each brain structure of interest. Tissue sections were stored at 4°C in a solution of 0.01% sodium azide in 1X PBS.

2.3. Immunohistochemistry

Free-floating tissue sections from an entire well were removed from storage, washed 4x in 1X PBS for 5 minutes each time, and then incubated with primary antibodies (Table 2) diluted in a solution of 0.1% Triton X-100 (MilliporeSigma) and 0.1% Tween-20 (Thermo Fisher Scientific) in 1X PBS overnight. Afterwards, tissue sections were washed 4x in 1X PBS for 5 minutes each time and then incubated with appropriate secondary antibodies conjugated with fluorescent markers (Table 2) and 4',6-diamidino-2-phenylindole (DAPI) diluted in another solution of 0.1% Triton X-100 and 0.1% Tween-20 in 1X PBS for 2 hours (protected from light). DAPI (MilliporeSigma) was diluted at 1:1000 from an original 1µg/mL solution. Tissue sections were then washed 4x in 1X PBS for 5 minutes each time before being mounted onto Fisherbrand™ Superfrost™ Plus Microscope Slides (Thermo Fisher Scientific) with Thermo Scientific™ Shandon™ Immu-Mount™ (Thermo Fisher Scientific). Different antibodies were sometimes used for the same targets depending on antibody combinations and/or commercial availability.

2.4. EdU Labelling

In vivo labelling of proliferating cells was achieved by giving an intraperitoneal injection of 10µL/g of a 5 mg/mL EdU (5-ethynyl-2'-deoxyuridine) solution (Clickbase, BCK647-IV-IM-M) to the adult mice, 2.5 hours prior to being sacrificed. Tissue sections from these mice were

Antibody	Host Species	Source	Dilution
Primary Antibodies			
Dcx	Goat	Santa Cruz Biotechnology (sc-8066)	1:500
	Rabbit	Cell Signaling Technology (4604S)	1:500
GFAP	Mouse	MilliporeSigma (MAB3402)	1:1000 (SGZ) 1:10 000 (SVZ)
Ki67	Rabbit	Cell Marque (SP6)	1:500 (SGZ)
Nestin	Goat	R&D Systems (AF2736)	1:1000
Sox2	Goat	Neuromics (GT15098)	1:500
	Rabbit	Abcam (ab97959) MilliporeSigma (AB5603)	1:500 1:500
Secondary Antibodies			
Anti-goat Cy3	Donkey	Jackson ImmunoResearch (705-165-147)	1:1000
Anti-mouse Alexa Fluor 647	Donkey	Jackson ImmunoResearch (715-605-151)	1:1000
Anti-rabbit Alexa Fluor 488	Donkey	Jackson ImmunoResearch (711-545-152)	1:1000

Table 2. Primary and secondary antibodies used for immunohistochemistry. Unless otherwise stated, listed antibody dilutions were used for both SVZ and SGZ tissue sections.

processed according to the manufacturer's instructions (Clickbase, BCK647-IV-IM-M). EdU label processing was done on tissue samples after being washed 4x in 1X PHS for 5 minutes each time, following incubation with primary and secondary antibodies for immunohistochemistry.

2.5 Microscopy, Cell Quantification, and Statistical Analysis

Mounted tissue sections were imaged by a Zeiss LSM800 AxioObserver Z1 confocal microscope using ZenPro (ZEISS) image acquisition software or a GE DeltaVision Elite (with microinjector) epifluorescence microscope using SoftWoRx (GE) image acquisition software. The entire structure of the SVZ or SGZ was imaged using an objective of 20X. Image processing and cell quantification was done using Fiji (ImageJ) software. Total SVZ or SGZ cell counts were extrapolated from cell counts performed on imaged sections. SVZ cell counts were based off of analyzing 2-3 SVZ sections per animal. SGZ cell counts were based off of analyzing 7-11 SGZ sections per animal (depending on the total number of serial sections obtained from each animal). Cell counts are represented as mean ± SD. Comparisons between WT and PINK1 groups were done using unpaired, two-tailed Student's t-tests. Comparisons between WT, parkin KO, and SODPAR mice were done using a one-way ANOVA followed by a post-hoc Tukey's test.

RESULTS

3.1. Role of PINK1 in SVZ Adult Neurogenesis

We decided to first examine how the absence of PINK1 affects adult neurogenesis in the SVZ. Immunohistochemical staining of SVZ sections from adult WT and PINK1 KO mice was done using markers for Sox2, nestin, and GFAP (Figure 5). By using this combination of markers, it was possible to identify specific populations of neural precursor cells. For instance, $Sox2^{+}/Nestin^{+}/GFAP^{+}$ cells would represent activated NSCs (Type B1 cells), while $Sox2^{+}/Nestin^{+}/GFAP^{-}$ cells would represent transit amplifying progenitors (Type C cells) (Codega et al., 2014; Doetsch et al., 1997; Graham et al., 2003; Imayoshi et al., 2011; Kazanis et al., 2010; Lagace et al., 2007; Lendahl et al., 1990). With respect to the overall number of neural precursor cells ($Sox2^{+}$ cells) in the SVZ, no significant difference was found between the WT and PINK1 KO groups (Figure 5B and Table S1). However, the PINK1 KO mice were found to have significantly fewer (~22.77% fewer) activated Type B1 cells than the WT mice (Figure 5B and Table S1). Potential causes for this finding include a defect in NSC self-renewal, decreased cellular proliferation, or increased cell death. However, there was no significant difference in the total number of nestin expressing neural precursor cells ($Sox2^{+}/Nestin^{+}$ cells) or Type C cells in the SVZ between the WT and PINK1 KO mice (Figure 5B and Table S1). Given that there was no difference between both experimental groups with respect to the total number of nestin expressing neural precursor cells in the SVZ, there should have been an increase in the total number of Type C cells for the PINK1 KO mice to compensate for the decrease in the total number of activated Type B1 cells. It is possible that there was indeed a potential difference in either the total number of neural precursor cells, the subpopulation of neural precursor cells expressing nestin, or Type C cells, which was obscured by high statistical variance.

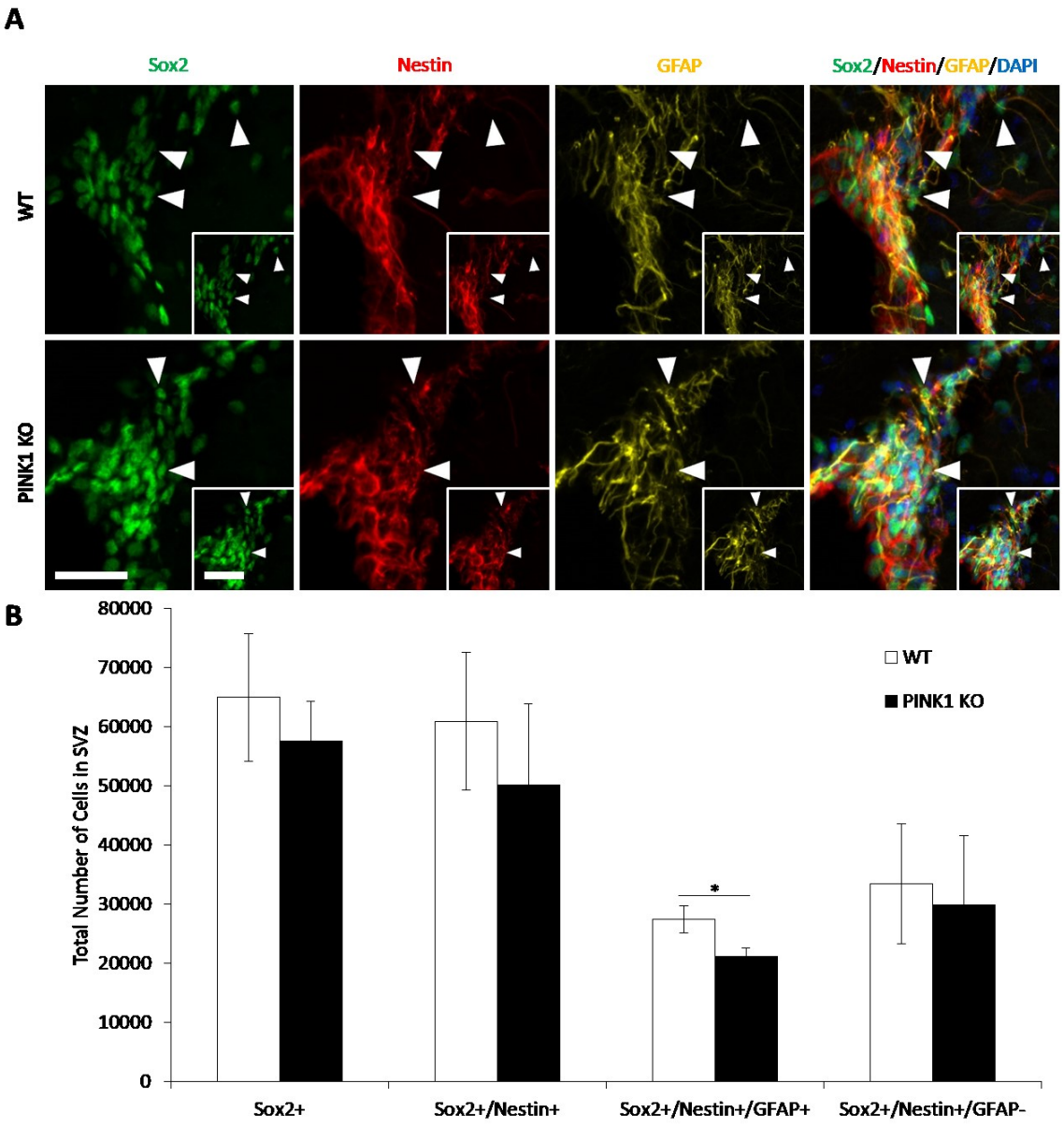

Figure 5: Loss of PINK1 causes a decrease in the total number of activated Type B1 cells in the adult SVZ. (A) Representative confocal images of the SVZ from 6 month old WT and PINK1 KO mice stained against Sox2, Nestin, GFAP, and DAPI. (B) Quantification of the $Sox2^+$, $Sox2^+/Nestin^+$, $Sox2^+/Nestin^+/GFAP^+$, and $Sox2^+/Nestin^+/GFAP^-$ cell populations in the SVZ. Arrowheads label activated Type B1 cells ($Sox2^+/Nestin^+/GFAP^+/DAPI^+$). Insets provide a zoomed-in view of the images. Quantitative data is represented as mean ± SD (n=3 per genotype, *$p<0.05$, Student's t-test). Scale bars = 50 μm.

In order to investigate the decline in the total number of activated Type B1 cells for the PINK1 KO mice (Figure 5), we decided to examine cellular proliferation in the SVZ (Figure 6). Prior to being sacrificed, the mice used for this study were given a single pulse of EdU, so that it would be possible to later assess cellular proliferation by counting the total number of EdU^+ cells (Chehrehasa et al., 2009). No significant difference was found between the WT and PINK1 KO mice with respect to the total number of EdU^+ cells in the SVZ (Figure 6B and Table S2). This result suggests that the decline in the total number of activated Type B1 cells for the PINK1 KO mice was not caused by a decrease in cellular proliferation.

While there were other possibilities that could have been explored with respect to the decreased number of activated Type B1 cells in the SVZ of the PINK1 KO mice (ie: self-renewal defect or increased cell death), we decided to shift our attention to studying the impact of a loss of PINK1 downstream of the neural precursor cell population in the SVZ. By using a marker for Dcx, a protein primarily expressed in neuroblasts (Type A cells) and newborn neurons (Koizumi et al., 2006; Lagace et al., 2007; Ming and Song, 2011), it was possible to compare the total number of Type A cells in the SVZ between WT and PINK1 KO mice (Figure 6). No significant difference was found between the WT and PINK1 KO mice with respect to the total number of Dcx^+ cells in the SVZ (Figure 6C and Table S2). This would imply that the loss of PINK1 (at least up until 6 months of age) does not affect the normal progression of neural precursor cells into becoming Type A cells in the SVZ.

Our analysis of neurogenesis in the adult SVZ revealed a decrease in the total number of activated Type B1 cells for the PINK1 KO mice (Figure 5). This decline was also found to not be the result of a defect in cellular proliferation (Figure 6). The loss of PINK1 was also found to not affect the total number of Type A cells in the SVZ (Figure 6). It is possible that the defect in the

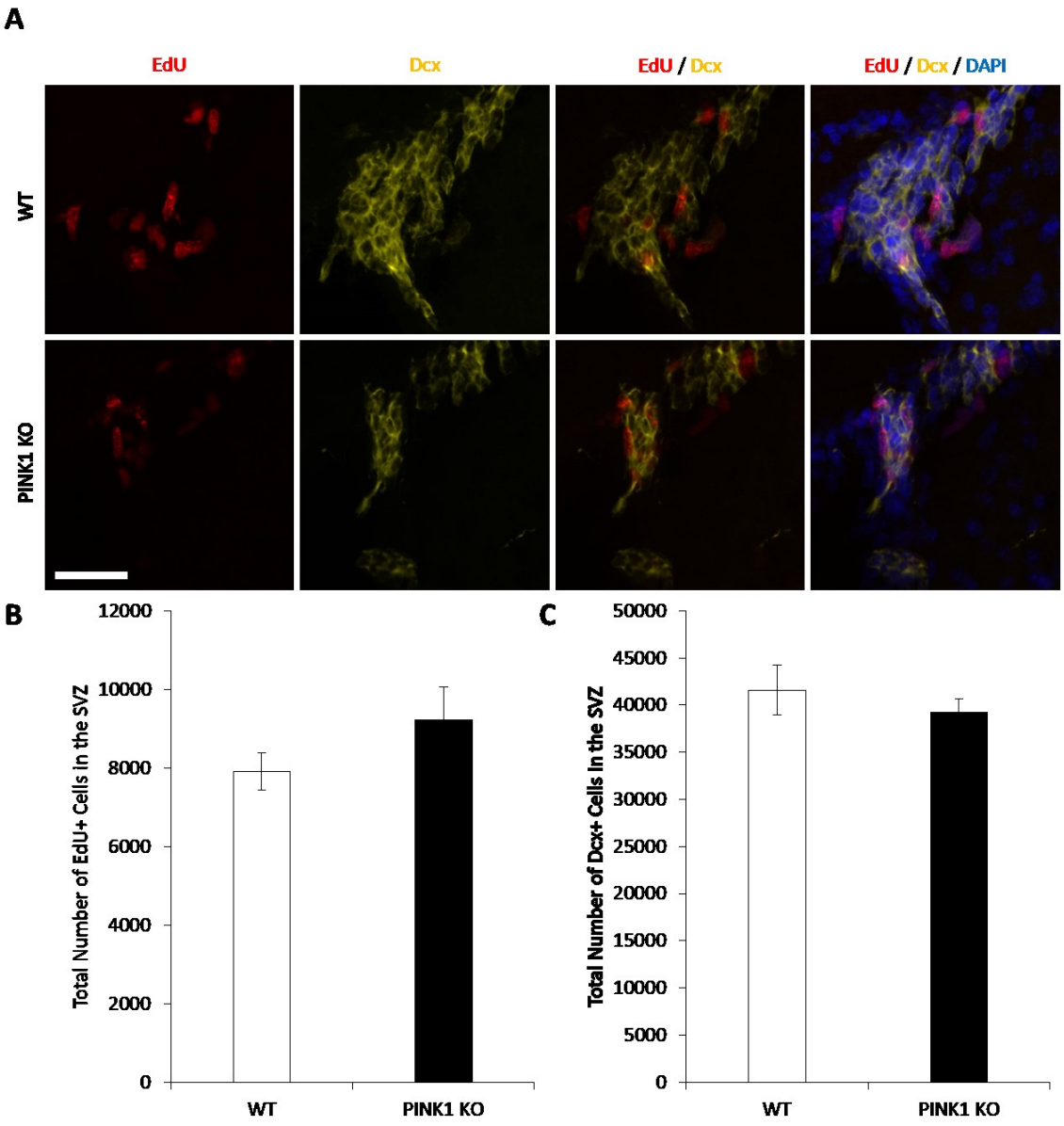

Figure 6: Loss of PINK1 does not affect cellular proliferation or the total number of Type A cells in the adult SVZ. (A) Representative confocal images of the SVZ from 6 month old WT and PINK1 KO mice stained against EdU, Dcx, and DAPI. (B) Quantification of the total number of EdU^+ cells in the SVZ. (C) Quantification of the total number of Dcx^+ cells in the SVZ. Quantitative data is represented as mean ± SD (n=3 per genotype, Student's t-test). Scale bar = 50 μm.

activated Type B1 cells was relatively recent, which would explain why there was no apparent change in the total number of Type A cells between both experimental groups.

3.2. Role of PINK1 in SGZ Adult Neurogenesis

After investigating the impact of a loss of PINK1 on adult neurogenesis in the SVZ, we decided to examine its impact on adult neurogenesis in the SGZ. The absence of PINK1 could have different effects on adult neurogenesis in the SVZ and SGZ because of niche specific differences (Bond et al., 2015; Ming and Song, 2011). Similar to our analysis in the SVZ, individual neural precursor cell populations in the SGZ were identified using markers for Sox2, nestin, and GFAP (Figure 7). In the SGZ, GFAP is expressed in NSCs (Type I cells), Sox2 is expressed in Type I cells and intermediate progenitor cells (Type II cells), and nestin is expressed in Type I cells and the Type IIa subpopulation of intermediate progenitors (Kronenberg et al., 2003; Lagace et al., 2007; Lugert et al., 2010, 2012; Seri et al., 2004; Shin et al., 2015; Suh et al., 2007). No significant differences were found between the WT and PINK1 KO mice with respect to the total number of neural precursor cells ($Sox2^+$ cells), nestin expressing neural precursor cells ($Sox2^+/Nestin^+$ cells), Type I cells ($Sox2^+/Nestin^+/GFAP^+$ cells), or Type IIa cells ($Sox2^+/Nestin^+/GFAP^-$ cells) in the SGZ (Figure 7B and Table S3). Based on these results, PINK1 is not necessary for the maintenance of the neural precursor cell population in the adult SGZ.

To determine if the absence of PINK1 causes aberrant cellular proliferation in the SGZ, we performed immunohistochemical staining of SGZ sections for Ki67 (Figure 8), a well-known marker of cellular proliferation (Scholzen and Gerdes, 2000). The PINK1 KO mice were found to have significantly fewer (~25.09% fewer) $Ki67^+$ cells in the SGZ than the WT mice (Figure 8B

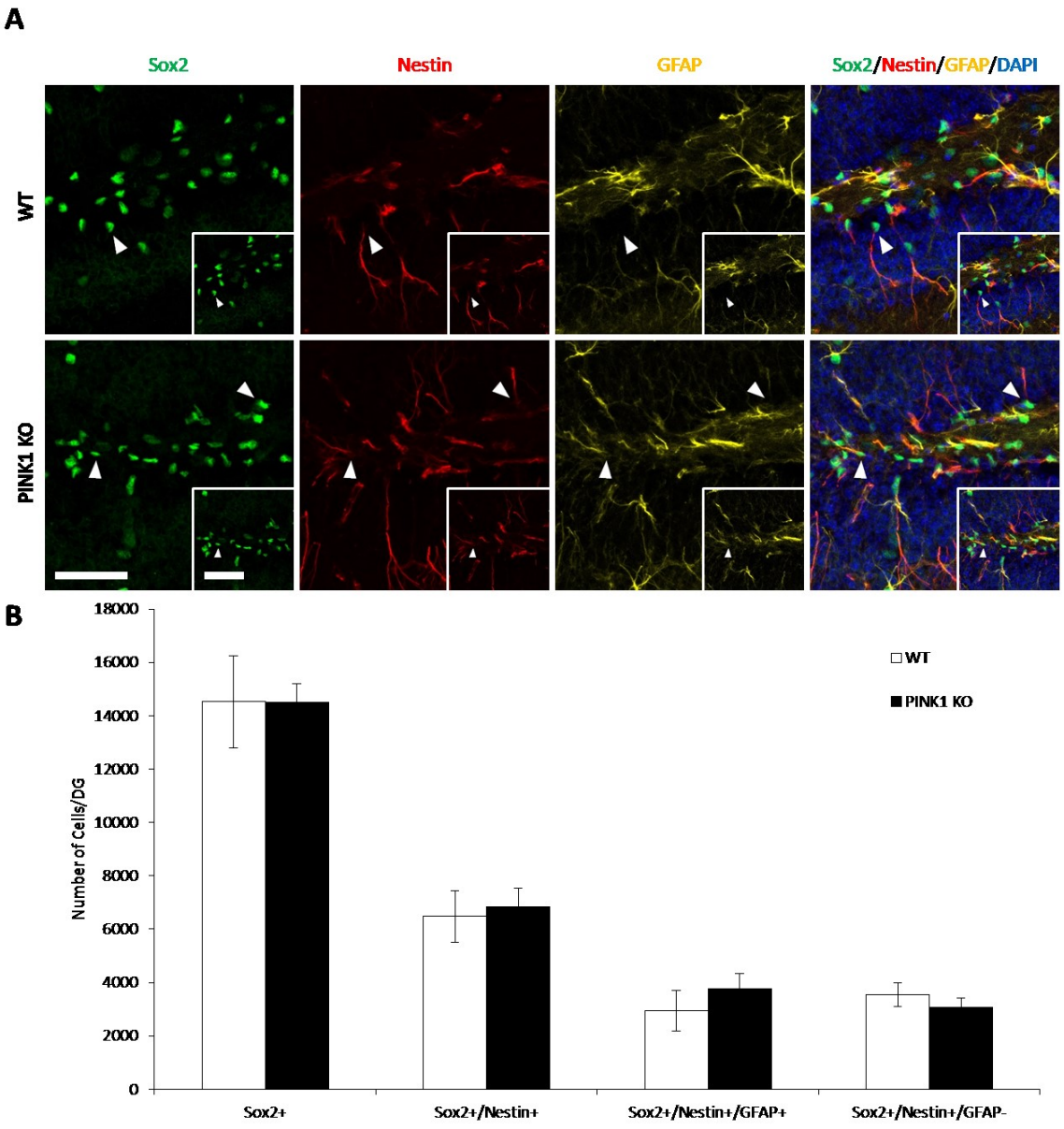

Figure 7: Loss of PINK1 does not affect the total number of neural precursor cells in the adult SGZ. (A) Representative confocal images of the SGZ from 6 month old WT and PINK1 KO mice stained against Sox2, Nestin, GFAP, and DAPI. (B) Quantification of the $Sox2^+$, $Sox2^+/Nestin^+$, $Sox2^+/Nestin^+/GFAP^+$, and $Sox2^+/Nestin^+/GFAP^-$ cell populations in the SGZ. Arrowheads label Type I cells ($Sox2^+/Nestin^+/GFAP^+/DAPI^+$). Insets provide a zoomed-in view of the images. Quantitative data is represented as mean ± SD (n=4 per genotype, Student's t-test). Scale bars = 50 μm.

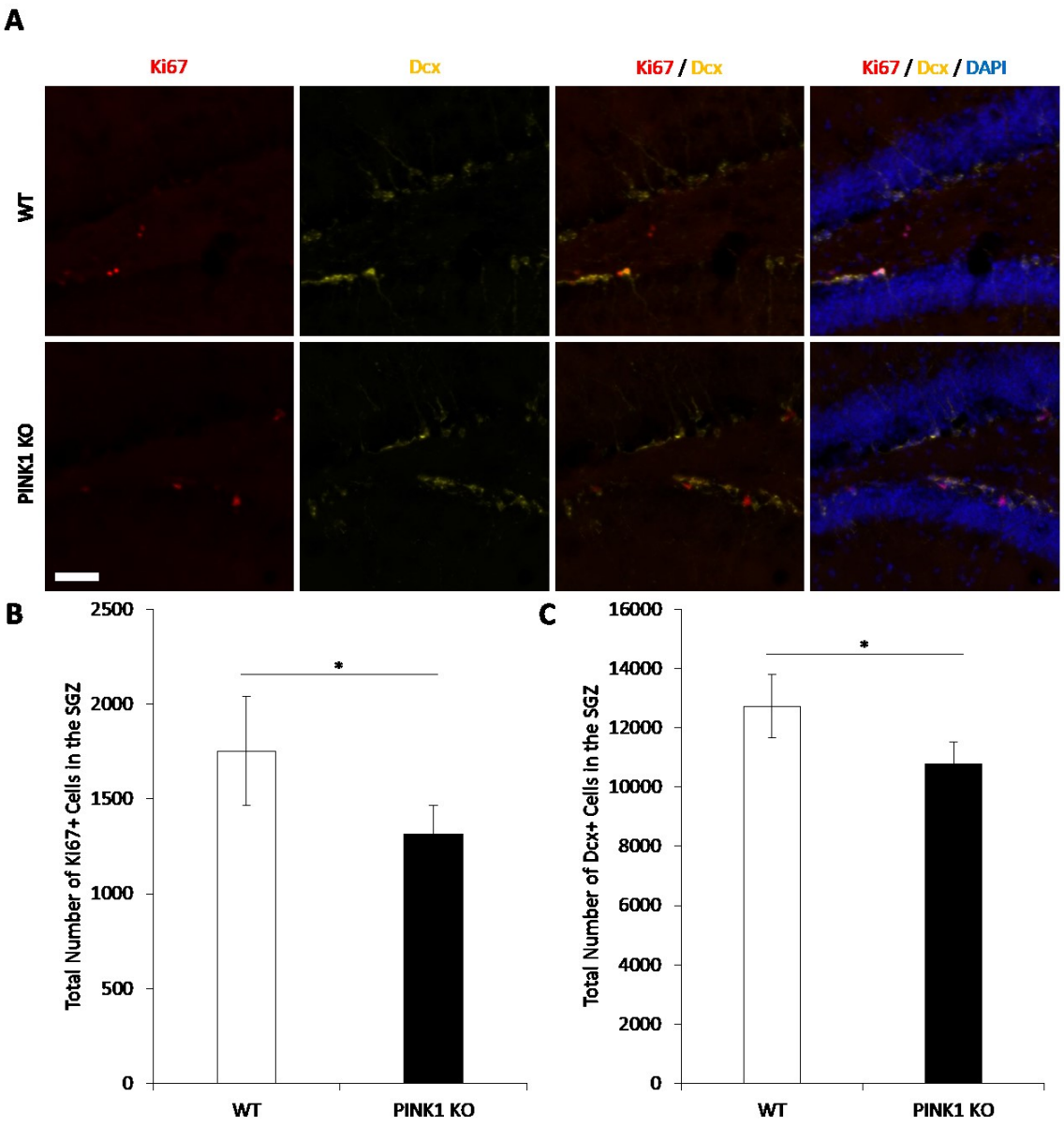

Figure 8: Loss of PINK1 causes a decrease in the total number of proliferating cells and newborn neurons in the adult SGZ. (A) Representative fluorescent images of the SGZ from 6 month old WT and PINK1 KO mice stained against Ki67, Dcx, and DAPI. (B) Quantification of the total number of $Ki67^+$ cells in the SGZ. (C) Quantification of the total number of Dcx^+ cells in the SGZ. Quantitative data is represented as mean ± SD (n=4 per genotype, *$p<0.05$, Student's t-test). Scale bar = 50 μm.

and Table S4). This suggests that PINK1 is required for proper cellular proliferation in the adult SGZ.

We decided to next investigate how the absence of PINK1 affects cells derived from neural precursor cells. This involved staining SGZ sections for Dcx (Figure 8), which is expressed in Type IIb cells, neuroblasts (Type III cells), and newborn neurons in the SGZ (Kronenberg et al., 2003; Lagace et al., 2007; Nicola et al., 2015). The PINK1 KO mice were found to have significantly fewer (~15.28% fewer) Dcx^+ cells, compared to the WT mice (Figure 8C and Table S4). The decrease in the total number of Dcx^+ cells in the SGZ for the PINK1 KO mice would indicate a decline in the total number of newborn neurons being produced, which would ultimately cause a decrease in the total number of mature neurons being produced by the SGZ over time. It is possible that this decline in the total number of newborn neurons could be caused in part by the previously mentioned deficit in cellular proliferation in the SGZ.

3.3. Role of Parkin in SVZ Adult Neurogenesis

For the second half of this investigation, we focused on studying the role of parkin in adult neurogenesis. While PINK1 and parkin have been shown to work together in several molecular pathways related to mitochondrial quality control, these two proteins have also been demonstrated in other molecular pathways to operate independently of each other (Exner et al., 2012; McWilliams and Muqit, 2017; Mouton-Liger et al., 2017; Park et al., 2018; Truban et al., 2017). This means that the absence of PINK1 or parkin could have different effects on adult neurogenesis. In order to study the role of parkin in adult neurogenesis, WT mice were compared to parkin KO and SODPAR (parkin KO with a hemizygous SOD2 KO) mice. SOD2 is an antioxidant enzyme responsible for reducing mitochondrial ROS in the cell (Lebovitz et al., 1996; Weisiger and

Fridovich, 1973; Zelko et al., 2002). By combing a loss of parkin with a reduction in SOD2, the SODPAR mice could potentially show more pronounced changes in adult neurogenesis than the parkin KO mice, which are known to display only minimal signs of PD pathology (Itier et al., 2003; Periquet et al., 2005). It was not possible to create homozygous SOD2 KO mice since the complete loss of SOD2 is lethal (Lebovitz et al., 1996).

The first question we asked when studying the role of parkin in adult neurogenesis, was how a loss of parkin would affect the neural precursor cell population in the SVZ. As with our analysis of the PINK1 KO mice (Figure 5), SVZ sections from WT, parkin KO, and SODPAR mice were stained with markers for Sox2, nestin, and GFAP (Figure 9) in order to identify specific populations of neural precursor cells. No significant differences were found between the WT, parkin KO, and SODPAR mice with respect to the total number of neural precursor cells ($Sox2^+$ cells), nestin expressing neural precursor cells ($Sox2^+/Nestin^+$ cells), activated Type B1 cells ($Sox2^+/Nestin^+/GFAP^+$ cells), or Type C cells ($Sox2^+/Nestin^+/GFAP^-$ cells) in the SVZ (Figure 9B and Table S5). These results suggest that parkin is not necessary for the maintenance of the neural precursor cell population in the adult SVZ.

The possibility of an absence of parkin affecting cellular proliferation in the adult SVZ was then examined by using EdU to quantify the number of proliferating cells in the SVZ (Figure 10). There were no significant differences between the WT, parkin KO, and SODPAR mice with respect to the total number of EdU^+ cells in the SVZ (Figure 10B and Table S6), suggesting that parkin is not required for cellular proliferation in the adult SVZ.

The impact of a loss of parkin on cells derived from the neural precursor cell population in the adult SVZ was then studied by using Dcx as a marker for Type A cells (Figure 10). There were no significant differences between the WT, parkin KO, and SODPAR mice with respect to the

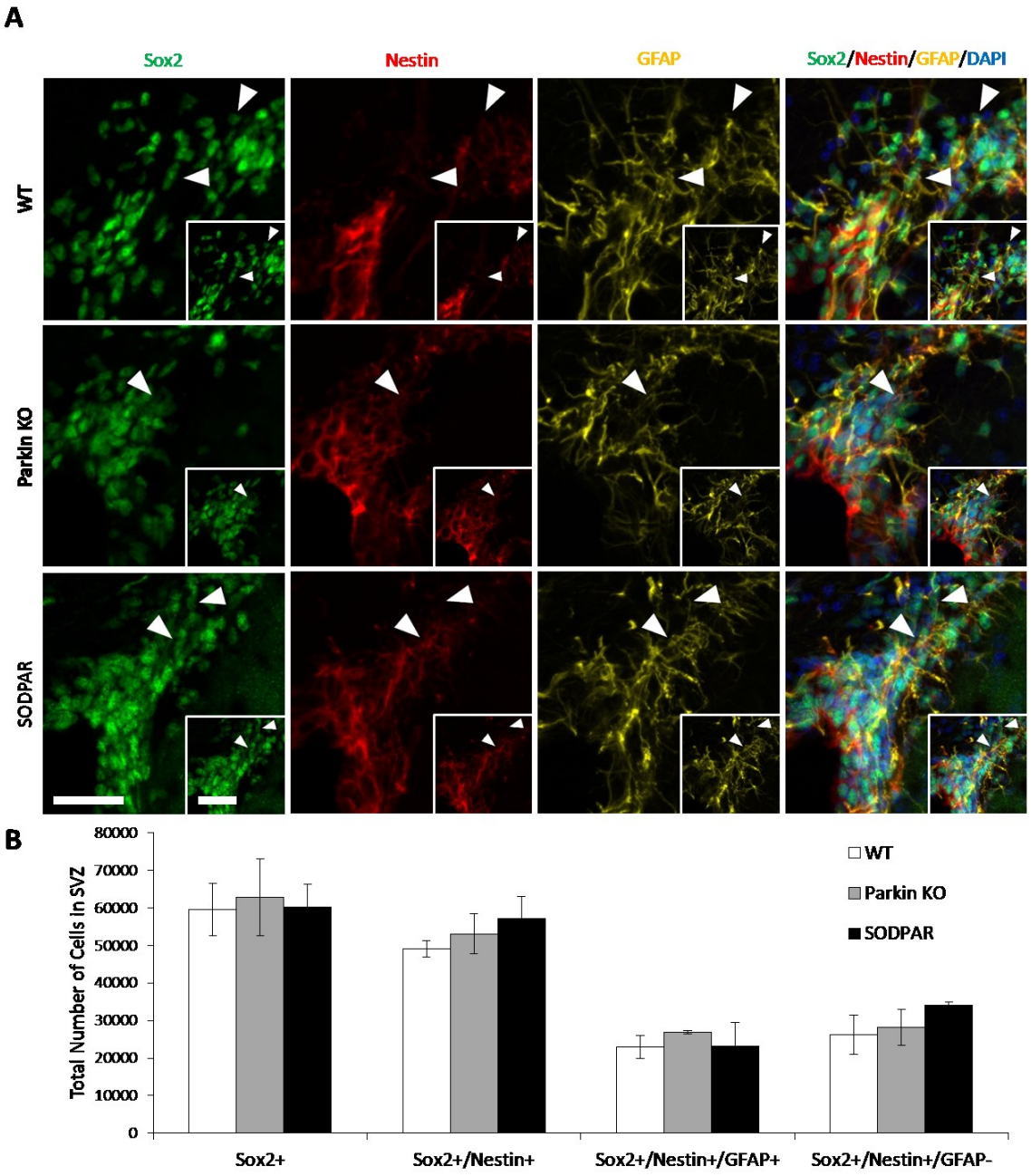

Figure 9: Loss of parkin does not affect the total number of neural precursor cells in the adult SVZ. (A) Representative confocal images of the SVZ from 6 month old WT, parkin KO, and SODPAR mice stained against Sox2, Nestin, GFAP, and DAPI. (B) Quantification of the $Sox2^{+}$, $Sox2^{+}/Nestin^{+}$, $Sox2^{+}/Nestin^{+}/GFAP^{+}$, and $Sox2^{+}/Nestin^{+}/GFAP^{-}$ cell populations in the SVZ. Arrowheads label activated Type B1 cells ($Sox2^{+}/Nestin^{+}/GFAP^{+}/DAPI^{+}$). Insets provide a zoomed-in view of the images. Quantitative data is represented as mean ± SD (n=3 per genotype, one-way ANOVA and Tukey's). Scale bars = 50 μm.

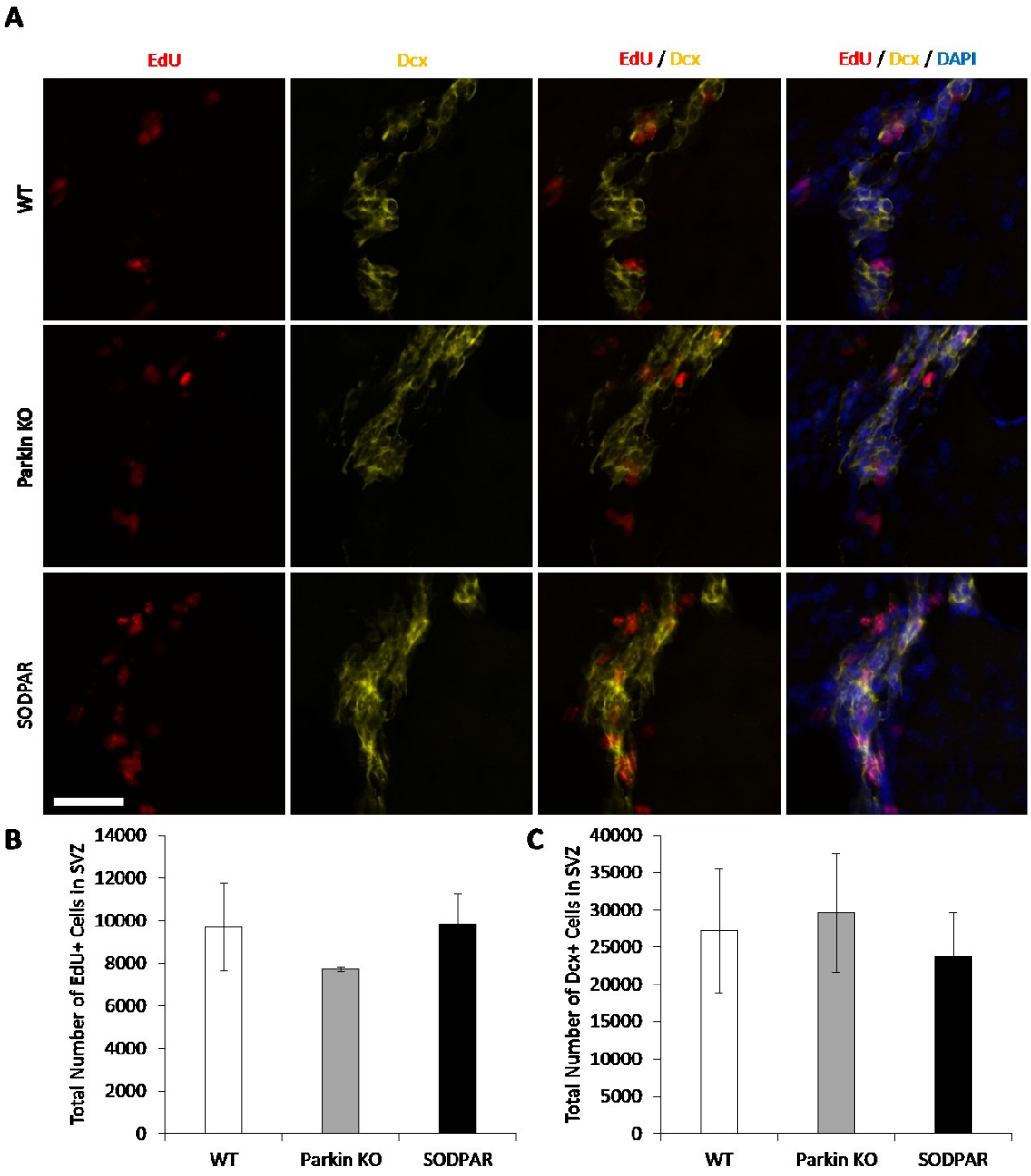

Figure 10: Loss of parkin does not affect cell proliferation or the total number of Type A cells in the adult SVZ. (A) Representative confocal images of the SVZ from 6 month old WT, parkin KO, and SODPAR mice stained against EdU, Dcx, and DAPI. (B) Quantification of the total number of EdU^+ cells in the SVZ. (C) Quantification of the total number of Dcx^+ cells in the SVZ. Quantitative data is represented as mean ± SD (n=3 per genotype, one-way ANOVA and Tukey's test). Scale bar = 50 μm.

total number of Dcx^+ (Type A) cells in the SVZ (Figure 10B and Table S6). This would imply that parkin is not necessary for the production of newborn neurons from the adult SVZ.

3.4. Role of Parkin in SGZ Adult Neurogenesis

Given the existence of niche specific differences between the SVZ and SGZ (Bond et al., 2015; Ming and Song, 2011), we decided to also investigate the role of parkin in SGZ adult neurogenesis. An analysis of the neural precursor cell population using markers for Sox2, nestin, and GFAP (Figure 11) revealed no significant differences between the WT, parkin KO, and SODPAR mice with respect to the total number of neural precursor cells (Sox2+ cells), nestin expressing neural precursor cells ($Sox2^+/Nestin^+$ cells), Type I cells ($Sox2^+/Nestin^+/GFAP^+$ cells), or Type IIa cells ($Sox2^+/Nestin^+/GFAP^-$ cells) in the SGZ (Figure 11B and Table S7). The impact of an absence of parkin on cellular proliferation and cells derived from the neural precursor cell population was examined by staining for Ki67 and Dcx (Figure 12). There was also no difference in the total number of $Ki67^+$ cells in the SGZ between the WT, parkin KO, and SODPAR mice (Figure 12B and Table S8), suggesting that parkin is not necessary for cellular proliferation in the adult SGZ. However, the parkin KO and SODPAR mice were found to have significantly fewer Dcx^+ cells in the SGZ (~19.25% fewer for the parkin KO mice and ~23.17% fewer for the SODPAR mice) compared to the WT control mice (Figure 12C and Table S8). The decrease in the total number of Dcx^+ cells in the SGZ would imply a decrease in the total number of newborn neurons being produced. There was no significant difference between the parkin KO and SODPAR mice with respect to the total number of Dcx^+ cells in the SGZ (Figure 12C and Table S8).

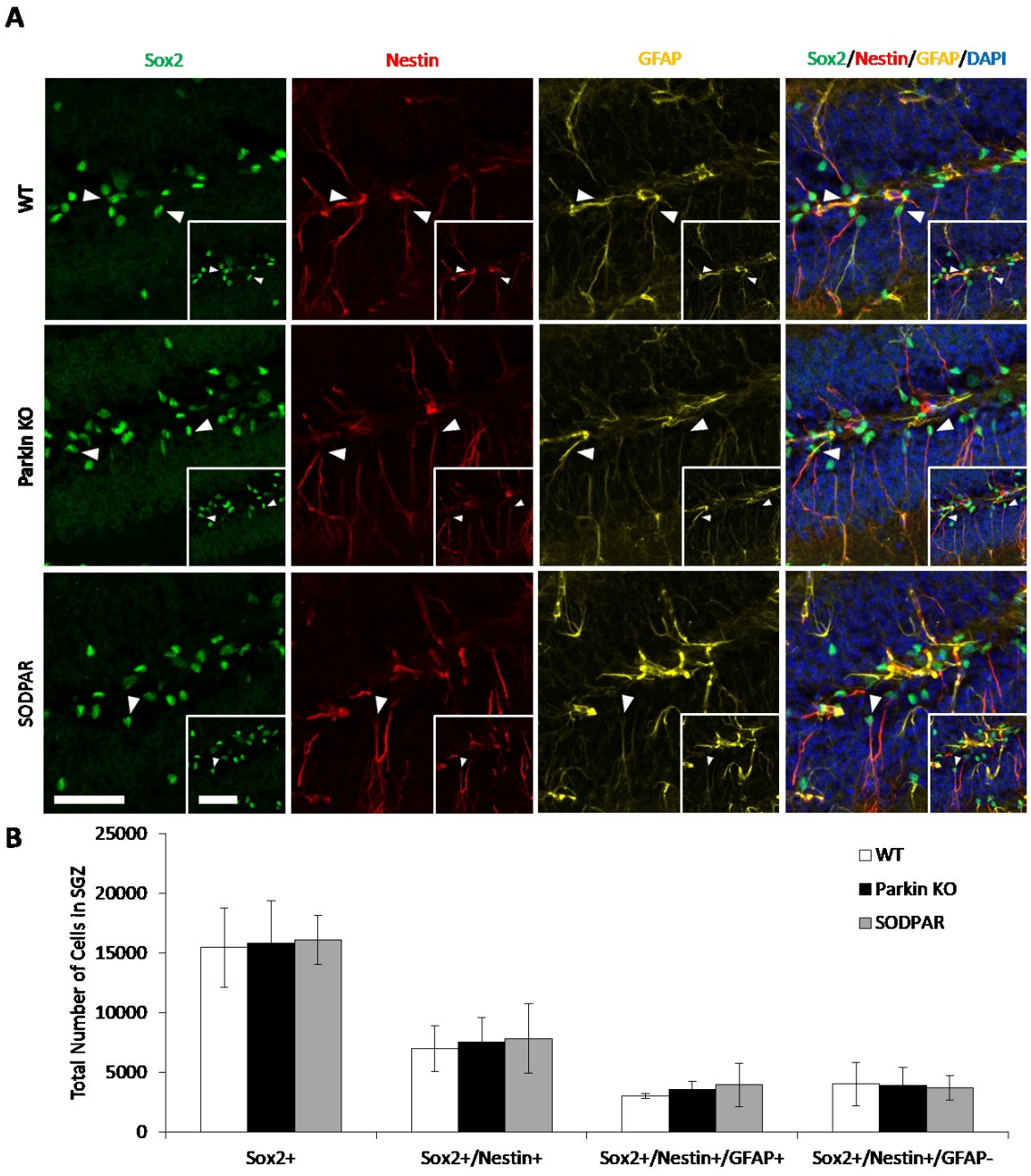

Figure 11: Loss of parkin does not affect the total number of neural precursor cells in the adult SGZ. (A) Representative confocal images of the SGZ from 6 month old WT, parkin KO, and SODPAR mice stained against Sox2, Nestin, GFAP, and DAPI. (B) Quantification of the $Sox2^+$, $Sox2^+/Nestin^+$, $Sox2^+/Nestin^+/GFAP^+$, and $Sox2^+/Nestin^+/GFAP^-$ cell populations in the SGZ. Arrowheads label Type I cells ($Sox2^+/Nestin^+/GFAP^+/DAPI^+$). Insets provide a zoomed-in view of the images. Quantitative data is represented as mean ± SD (n=3 per genotype, one-way ANOVA and Tukey's test). Scale bars = 50 µm.

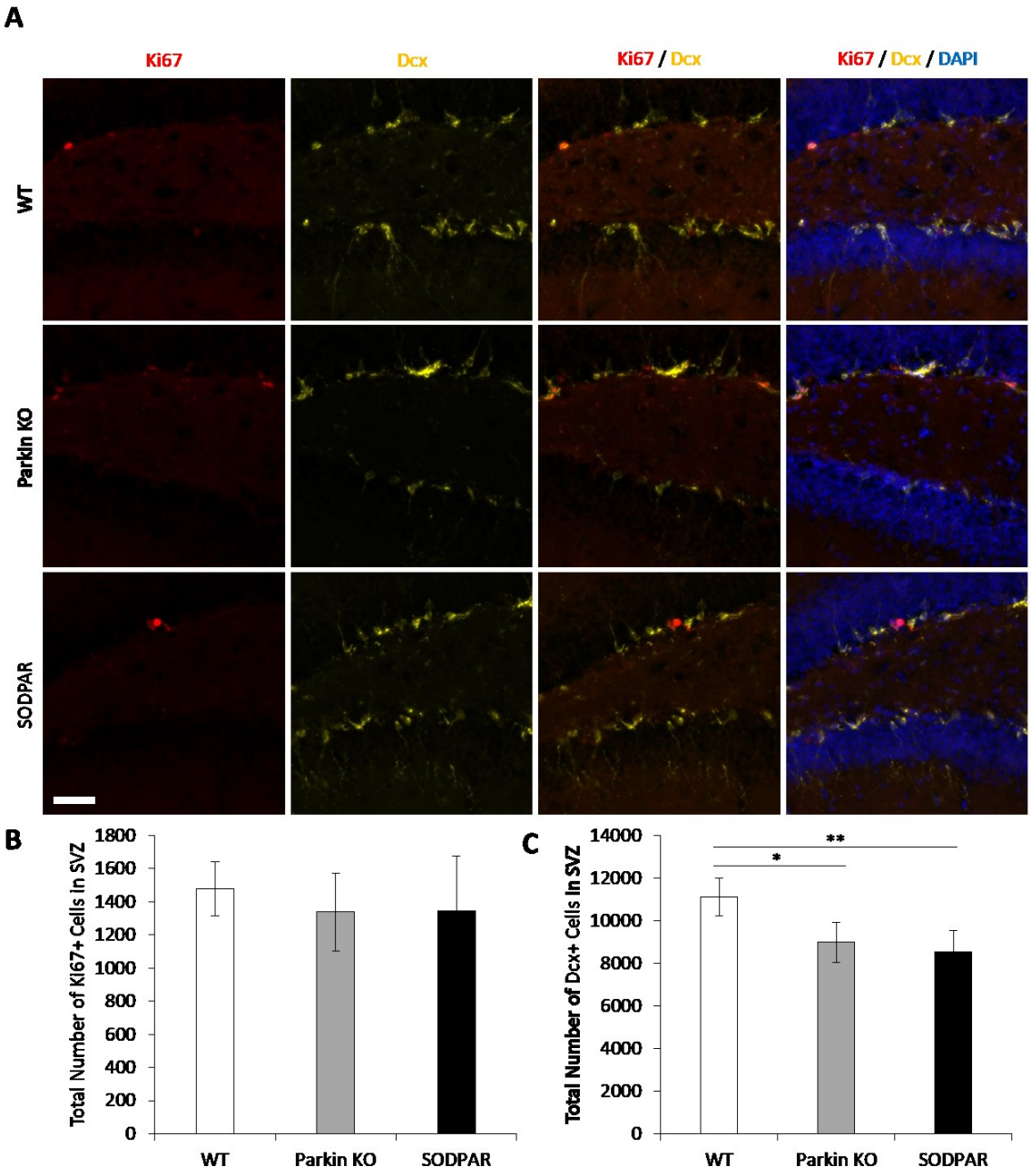

Figure 12: Loss of parkin causes a decrease in the total number of newborn neurons in the adult SGZ. (A) Representative fluorescent images of the SGZ from 6 month old WT, parkin KO, and SODPAR mice stained against Ki67, Dcx, and DAPI. (B) Quantification of the total number of $Ki67^+$ cells in the SGZ. (C) Quantification of the total number of Dcx^+ cells in the SGZ. Quantitative data is represented as mean ± SD (n=3-4 per genotype, *p<0.05, **p<0.01, one-way ANOVA and Tukey's test). Scale bar = 50 μm.

DISCUSSION

4.1. Summary of Results

Given the central role of mitochondria in regulating NSC fate decisions, one would expect PD associated mitochondrial dysfunction to cause defects in adult neurogenesis (Khacho et al., 2019). The goal of this thesis was to assess the impact of a loss of PINK1 or parkin on adult neurogenesis. The results of this thesis support a number of conclusions:

1) PINK1 is required for the maintenance of the activated Type B1 (NSC) population in the adult SVZ.

2) PINK1 is required for cellular proliferation in the SGZ.

3) PINK1 is required for the maintenance of newborn neurons in the SGZ.

4) Parkin is required for the maintenance of newborn neurons in the SGZ.

5) A hemizygous KO of SOD2 is an insufficient stressor to exacerbate defects in adult neurogenesis in a parkin KO mouse model.

Collectively, these findings support the hypothesis that a loss of either PINK1 or parkin would impair adult neurogenesis. The following sections of this thesis discuss the major findings from this study and their implications.

4.2. Role of PINK1 in SVZ Adult Neurogenesis

Our investigation into the role of PINK1 in adult neurogenesis began with determining the impact an absence of PINK1 would have on the neural precursor cell population in the SVZ (Figure 5). Through immunohistochemistry, 6 month old PINK1 KO mice were found to have ~22.77% fewer activated Type B1 cells ($Sox2^+/Nestin^+/GFAP^+$ cells) in the SVZ compared to age matched WT controls (Figure 5B and Table S1). EdU labelling was conducted to test the possibility that a loss of PINK1 impairs cellular proliferation in the SVZ (Figure 6). However, no significant difference in the total number of EdU^+ cells in the SVZ was found between the WT and PINK1 KO mice (Figure 6B and Table S2), suggesting that the absence of PINK1 does not affect cellular proliferation in the SVZ. The observed decline in the activated Type B1 cell population could also have been the result of increased cell death or a defect in NSC activation. Numerous studies have demonstrated PINK1 to promote cell survival under various conditions by preventing apoptosis (Arena et al., 2013; Deng et al., 2005; Gandhi et al., 2009; Haque et al., 2008; Marongiu et al., 2009; Petit et al., 2005; Pridgeon et al., 2007; Wood-Kaczmar et al., 2008). A defect in NSC activation would lead to an increase in the total number of quiescent Type B1 cells at the expense of the activated Type B1 cell population (Daynac et al., 2016).

No significant differences were found between the WT and PINK1 KO mice with respect to the total number of neural precursor cells ($Sox2^+$ cells), nestin expressing neural precursor cells ($Sox2^+/Nestin^+$ cells), or Type C cells ($Sox2^+/Nestin^+/GFAP^-$ cells) in the SVZ (Figure 5B and Table S1). Given that the activated Type B1 cell population is a part of the overall neural precursor cell population, a decline in the total number of activated Type B1 cells should have also affected the total number of neural precursor cells and/or other neural precursor subpopulations (Bond et al., 2015). It is possible that changes in these other cell populations were not detected due to the

presence of high statistical variance. This issue could be resolved by increasing the sample size used to compare the WT and PINK1 KO mice.

After examining the neural precursor cell population and cellular proliferation, we decided to investigate the impact an absence of PINK1 would have on the Type A cell (neuroblast) population in the SVZ (Figure 6). No significant difference was found between WT and PINK1 KO mice with respect to the total number of Dcx^+ cells in the SVZ (Figure 6C and Table S2). This result suggests that the loss of PINK1 does not affect the Type A cell population in the adult SVZ (at least until 6 months of age). Given that there was no significant difference in the total number of Type A cells in the SVZ between the WT and PINK1 KO mice, the observed decrease in the activated Type B1 cell population would likely have occurred relatively recently. In this case, examining additional time-points would allow for the observation of changes in cell populations derived from the activated Type B1 cell population.

4.3. Role of PINK1 in SGZ Adult Neurogenesis

The role of PINK1 in SGZ adult neurogenesis was also investigated by examining the impact an absence of PINK1 would have on the neural precursor cell population, cellular proliferation, and neural precursor derived cells. The absence of PINK1 was not found to affect the neural precursor cell population (Figure 7), with respect to the total number of neural precursor cells ($Sox2^+$ cells), nestin expressing neural precursor cells ($Sox2^+/Nestin^+$ cells), Type I cells ($Sox2^+/Nestin^+/GFAP^+$ cells), or Type IIa cells ($Sox2^+/Nestin^+/GFAP^-$ cells) in the SGZ (Figure 7B and Table S3). Analysis of cellular proliferation in the SGZ (Figure 8) revealed that the PINK1 KO mice had ~25.09% fewer $Ki67^+$ cells (Figure 8B and Table S4) than the WT mice, suggesting that the loss of PINK1 resulted in a cellular proliferation defect in the SGZ. Analysis of cell

populations derived from the neural precursor cell population in the SGZ (Figure 8) revealed that the PINK1 KO mice had ~15.28% fewer Dcx^+ cells (Figure 8C and Table S4) than the WT mice, suggesting that PINK1 is involved with the production of newborn neurons in the SGZ.

Similar results have been reported in a study by Beckervordersandforth et al. (2017), which examined the importance of mitochondrial function in regulating adult hippocampal neurogenesis. In their study, defects in proliferation, differentiation, maturation, and survival were found in the SGZ of adult conditional TFAM KO mice. These defects were discovered to be the result of mitochondrial dysfunction negatively affecting committed neuronal cell-types that primarily rely on OXPHOS more than NSCs that primarily rely on glycolysis (Beckervordersandforth et al., 2017). Given that mitochondrial dysfunction has been observed in adult hippocampal NSCs derived from PINK1 KO mice (Agnihotri et al., 2017), the possibility of mitochondrial dysfunction preferentially affecting committed neuronal cell populations (that rely on OXPHOS) in the SGZ of PINK1 KO mice should be investigated (Beckervordersandforth et al., 2017).

4.4. Role of Parkin in SVZ Adult Neurogenesis

After studying the impact of a loss of PINK1 on adult neurogenesis, we decided to investigate the impact a loss of parkin would have on adult neurogenesis. PINK1 and parkin are known to cooperate in several pathways related to mitochondrial quality control, but independent functions for these proteins have been identified (Exner et al., 2012; McWilliams and Muqit, 2017; Mouton-Liger et al., 2017; Park et al., 2018; Truban et al., 2017). Analysis of the neural precursor cell population (Figure 9) in the SVZ revealed no significant differences between 6 month old WT, parkin KO, and SODPAR mice with respect to the total number of neural precursor cells ($Sox2^+$ cells), nestin expressing neural precursor cells ($Sox2^+$/$Nestin^+$ cells), activated Type B1 cells

(Sox2$^+$/Nestin$^+$/GFAP$^+$ cells), or Type C cells (Sox2$^+$/Nestin$^+$/GFAP$^-$ cells) in the SVZ (Figure 9B and Table S5). No significant differences in cellular proliferation (EdU$^+$ cells) or the total number of Type A cells (Dcx$^+$ cells) in the SVZ were found between the WT, parkin KO, and SODPAR mice (Figure 10 and Table S6). Based on these findings, the loss of parkin (even when combined with a reduction in SOD2) does not seem to affect the neural precursor cell population, cellular proliferation, or the production of newborn neurons in the SVZ (at least up to 6 months of age).

4.5. Role of Parkin in SGZ Adult Neurogenesis

The final part of this thesis involved studying the impact of a loss of parkin on SGZ adult neurogenesis. Examination of the neural precursor cell population in the SGZ revealed no significant differences between WT, parkin KO, and SODPAR mice (Figure 11), with respect to the total number of neural precursor cells (Sox2+ cells), nestin expressing neural precursor cells (Sox2$^+$/Nestin$^+$ cells), Type I cells (Sox2$^+$/Nestin$^+$/GFAP$^+$ cells), or Type IIa cells (Sox2$^+$/Nestin$^+$/GFAP$^-$ cells) in the SGZ (Figure 11B and Table S7). There was also no significant difference in cellular proliferation in the SGZ found between WT, parkin KO, and SODPAR mice (Figure 12B and Table S8). However, the parkin KO and SODPAR mice were found to have significantly fewer (~19.25% and ~23.17% fewer, respectively) Dcx$^+$ cells in the SGZ, compared to the WT mice (Figure 12C and Table S8). This would imply a decline in the production of newborn neurons, which could be the result of a defect in neuronal differentiation and/or survival in the SGZ. Similar to the PINK1 KO data, the decline in newborn neurons could be the result of mitochondrial dysfunction (caused by a loss of Parkin) impairing differentiation and survival in committed neuronal cell populations that rely primarily on OXPHOS (Beckervordersandforth et al., 2017).

4.6. No Observable Differences between Parkin KO and SODPAR Mice

While investigating the role of parkin in adult neurogenesis, no significant differences in SVZ or SGZ cell populations were observed between the parkin KO and SODPAR mice. The SODPAR mice were initially expected to display more pronounced changes in adult neurogenesis than the parkin KO mice, due to the combination of an absence of parkin with a reduction in the mitochondrial antioxidant enzyme SOD2 (Lebovitz et al., 1996; Weisiger and Fridovich, 1973; Zelko et al., 2002). However, the results of this thesis do not support the notation of a hemizygous KO of SOD2 as being a sufficient cellular stressor to exacerbate defects in adult neurogenesis caused by a loss of parkin.

4.7. Future Directions

In this thesis, adult neurogenesis in the SVZ and SGZ was examined at a single time-point (6 months of age) using immunohistochemistry to label a limited number of different cell populations. While this approach has provided some preliminary data on the importance of PINK1 and parkin in adult neurogenesis, a complete characterization of adult neurogenesis in our PD mouse models is still required. This would involve using additional immunohistochemical markers to refine the analysis of specific subpopulations of cells within the SVZ and SGZ. For example, markers for GFAP, prominin-1 (CD133), and the epidermal growth factor receptor (EGFR) can be used to differentiate quiescent NSCs ($GFAP^+/CD133^+/EGFR^-$ cells) from activated NSCs ($GFAP^+/CD133^+/EGFR^+$ cells) in the adult SVZ (Codega et al., 2014). Multiple time-points would need to be examined since PD is a progressive neurodegenerative disease (Exner et al., 2012), meaning that defects in adult neurogenesis may only become salient at an advanced age (Marxreiter et al., 2013b). Cell intrinsic and extrinsic regulation of NSC self-renewal,

differentiation, proliferation, and survival can also be assessed *in vitro* by studying neurospheres (Pastrana et al., 2011).

Several studies have shown PINK1 and parkin to be necessary for neuronal survival and maturation (Agnihotri et al., 2017; Dagda et al., 2014; Giguère et al., 2018; Wood-Kaczmar et al., 2008). Defects in survival have been reported for NSC derived human dopaminergic neurons with a knockdown in PINK1 and primary cortical neurons from PINK1 KO mice (Wood-Kaczmar et al., 2008). Another study, using primary cortical and midbrain neurons from PINK1 KO mice, found PINK1 to be involved with the regulation of dendritic length and complexity (Dagda et al., 2014). In the context of adult hippocampal neurogenesis, Dcx^+ newborn neurons in the SGZ of 10-12 week old PINK1 KO mice were discovered to have dendrites that were structurally less complex than those from WT control mice (Agnihotri et al., 2017). In a study by Giguère et al. (2018), cultured dopaminergic neurons from the SN of newborn parkin KO mouse pups (P0-P2) were found to develop defects in survival and axonal arborization over time. In this thesis, we did not assess long-term survival or activity of neurons being produced in the adult SVZ or SGZ from our PD mouse models. Lineage tracing experiments will need to be conducted in order to answer questions related to neuronal differentiation, survival, and functionality *in vivo* (Enikolopov et al., 2015).

Given the lack of robust changes in adult neurogenesis observed in the 6 month old PD mouse models used for this project, it may be prudent to use a different PD mouse model. For example, the parkin-mutator mouse model [which lacks parkin and has a mutated version of the mitochondrial DNA polymerase γ (POLG) with impaired proofreading function] has been reported to display progressive dopaminergic neurodegeneration (Goldberg et al., 2003; Kujoth et al., 2005; Pickrell et al., 2015). Evidence of aberrant changes to neurogenesis have been reported in the

progeroid POLG mutator mouse model (Ahlqvist et al., 2012). Neurospheres derived from embryonic POLG mutator mice have shown impaired self-renewal. In adult POLG mutator mice over 40 weeks of age, a decrease in the total number of nestin^{+} neural precursor cells in the SVZ has been observed, with no changes in the total number of proliferating cells in the SVZ or calbindin^{+} periglomerular interneurons in the OBs (Ahlqvist et al., 2012). The mitochondrial dysfunction associated with an absence of parkin combined with a defect in POLG activity (Pickrell et al., 2015) could potentially be severe enough to cause aberrant changes to adult neurogenesis (Beckervordersandforth et al., 2017; Khacho et al., 2016). Having a PD mouse model that displays robust changes in adult neurogenesis (especially at an earlier age of onset) would help to streamline research on the impact of PD associated mitochondrial dysfunction on adult neurogenesis.

After a detailed characterization of adult neurogenesis in a suitable PD mouse model, the next phase of this project would be to determine if observed changes in adult neurogenesis are the result of mitochondrial dysfunction. This would involve three key steps: (1) assessing mitochondrial integrity, (2) identifying molecular pathways responsible for aberrant changes to adult neurogenesis, and (3) performing phenotypic rescue experiments.

There are several parameters that can be examined in order to assess mitochondrial integrity within a suitable PD mouse model. Measuring the lengths of individual mitochondria in cells, through immunohistochemical staining of Tom20 (translocase of the outer membrane subunit 20), can be done to assess mitochondrial dynamics (Khacho et al., 2016). Parameters such as mitochondrial membrane potential ($\Delta\Psi m$), ATP production, cellular and mitochondrial ROS levels, oxygen consumption rate (OCR), and extracellular acidification rate (ECAR) can be examined in order to assess mitochondrial function and cellular metabolism (Agnihotri et al., 2017;

Khacho et al., 2016). Mitophagy can also be examined given the potential involvement of dysfunctional mitophagy in PD pathology (Cummins and Götz, 2018; Pickrell and Youle, 2015) and the mounting evidence supporting an involvement of autophagy in adult neurogenesis (Casares-Crespo et al., 2018). It is expected that there would be signs of mitochondrial dysfunction and altered cellular metabolism in the chosen PD mouse model, given the intimate relationship between PD pathology and mitochondrial dysfunction (Exner et al., 2012). In the study by Agnihotri et al. (2017), analysis of mitochondrial potential, OCR, and ECAR found evidence of mitochondrial dysfunction and a greater reliance on glycolysis for energy production in hippocampal NSCs derived from 10-12 week old PINK1 KO mice. A careful analysis of mitochondrial integrity would be necessary in order to be able to later identify key molecular pathways and plan appropriate phenotypic rescue experiments.

The identification of molecular pathways responsible for aberrant changes to adult neurogenesis can be achieved by using RNA-sequencing (RNA-seq) to identify potential genes of interest. In the study by Khacho et al. (2016), RNA-seq was used to study the mechanism behind the regulation of embryonic neurogenesis through mitochondrial dynamics. Over-represented transcription factors revealed through RNA-seq were analyzed in order to identify changes in groups of genes, involved with a particular cellular function (Khacho et al., 2016). A similar approach could be applied to studying the impact of PD associated mitochondrial dysfunction on adult neurogenesis. Single-cell RNA-seq, which has been successfully used to study the progression of neurogenesis in the adult SGZ (Shin et al., 2015) and SVZ (Zywitza et al., 2018), can be used to identify changes in specific cell populations.

Lastly, phenotypic rescue experiments will need to be conducted in order to demonstrate the possibility of rescuing adult neurogenesis in a chosen PD mouse model through the restoration

of mitochondrial integrity. Transgenic and pharmacological approaches can be used to restore mitochondrial integrity both *in vitro* and *in vivo* (Khacho et al., 2016; Sorrentino et al., 2018). Transgenic rescue experiments will involve modulating the expression of specific genes (levels of specific proteins) to improve mitochondrial dynamics and function. For example, the PD model mice could be crossed with mice overexpressing Opa1 ($Opa1^{tg}$ mice), which are reported to have improved OXPHOS efficiency as a result of mitochondrial elongation and improved mitochondrial cristae structure organization (Civiletto et al., 2015; Varanita et al., 2015). Pharmacological rescue experiments will involve the use of small molecule compounds to improve mitochondrial dynamics and function. Potential compounds that can be tested include mdivi-1 (Mitochondrial Division Inhibitor 1), nicotinamide riboside (NR), and piracetam (Beckervordersandforth et al., 2017; Cassidy-Stone et al., 2008; Cui et al., 2010; Zhang et al., 2016). Mdivi-1 promotes mitochondrial fusion by inhibiting the mitochondrial fission protein Drp1 (Cassidy-Stone et al., 2008). This compound was found to ameliorate aberrant changes to mitochondrial dynamics and function in PINK1 mutant B27 cells (Cui et al., 2010). NR supplementation in aged mice has been reported to revitalize muscle, neural, and melanocyte stem cell populations (Zhang et al., 2016). Piracetam treatment of aged mice was found to improve adult hippocampal neurogenesis by enhancing neuronal differentiation and maturation (Beckervordersandforth et al., 2017). The efficacy of all phenotypic rescue experiments can be assessed using metrics previously mentioned in this thesis.

CONCLUSION

The purpose of this thesis was to investigate the impact a loss of PINK1 or parkin would have on adult neurogenesis in the SVZ and SGZ neurogenic niches. In the adult SVZ, PINK1 was found to be required for the maintenance of the activated Type B1 cell population. In the adult SGZ, PINK1 was found to be involved with cellular proliferation and the production of newborn neurons. The loss of parkin was not found to affect neurogenesis in the adult SVZ, but did cause a decline in the production of newborn neurons in the SGZ. These findings support our hypothesis that a loss of either PINK1 or parkin would impair adult neurogenesis. Future studies should focus on completing the characterization of adult neurogenesis in these PD mouse models and investigating the potential involvement of mitochondrial dysfunction. In order to streamline this research, a different mouse model of PD that displays robust changes in adult neurogenesis could be studied. Understanding the effect of PD on endogenous neural regeneration is essential for the development of effective therapeutic treatments for PD patients that are based on the use of NSCs to replace lost neurons.

REFERENCES

Agnihotri, S.K., Shen, R., Li, J., Gao, X., and Büeler, H. (2017). Loss of PINK1 leads to metabolic deficits in adult neural stem cells and impedes differentiation of newborn neurons in the mouse hippocampus. FASEB J. *31*, 2839–2853.

Ahlqvist, K.J., Hämäläinen, R.H., Yatsuga, S., Uutela, M., Terzioglu, M., Götz, A., Forsström, S., Salven, P., Angers-Loustau, A., Kopra, O.H., et al. (2012). Somatic progenitor cell vulnerability to mitochondrial DNA mutagenesis underlies progeroid phenotypes in Polg mutator mice. Cell Metab. *15*, 100–109.

Akundi, R.S., Huang, Z., Eason, J., Pandya, J.D., Zhi, L., Cass, W.A., Sullivan, P.G., and Büeler, H. (2011). Increased Mitochondrial Calcium Sensitivity and Abnormal Expression of Innate Immunity Genes Precede Dopaminergic Defects in Pink1-Deficient Mice. PLoS One *6*, e16038.

Allen, G.F.G., Toth, R., James, J., and Ganley, I.G. (2013). Loss of iron triggers PINK1/Parkin-independent mitophagy. EMBO Rep. *14*, 1127–1135.

Almeida, A., Almeida, J., Bolanos, J.P., and Moncada, S. (2001). Different responses of astrocytes and neurons to nitric oxide: The role of glycolytically generated ATP in astrocyte protection. Proc. Natl. Acad. Sci. *98*, 15294–15299.

Almeida, A., Moncada, S., and Bolaños, J.P. (2004). Nitric oxide switches on glycolysis through the AMP protein kinase and 6-phosphofructo-2-kinase pathway. Nat. Cell Biol. *6*, 45–51.

Altman, J. (1969). Autoradiographic and histological studies of postnatal neurogenesis. IV. Cell proliferation and migration in the anterior forebrain, with special reference to persisting neurogenesis in the olfactory bulb. J. Comp. Neurol. *137*, 433–457.

Altman, J., and Das, G.D. (1965). Autoradiographic and histological evidence of postnatal hippocampal neurogenesis in rats. J. Comp. Neurol. *124*, 319–335.

Anderson, D., Beecher, G., and Ba, F. (2017). Deep Brain Stimulation in Parkinson's Disease: New and Emerging Targets for Refractory Motor and Nonmotor Symptoms. Parkinsons. Dis. *2017*, 1–13.

Andersson, U., and Scarpulla, R.C. (2001). PGC-1-Related Coactivator, a Novel, Serum-Inducible Coactivator of Nuclear Respiratory Factor 1-Dependent Transcription in Mammalian Cells. Mol. Cell. Biol. *21*, 3738–3749.

Arena, G., Gelmetti, V., Torosantucci, L., Vignone, D., Lamorte, G., De Rosa, P., Cilia, E., Jonas, E.A., and Valente, E.M. (2013). PINK1 protects against cell death induced by mitochondrial depolarization, by phosphorylating Bcl-xL and impairing its pro-apoptotic cleavage. Cell Death Differ. *20*, 920–930.

Ashrafi, G., Schlehe, J.S., LaVoie, M.J., and Schwarz, T.L. (2014). Mitophagy of damaged mitochondria occurs locally in distal neuronal axons and requires PINK1 and Parkin. J. Cell Biol. *206*, 655–670.

Baker, S.A., Baker, K.A., and Hagg, T. (2004). Dopaminergic nigrostriatal projections regulate neural precursor proliferation in the adult mouse subventricular zone. Eur. J. Neurosci. *20*, 575–

579.

Beal, M.F. (2005). Mitochondria take center stage in aging and neurodegeneration. Ann. Neurol. *58*, 495–505.

Beckervordersandforth, R., Ebert, B., Schäffner, I., Moss, J., Fiebig, C., Shin, J., Moore, D.L., Ghosh, L., Trinchero, M.F., Stockburger, C., et al. (2017). Role of Mitochondrial Metabolism in the Control of Early Lineage Progression and Aging Phenotypes in Adult Hippocampal Neurogenesis. Neuron *93*, 560-573.e6.

Beilina, A., Van Der Brug, M., Ahmad, R., Kesavapany, S., Miller, D.W., Petsko, G.A., and Cookson, M.R. (2005). Mutations in PTEN-induced putative kinase 1 associated with recessive parkinsonism have differential effects on protein stability. Proc. Natl. Acad. Sci. *102*, 5703–5708.

Le Belle, J.E., Orozco, N.M., Paucar, A.A., Saxe, J.P., Mottahedeh, J., Pyle, A.D., Wu, H., and Kornblum, H.I. (2011). Proliferative Neural Stem Cells Have High Endogenous ROS Levels that Regulate Self-Renewal and Neurogenesis in a PI3K/Akt-Dependant Manner. Cell Stem Cell *8*, 59–71.

Benard, G., and Rossignol, R. (2008). Ultrastructure of the Mitochondrion and Its Bearing on Function and Bioenergetics. Antioxid. Redox Signal. *10*, 1313–1342.

Bender, A., Krishnan, K.J., Morris, C.M., Taylor, G.A., Reeve, A.K., Perry, R.H., Jaros, E., Hersheson, J.S., Betts, J., Klopstock, T., et al. (2006). High levels of mitochondrial DNA deletions in substantia nigra neurons in aging and Parkinson disease. Nat. Genet. *38*, 515–517.

Berg, D.A., Belnoue, L., Song, H., and Simon, A. (2013). Neurotransmitter-mediated control of neurogenesis in the adult vertebrate brain. Development *140*, 2548–2561.

Bergmann, O., Liebl, J., Bernard, S., Alkass, K., Yeung, M.S.Y., Steier, P., Kutschera, W., Johnson, L., Landén, M., Druid, H., et al. (2012). The Age of Olfactory Bulb Neurons in Humans. Neuron *74*, 634–639.

Bergmann, O., Spalding, K.L., and Frisén, J. (2015). Adult Neurogenesis in Humans. Cold Spring Harb. Perspect. Biol. *7*, a018994.

Bertolin, G., Ferrando-Miguel, R., Jacoupy, M., Traver, S., Grenier, K., Greene, A.W., Dauphin, A., Waharte, F., Bayot, A., Salamero, J., et al. (2013). The TOMM machinery is a molecular switch in PINK1 and PARK2/PARKIN-dependent mitochondrial clearance. Autophagy *9*, 1801–1817.

Bhujabal, Z., Birgisdottir, Å.B., Sjøttem, E., Brenne, H.B., Øvervatn, A., Habisov, S., Kirkin, V., Lamark, T., and Johansen, T. (2017). FKBP8 recruits LC3A to mediate Parkin-independent mitophagy. EMBO Rep. *18*, 947–961.

Bingol, B., Tea, J.S., Phu, L., Reichelt, M., Bakalarski, C.E., Song, Q., Foreman, O., Kirkpatrick, D.S., and Sheng, M. (2014). The mitochondrial deubiquitinase USP30 opposes parkin-mediated mitophagy. Nature *510*, 370–375.

Boekhoorn, K., Joels, M., and Lucassen, P.J. (2006). Increased proliferation reflects glial and vascular-associated changes, but not neurogenesis in the presenile Alzheimer hippocampus.

Neurobiol. Dis. *24*, 1–14.

Bolaños, J.P. (2016). Bioenergetics and redox adaptations of astrocytes to neuronal activity. J. Neurochem. *139 Suppl 2*, 115–125.

Boldrini, M., Fulmore, C.A., Tartt, A.N., Simeon, L.R., Pavlova, I., Poposka, V., Rosoklija, G.B., Stankov, A., Arango, V., Dwork, A.J., et al. (2018). Human Hippocampal Neurogenesis Persists throughout Aging. Cell Stem Cell *22*, 589-599.e5.

Bond, A.M., Ming, G.L., and Song, H. (2015). Adult Mammalian Neural Stem Cells and Neurogenesis: Five Decades Later. Cell Stem Cell *17*, 385–395.

Bonifati, V., Rizzu, P., Van Baren, M.J., Schaap, O., Breedveld, G.J., Krieger, E., Dekker, M.C.J., Squitieri, F., Ibanez, P., Joosse, M., et al. (2003). Mutations in the DJ-1 gene associated with autosomal recessive early-onset parkinsonism. Science *299*, 256–259.

Braak, H., and Braak, E. (2000). Pathoanatomy of Parkinson's disease. J. Neurol. *247*, II3–II10.

Braak, H., Ghebremedhin, E., Rüb, U., Bratzke, H., and Del Tredici, K. (2004). Stages in the development of Parkinson's disease-related pathology. Cell Tissue Res. *318*, 121–134.

Braschi, E., Goyon, V., Zunino, R., Mohanty, A., Xu, L., and McBride, H.M. (2010). Vps35 Mediates Vesicle Transport between the Mitochondria and Peroxisomes. Curr. Biol. *20*, 1310–1315.

Breton-Provencher, V., Lemasson, M., Peralta, M.R., and Saghatelyan, A. (2009). Interneurons Produced in Adulthood Are Required for the Normal Functioning of the Olfactory Bulb Network and for the Execution of Selected Olfactory Behaviors. J. Neurosci. *29*, 15245–15257.

Brickley, K., and Stephenson, F.A. (2011). Trafficking Kinesin Protein (TRAK)-mediated Transport of Mitochondria in Axons of Hippocampal Neurons. J. Biol. Chem. *286*, 18079–18092.

Buhlman, L., Damiano, M., Bertolin, G., Ferrando-Miguel, R., Lombès, A., Brice, A., and Corti, O. (2014). Functional interplay between Parkin and Drp1 in mitochondrial fission and clearance. Biochim. Biophys. Acta - Mol. Cell Res. *1843*, 2012–2026.

Cai, Q., Zakaria, H.M., Simone, A., and Sheng, Z.-H. (2012). Spatial Parkin Translocation and Degradation of Damaged Mitochondria via Mitophagy in Live Cortical Neurons. Curr. Biol. *22*, 545–552.

Capilla-Gonzalez, V., Cebrian-Silla, A., Guerrero-Cazares, H., Garcia-Verdugo, J.M., and Quiñones-Hinojosa, A. (2014). Age-related changes in astrocytic and ependymal cells of the subventricular zone. Glia *62*, 790–803.

Casares-Crespo, L., Calatayud-Baselga, I., García-Corzo, L., and Mira, H. (2018). On the Role of Basal Autophagy in Adult Neural Stem Cells and Neurogenesis. Front. Cell. Neurosci. *12*, 339.

Cassidy-Stone, A., Chipuk, J.E., Ingerman, E., Song, C., Yoo, C., Kuwana, T., Kurth, M.J., Shaw, J.T., Hinshaw, J.E., Green, D.R., et al. (2008). Chemical Inhibition of the Mitochondrial Division Dynamin Reveals Its Role in Bax/Bak-Dependent Mitochondrial Outer Membrane

Permeabilization. Dev. Cell *14*, 193–204.

Chartier-Harlin, M.-C., Kachergus, J., Roumier, C., Mouroux, V., Douay, X., Lincoln, S., Levecque, C., Larvor, L., Andrieux, J., Hulihan, M., et al. (2004). α-synuclein locus duplication as a cause of familial Parkinson's disease. Lancet *364*, 1167–1169.

Chehrehasa, F., Meedeniya, A.C.B., Dwyer, P., Abrahamsen, G., and Mackay-Sim, A. (2009). EdU, a new thymidine analogue for labelling proliferating cells in the nervous system. J. Neurosci. Methods *177*, 122–130.

Chen, C.-T., Shih, Y.-R. V., Kuo, T.K., Lee, O.K., and Wei, Y.-H. (2008). Coordinated Changes of Mitochondrial Biogenesis and Antioxidant Enzymes During Osteogenic Differentiation of Human Mesenchymal Stem Cells. Stem Cells *26*, 960–968.

Chen, H., Detmer, S.A., Ewald, A.J., Griffin, E.E., Fraser, S.E., and Chan, D.C. (2003). Mitofusins Mfn1 and Mfn2 coordinately regulate mitochondrial fusion and are essential for embryonic development. J. Cell Biol. *160*, 189–200.

Cheung, E.C.C., Joza, N., Steenaart, N.A.E., McClellan, K.A., Neuspiel, M., McNamara, S., MacLaurin, J.G., Rippstein, P., Park, D.S., Shore, G.C., et al. (2006). Dissociating the dual roles of apoptosis-inducing factor in maintaining mitochondrial structure and apoptosis. EMBO J. *25*, 4061–4073.

Chi, Z., Zhang, J., Tokunaga, A., Harraz, M.M., Byrne, S.T., Dolinko, A., Xu, J., Blackshaw, S., Gaiano, N., Dawson, T.M., et al. (2012). Botch Promotes Neurogenesis by Antagonizing Notch. Dev. Cell *22*, 707–720.

Cho, Y.M., Kwon, S., Pak, Y.K., Seol, H.W., Choi, Y.M., Park, D.J., Park, K.S., and Lee, H.K. (2006). Dynamic changes in mitochondrial biogenesis and antioxidant enzymes during the spontaneous differentiation of human embryonic stem cells. Biochem. Biophys. Res. Commun. *348*, 1472–1478.

Choe, Y., Pleasure, S.J., and Mira, H. (2016). Control of Adult Neurogenesis by Short-Range Morphogenic-Signaling Molecules. Cold Spring Harb. Perspect. Biol. *8*, a018887.

Chu, C.T., Ji, J., Dagda, R.K., Jiang, J.F., Tyurina, Y.Y., Kapralov, A.A., Tyurin, V.A., Yanamala, N., Shrivastava, I.H., Mohammadyani, D., et al. (2013). Cardiolipin externalization to the outer mitochondrial membrane acts as an elimination signal for mitophagy in neuronal cells. Nat. Cell Biol. *15*, 1197–1205.

Chung, S., Dzeja, P.P., Faustino, R.S., Perez-Terzic, C., Behfar, A., and Terzic, A. (2007). Mitochondrial oxidative metabolism is required for the cardiac differentiation of stem cells. Nat. Clin. Pract. Cardiovasc. Med. *4*, S60-7.

Chung, S., Arrell, D.K., Faustino, R.S., Terzic, A., and Dzeja, P.P. (2010). Glycolytic network restructuring integral to the energetics of embryonic stem cell cardiac differentiation. J. Mol. Cell. Cardiol. *48*, 725–734.

Cipolat, S., de Brito, O.M., Dal Zilio, B., and Scorrano, L. (2004). OPA1 requires mitofusin 1 to promote mitochondrial fusion. Proc. Natl. Acad. Sci. *101*, 15927–15932.

Cipolat, S., Rudka, T., Hartmann, D., Costa, V., Serneels, L., Craessaerts, K., Metzger, K.,

Frezza, C., Annaert, W., D'Adamio, L., et al. (2006). Mitochondrial Rhomboid PARL Regulates Cytochrome c Release during Apoptosis via OPA1-Dependent Cristae Remodeling. Cell *126*, 163–175.

Civiletto, G., Varanita, T., Cerutti, R., Gorletta, T., Barbaro, S., Marchet, S., Lamperti, C., Viscomi, C., Scorrano, L., and Zeviani, M. (2015). Opa1 overexpression ameliorates the phenotype of two mitochondrial disease mouse models. Cell Metab. *21*, 845–854.

Clark, I.E., Dodson, M.W., Jiang, C., Cao, J.H., Huh, J.R., Seol, J.H., Yoo, S.J., Hay, B.A., and Guo, M. (2006). Drosophila pink1 is required for mitochondrial function and interacts genetically with parkin. Nature *441*, 1162–1166.

Clelland, C.D., Choi, M., Romberg, C., Clemenson, G.D., Fragniere, A., Tyers, P., Jessberger, S., Saksida, L.M., Barker, R.A., Gage, F.H., et al. (2009). A functional role for adult hippocampal neurogenesis in spatial pattern separation. Science *325*, 210–213.

Codega, P., Silva-Vargas, V., Paul, A., Maldonado-Soto, A.R., DeLeo, A.M., Pastrana, E., and Doetsch, F. (2014). Prospective Identification and Purification of Quiescent Adult Neural Stem Cells from Their In Vivo Niche. Neuron *82*, 545–559.

von Coelln, R., Thomas, B., Savitt, J.M., Lim, K.L., Sasaki, M., Hess, E.J., Dawson, V.L., and Dawson, T.M. (2004). Loss of locus coeruleus neurons and reduced startle in parkin null mice. Proc. Natl. Acad. Sci. *101*, 10744–10749.

Cogliati, S., Frezza, C., Soriano, M.E., Varanita, T., Quintana-Cabrera, R., Corrado, M., Cipolat, S., Costa, V., Casarin, A., Gomes, L.C., et al. (2013). Mitochondrial Cristae Shape Determines Respiratory Chain Supercomplexes Assembly and Respiratory Efficiency. Cell *155*, 160–171.

Conover, J.C., and Notti, R.Q. (2008). The neural stem cell niche. Cell Tissue Res. *331*, 211–224.

Cornelissen, T., Vilain, S., Vints, K., Gounko, N., Verstreken, P., and Vandenberghe, W. (2018). Deficiency of parkin and PINK1 impairs age-dependent mitophagy in drosophila. Elife *7*, e35878.

Costa, R.A.P., Fernandes, M.P., de Souza-Pinto, N.C., and Vercesi, A.E. (2013). Protective effects of l-carnitine and piracetam against mitochondrial permeability transition and PC3 cell necrosis induced by simvastatin. Eur. J. Pharmacol. *701*, 82–86.

Cui, M., Tang, X., Christian, W. V, Yoon, Y., and Tieu, K. (2010). Perturbations in mitochondrial dynamics induced by human mutant PINK1 can be rescued by the mitochondrial division inhibitor mdivi-1. J. Biol. Chem. *285*, 11740–11752.

Cummins, N., and Götz, J. (2018). Shedding light on mitophagy in neurons: what is the evidence for PINK1/Parkin mitophagy in vivo? Cell. Mol. Life Sci. *75*, 1151–1162.

Curtis, M.A., Kam, M., Nannmark, U., Anderson, M.F., Axell, M.Z., Wikkelso, C., Holtas, S., van Roon-Mom, W.M.C., Bjork-Eriksson, T., Nordborg, C., et al. (2007). Human Neuroblasts Migrate to the Olfactory Bulb via a Lateral Ventricular Extension. Science *315*, 1243–1249.

Dagda, R.K., Cherra, S.J., Kulich, S.M., Tandon, A., Park, D., and Chu, C.T. (2009). Loss of PINK1 Function Promotes Mitophagy through Effects on Oxidative Stress and Mitochondrial

Fission. J. Biol. Chem. *284*, 13843–13855.

Dagda, R.K., Pien, I., Wang, R., Zhu, J., Wang, K.Z.Q., Callio, J., Banerjee, T. Das, Dagda, R.Y., and Chu, C.T. (2014). Beyond the mitochondrion: cytosolic PINK1 remodels dendrites through protein kinase A. J. Neurochem. *128*, 864–877.

Davis, G.C., Williams, A.C., Markey, S.P., Ebert, M.H., Caine, E.D., Reichert, C.M., and Kopin, I.J. (1979). Chronic Parkinsonism secondary to intravenous injection of meperidine analogues. Psychiatry Res. *1*, 249–254.

Daynac, M., Tirou, L., Faure, H., Mouthon, M.-A., Gauthier, L.R., Hahn, H., Boussin, F.D., and Ruat, M. (2016). Hedgehog Controls Quiescence and Activation of Neural Stem Cells in the Adult Ventricular-Subventricular Zone. Stem Cell Reports *7*, 735–748.

Deng, H., Jankovic, J., Guo, Y., Xie, W., and Le, W. (2005). Small interfering RNA targeting the PINK1 induces apoptosis in dopaminergic cells SH-SY5Y. Biochem. Biophys. Res. Commun. *337*, 1133–1138.

Deng, H., Dodson, M.W., Huang, H., and Guo, M. (2008). The Parkinson's disease genes pink1 and parkin promote mitochondrial fission and/or inhibit fusion in Drosophila. Proc. Natl. Acad. Sci. *105*, 14503–14508.

Doetsch, F., and Alvarez-Buylla, A. (1996). Network of tangential pathways for neuronal migration in adult mammalian brain. Proc. Natl. Acad. Sci. U. S. A. *93*, 14895–14900.

Doetsch, F., García-Verdugo, J.M., and Alvarez-Buylla, A. (1997). Cellular composition and three-dimensional organization of the subventricular germinal zone in the adult mammalian brain. J. Neurosci. *17*, 5046–5061.

Doetsch, F., García-Verdugo, J.M., and Alvarez-Buylla, A. (1999a). Regeneration of a germinal layer in the adult mammalian brain. Proc. Natl. Acad. Sci. U. S. A. *96*, 11619–11624.

Doetsch, F., Caillé, I., Lim, D.A., García-Verdugo, J.M., and Alvarez-Buylla, A. (1999b). Subventricular zone astrocytes are neural stem cells in the adult mammalian brain. Cell *97*, 703–716.

Enikolopov, G., Overstreet-Wadiche, L., and Ge, S. (2015). Viral and transgenic reporters and genetic analysis of adult neurogenesis. Cold Spring Harb. Perspect. Biol. *7*, a018804.

Eriksson, P.S., Perfilieva, E., Björk-Eriksson, T., Alborn, A.M., Nordborg, C., Peterson, D.A., and Gage, F.H. (1998). Neurogenesis in the adult human hippocampus. Nat. Med. *4*, 1313–1317.

Ernst, A., and Frisén, J. (2015). Adult Neurogenesis in Humans- Common and Unique Traits in Mammals. PLOS Biol. *13*, e1002045.

Ernst, A., Alkass, K., Bernard, S., Salehpour, M., Perl, S., Tisdale, J., Possnert, G., Druid, H., and Frisén, J. (2014). Neurogenesis in the striatum of the adult human brain. Cell *156*, 1072–1083.

Exner, N., Treske, B., Paquet, D., Holmstrom, K., Schiesling, C., Gispert, S., Carballo-Carbajal, I., Berg, D., Hoepken, H.-H., Gasser, T., et al. (2007). Loss-of-Function of Human PINK1 Results in Mitochondrial Pathology and Can Be Rescued by Parkin. J. Neurosci. *27*, 12413–

12418.

Exner, N., Lutz, A.K., Haass, C., Winklhofer, K.F., Abeliovich, A., Schmitz, Y., Farinas, I., Choi-Lundberg, D., Ho, W., Castillo, P., et al. (2012). Mitochondrial dysfunction in Parkinson's disease: molecular mechanisms and pathophysiological consequences. EMBO J. *31*, 3038–3062.

Facucho-Oliveira, J.M., Alderson, J., Spikings, E.C., Egginton, S., and St. John, J.C. (2007). Mitochondrial DNA replication during differentiation of murine embryonic stem cells. J. Cell Sci. *120*, 4025–4034.

Faigle, R., and Song, H. (2013). Signaling mechanisms regulating adult neural stem cells and neurogenesis. Biochim. Biophys. Acta *1830*, 2435–2448.

Folmes, C.D.L., Nelson, T.J., Martinez-Fernandez, A., Arrell, D.K., Lindor, J.Z., Dzeja, P.P., Ikeda, Y., Perez-Terzic, C., and Terzic, A. (2011). Somatic Oxidative Bioenergetics Transitions into Pluripotency-Dependent Glycolysis to Facilitate Nuclear Reprogramming. Cell Metab. *14*, 264–271.

Fournier, M., Vitte, J., Garrigue, J., Langui, D., Dullin, J.-P., Saurini, F., Hanoun, N., Perez-Diaz, F., Cornilleau, F., Joubert, C., et al. (2009). Parkin Deficiency Delays Motor Decline and Disease Manifestation in a Mouse Model of Synucleinopathy. PLoS One *4*, e6629.

Frank, S., Gaume, B., Bergmann-Leitner, E.S., Leitner, W.W., Robert, E.G., Catez, F., Smith, C.L., and Youle, R.J. (2001). The role of dynamin-related protein 1, a mediator of mitochondrial fission, in apoptosis. Dev. Cell *1*, 515–525.

Fu, M., St-Pierre, P., Shankar, J., Wang, P.T.C., Joshi, B., and Nabi, I.R. (2013). Regulation of mitophagy by the Gp78 E3 ubiquitin ligase. Mol. Biol. Cell *24*, 1153–1162.

Funayama, M., Hasegawa, K., Kowa, H., Saito, M., Tsuji, S., and Obata, F. (2002). A new locus for Parkinson's disease (PARK8) maps to chromosome 12p11.2-q13.1. Ann. Neurol. *51*, 296–301.

Gage, F.H., Coates, P.W., Palmer, T.D., Kuhn, H.G., Fisher, L.J., Suhonen, J.O., Peterson, D.A., Suhr, S.T., and Ray, J. (1995). Survival and differentiation of adult neuronal progenitor cells transplanted to the adult brain. Proc. Natl. Acad. Sci. U. S. A. *92*, 11879–11883.

Gandhi, S., Wood-Kaczmar, A., Yao, Z., Plun-Favreau, H., Deas, E., Klupsch, K., Downward, J., Latchman, D.S., Tabrizi, S.J., Wood, N.W., et al. (2009). PINK1-Associated Parkinson's Disease Is Caused by Neuronal Vulnerability to Calcium-Induced Cell Death. Mol. Cell *33*, 627–638.

García-Verdugo, J.M., Doetsch, F., Wichterle, H., Lim, D.A., and Alvarez-Buylla, A. (1998). Architecture and cell types of the adult subventricular zone: in search of the stem cells. J. Neurobiol. *36*, 234–248.

Gautier, C.A., Kitada, T., and Shen, J. (2008). Loss of PINK1 causes mitochondrial functional defects and increased sensitivity to oxidative stress. Proc. Natl. Acad. Sci. *105*, 11364–11369.

Gegg, M.E., Cooper, J.M., Chau, K.-Y., Rojo, M., Schapira, A.H.V., and Taanman, J.-W. (2010). Mitofusin 1 and mitofusin 2 are ubiquitinated in a PINK1/parkin-dependent manner upon induction of mitophagy. Hum. Mol. Genet. *19*, 4861–4870.

Gehrke, S., Wu, Z., Klinkenberg, M., Sun, Y., Auburger, G., Guo, S., and Lu, B. (2015). PINK1 and Parkin Control Localized Translation of Respiratory Chain Component mRNAs on Mitochondria Outer Membrane. Cell Metab. *21*, 95–108.

Georgakopoulos, N.D., Wells, G., and Campanella, M. (2017). The pharmacological regulation of cellular mitophagy. Nat. Chem. Biol. *13*, 136–146.

Germain, M., Mathai, J.P., McBride, H.M., and Shore, G.C. (2005). Endoplasmic reticulum BIK initiates DRP1-regulated remodelling of mitochondrial cristae during apoptosis. EMBO J. *24*, 1546–1556.

Germain, M., Nguyen, A.P., Khacho, M., Patten, D.A., Screaton, R.A., Park, D.S., and Slack, R.S. (2013). LKB1-regulated adaptive mechanisms are essential for neuronal survival following mitochondrial dysfunction. Hum. Mol. Genet. *22*, 952–962.

Giguère, N., Burke Nanni, S., and Trudeau, L.-E. (2018). On Cell Loss and Selective Vulnerability of Neuronal Populations in Parkinson's Disease. Front. Neurol. *9*, 455.

Gladkova, C., Maslen, S.L., Skehel, J.M., and Komander, D. (2018). Mechanism of parkin activation by PINK1. Nature *559*, 410–414.

Glater, E.E., Megeath, L.J., Stowers, R.S., and Schwarz, T.L. (2006). Axonal transport of mitochondria requires milton to recruit kinesin heavy chain and is light chain independent. J. Cell Biol. *173*, 545–557.

Goldberg, M.S., Fleming, S.M., Palacino, J.J., Cepeda, C., Lam, H.A., Bhatnagar, A., Meloni, E.G., Wu, N., Ackerson, L.C., Klapstein, G.J., et al. (2003). Parkin-deficient Mice Exhibit Nigrostriatal Deficits but Not Loss of Dopaminergic Neurons. J. Biol. Chem. *278*, 43628–43635.

Gomes, L.C., Benedetto, G. Di, and Scorrano, L. (2011). During autophagy mitochondria elongate, are spared from degradation and sustain cell viability. Nat. Cell Biol. *13*, 589–598.

Göritz, C., and Frisén, J. (2012). Neural stem cells and neurogenesis in the adult. Cell Stem Cell *10*, 657–659.

Graham, V., Khudyakov, J., Ellis, P., and Pevny, L. (2003). SOX2 functions to maintain neural progenitor identity. Neuron *39*, 749–765.

Greene, A.W., Grenier, K., Aguileta, M.A., Muise, S., Farazifard, R., Haque, M.E., McBride, H.M., Park, D.S., and Fon, E.A. (2012). Mitochondrial processing peptidase regulates PINK1 processing, import and Parkin recruitment. EMBO Rep. *13*, 378–385.

Greene, J.C., Whitworth, A.J., Kuo, I., Andrews, L.A., Feany, M.B., and Pallanck, L.J. (2003). Mitochondrial pathology and apoptotic muscle degeneration in Drosophila parkin mutants. Proc. Natl. Acad. Sci. *100*, 4078–4083.

Guo, X., Macleod, G.T., Wellington, A., Hu, F., Panchumarthi, S., Schoenfield, M., Marin, L., Charlton, M.P., Atwood, H.L., and Zinsmaier, K.E. (2005). The GTPase dMiro Is Required for Axonal Transport of Mitochondria to Drosophila Synapses. Neuron *47*, 379–393.

Haque, M.E., Thomas, K.J., D'Souza, C., Callaghan, S., Kitada, T., Slack, R.S., Fraser, P., Cookson, M.R., Tandon, A., and Park, D.S. (2008). Cytoplasmic Pink1 activity protects neurons

from dopaminergic neurotoxin MPTP. Proc. Natl. Acad. Sci. *105*, 1716–1721.

Haque, M.E., Mount, M.P., Safarpour, F., Abdel-Messih, E., Callaghan, S., Mazerolle, C., Kitada, T., Slack, R.S., Wallace, V., Shen, J., et al. (2012). Inactivation of *Pink1* Gene *in Vivo* Sensitizes Dopamine-producing Neurons to 1-Methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) and Can Be Rescued by Autosomal Recessive Parkinson Disease Genes, *Parkin* or *DJ-1*. J. Biol. Chem. *287*, 23162–23170.

Heo, J.-M., Ordureau, A., Paulo, J.A., Rinehart, J., and Harper, J.W. (2015). The PINK1-PARKIN Mitochondrial Ubiquitylation Pathway Drives a Program of OPTN/NDP52 Recruitment and TBK1 Activation to Promote Mitophagy. Mol. Cell *60*, 7–20.

Höglinger, G.U., Rizk, P., Muriel, M.P., Duyckaerts, C., Oertel, W.H., Caille, I., and Hirsch, E.C. (2004). Dopamine depletion impairs precursor cell proliferation in Parkinson disease. Nat. Neurosci. *7*, 726–735.

Hristova, V.A., Beasley, S.A., Rylett, R.J., and Shaw, G.S. (2009). Identification of a novel Zn2+-binding domain in the autosomal recessive juvenile Parkinson-related E3 ligase parkin. J. Biol. Chem. *284*, 14978–14986.

Hsieh, J. (2012). Orchestrating transcriptional control of adult neurogenesis. Genes Dev. *26*, 1010–1021.

Hsieh, J., and Zhao, X. (2016). Genetics and epigenetics in adult neurogenesis. Cold Spring Harb. Perspect. Biol. *8*, a018911.

Hsieh, C.-H., Shaltouki, A., Gonzalez, A.E., Bettencourt da Cruz, A., Burbulla, L.F., St Lawrence, E., Schüle, B., Krainc, D., Palmer, T.D., and Wang, X. (2016). Functional Impairment in Miro Degradation and Mitophagy Is a Shared Feature in Familial and Sporadic Parkinson's Disease. Cell Stem Cell *19*, 709–724.

Hwang, S., Disatnik, M.-H., and Mochly-Rosen, D. (2015). Impaired GAPDH-induced mitophagy contributes to the pathology of Huntington's disease. EMBO Mol. Med. *7*, 1307–1326.

Imai, S., and Guarente, L. (2014). NAD+ and sirtuins in aging and disease. Trends Cell Biol. *24*, 464–471.

Imayoshi, I., Sakamoto, M., and Kageyama, R. (2011). Genetic Methods to Identify and Manipulate Newly Born Neurons in the Adult Brain. Front. Neurosci. *5*, 64.

Itier, J.-M., Ibanez, P., Mena, M.A., Abbas, N., Cohen-Salmon, C., Bohme, G.A., Laville, M., Pratt, J., Corti, O., Pradier, L., et al. (2003). Parkin gene inactivation alters behaviour and dopamine neurotransmission in the mouse. Hum. Mol. Genet. *12*, 2277–2291.

Ito, K., and Suda, T. (2014). Metabolic requirements for the maintenance of self-renewing stem cells. Nat. Rev. Mol. Cell Biol. *15*, 243–256.

Javitch, J.A., D'Amato, R.J., Strittmatter, S.M., and Snyder, S.H. (1985). Parkinsonism-inducing neurotoxin, N-methyl-4-phenyl-1,2,3,6 -tetrahydropyridine: uptake of the metabolite N-methyl-4-phenylpyridine by dopamine neurons explains selective toxicity. Proc. Natl. Acad. Sci. U. S. A. *82*, 2173–2177.

Jin, S.M., Lazarou, M., Wang, C., Kane, L.A., Narendra, D.P., and Youle, R.J. (2010). Mitochondrial membrane potential regulates PINK1 import and proteolytic destabilization by PARL. J. Cell Biol. *191*, 933–942.

Johansson, C.B., Svensson, M., Wallstedt, L., Janson, A.M., and Frisén, J. (1999). Neural Stem Cells in the Adult Human Brain. Exp. Cell Res. *253*, 733–736.

St. John, J.C., Ramalho-Santos, J., Gray, H.L., Petrosko, P., Rawe, V.Y., Navara, C.S., Simerly, C.R., and Schatten, G.P. (2005). The Expression of Mitochondrial DNA Transcription Factors during Early Cardiomyocyte *In Vitro* Differentiation from Human Embryonic Stem Cells. Cloning Stem Cells *7*, 141–153.

Jornayvaz, F.R., and Shulman, G.I. (2010). Regulation of mitochondrial biogenesis. Essays Biochem. *47*, 69–84.

Kaelin, W.G., McKnight, S.L., and McKnight, S.L. (2013). Influence of metabolism on epigenetics and disease. Cell *153*, 56–69.

Kalia, L. V., and Lang, A.E. (2015). Parkinson's disease. Lancet *386*, 896–912.

Kane, L.A., Lazarou, M., Fogel, A.I., Li, Y., Yamano, K., Sarraf, S.A., Banerjee, S., and Youle, R.J. (2014). PINK1 phosphorylates ubiquitin to activate Parkin E3 ubiquitin ligase activity. J. Cell Biol. *205*, 143–153.

Karbowski, M., and Youle, R.J. (2011). Regulating mitochondrial outer membrane proteins by ubiquitination and proteasomal degradation. Curr. Opin. Cell Biol. *23*, 476–482.

Kazanis, I., Lathia, J.D., Vadakkan, T.J., Raborn, E., Wan, R., Mughal, M.R., Eckley, D.M., Sasaki, T., Patton, B., Mattson, M.P., et al. (2010). Quiescence and activation of stem and precursor cell populations in the subependymal zone of the mammalian brain are associated with distinct cellular and extracellular matrix signals. J. Neurosci. *30*, 9771–9781.

Kazlauskaite, A., Kondapalli, C., Gourlay, R., Campbell, D.G., Ritorto, M.S., Hofmann, K., Alessi, D.R., Knebel, A., Trost, M., and Muqit, M.M.K. (2014). Parkin is activated by PINK1-dependent phosphorylation of ubiquitin at Ser65. Biochem. J. *460*, 127–139.

Keil, U., Scherping, I., Hauptmann, S., Schuessel, K., Eckert, A., and Müller, W.E. (2006). Piracetam improves mitochondrial dysfunction following oxidative stress. Br. J. Pharmacol. *147*, 199–208.

Kempermann, G., Jessberger, S., Steiner, B., and Kronenberg, G. (2004). Milestones of neuronal development in the adult hippocampus. Trends Neurosci. *27*, 447–452.

Kempermann, G., Gage, F.H., Aigner, L., Song, H., Curtis, M.A., Thuret, S., Kuhn, H.G., Jessberger, S., Frankland, P.W., Cameron, H.A., et al. (2018). Human Adult Neurogenesis: Evidence and Remaining Questions. Cell Stem Cell *23*, 25–30.

Kessova, I.G., and Cederbaum, A.I. (2007). Mitochondrial alterations in livers of Sod1-/- mice fed alcohol. Free Radic. Biol. Med. *42*, 1470–1480.

Khacho, M., and Slack, R.S. (2017). Mitochondrial activity in the regulation of stem cell self-renewal and differentiation. Curr. Opin. Cell Biol. *49*, 1–8.

Khacho, M., Tarabay, M., Patten, D., Khacho, P., MacLaurin, J.G., Guadagno, J., Bergeron, R., Cregan, S.P., Harper, M.-E., Park, D.S., et al. (2014). Acidosis overrides oxygen deprivation to maintain mitochondrial function and cell survival. Nat. Commun. *5*, 3550.

Khacho, M., Clark, A., Svoboda, D.S., Azzi, J., MacLaurin, J.G., Meghaizel, C., Sesaki, H., Lagace, D.C., Germain, M., Harper, M.E., et al. (2016). Mitochondrial Dynamics Impacts Stem Cell Identity and Fate Decisions by Regulating a Nuclear Transcriptional Program. Cell Stem Cell *19*, 232–247.

Khacho, M., Clark, A., Svoboda, D.S., MacLaurin, J.G., Lagace, D.C., Park, D.S., and Slack, R.S. (2017). Mitochondrial dysfunction underlies cognitive defects as a result of neural stem cell depletion and impaired neurogenesis. Hum. Mol. Genet. *26*, 3327–3341.

Khacho, M., Harris, R., and Slack, R.S. (2019). Mitochondria as central regulators of neural stem cell fate and cognitive function. Nat. Rev. Neurosci. *20*, 34–48.

Kim, W.S., Kågedal, K., and Halliday, G.M. (2014). Alpha-synuclein biology in Lewy body diseases. Alzheimers. Res. Ther. *6*, 73.

Kitada, T., Asakawa, S., Hattori, N., Matsumine, H., Yamamura, Y., Minoshima, S., Yokochi, M., Mizuno, Y., and Shimizu, N. (1998). Mutations in the parkin gene cause autosomal recessive juvenile parkinsonism. Nature *392*, 605–608.

Kitada, T., Pisani, A., Porter, D.R., Yamaguchi, H., Tscherter, A., Martella, G., Bonsi, P., Zhang, C., Pothos, E.N., and Shen, J. (2007). Impaired dopamine release and synaptic plasticity in the striatum of PINK1-deficient mice. Proc. Natl. Acad. Sci. *104*, 11441–11446.

Kitada, T., Tong, Y., Gautier, C.A., and Shen, J. (2009). Absence of nigral degeneration in aged parkin/DJ-1/PINK1 triple knockout mice. J. Neurochem. *111*, 696–702.

Klein, J.A., Longo-Guess, C.M., Rossmann, M.P., Seburn, K.L., Hurd, R.E., Frankel, W.N., Bronson, R.T., and Ackerman, S.L. (2002). The harlequin mouse mutation downregulates apoptosis-inducing factor. Nature *419*, 367–374.

Knoth, R., Singec, I., Ditter, M., Pantazis, G., Capetian, P., Meyer, R.P., Horvat, V., Volk, B., and Kempermann, G. (2010). Murine Features of Neurogenesis in the Human Hippocampus across the Lifespan from 0 to 100 Years. PLoS One *5*, e8809.

Ko, H.-G., Jang, D.-J., Son, J., Kwak, C., Choi, J.-H., Ji, Y.-H., Lee, Y.-S., Son, H., and Kaang, B.-K. (2009). Effect of ablated hippocampal neurogenesis on the formation and extinction of contextual fear memory. Mol. Brain *2*, 1.

Kohl, Z., Winner, B., Ubhi, K., Rockenstein, E., Mante, M., Münch, M., Barlow, C., Carter, T., Masliah, E., and Winkler, J. (2012). Fluoxetine rescues impaired hippocampal neurogenesis in a transgenic A53T synuclein mouse model. Eur. J. Neurosci. *35*, 10–19.

Koizumi, H., Higginbotham, H., Poon, T., Tanaka, T., Brinkman, B.C., and Gleeson, J.G. (2006). Doublecortin maintains bipolar shape and nuclear translocation during migration in the adult forebrain. Nat. Neurosci. *9*, 779–786.

Kondapalli, C., Kazlauskaite, A., Zhang, N., Woodroof, H.I., Campbell, D.G., Gourlay, R., Burchell, L., Walden, H., MacArtney, T.J., Deak, M., et al. (2012). PINK1 is activated by

mitochondrial membrane potential depolarization and stimulates Parkin E3 ligase activity by phosphorylating Serine 65. Open Biol. *2*, 120080.

Kondoh, H., Lleonart, M.E., Nakashima, Y., Yokode, M., Tanaka, M., Bernard, D., Gil, J., and Beach, D. (2007). A High Glycolytic Flux Supports the Proliferative Potential of Murine Embryonic Stem Cells. Antioxid. Redox Signal. *9*, 293–299.

Koyano, F., Okatsu, K., Kosako, H., Tamura, Y., Go, E., Kimura, M., Kimura, Y., Tsuchiya, H., Yoshihara, H., Hirokawa, T., et al. (2014). Ubiquitin is phosphorylated by PINK1 to activate parkin. Nature *510*, 162–166.

Kraytsberg, Y., Kudryavtseva, E., McKee, A.C., Geula, C., Kowall, N.W., and Khrapko, K. (2006). Mitochondrial DNA deletions are abundant and cause functional impairment in aged human substantia nigra neurons. Nat. Genet. *38*, 518–520.

Kronenberg, G., Reuter, K., Steiner, B., Brandt, M.D., Jessberger, S., Yamaguchi, M., and Kempermann, G. (2003). Subpopulations of proliferating cells of the adult hippocampus respond differently to physiologic neurogenic stimuli. J. Comp. Neurol. *467*, 455–463.

Kujoth, G.C., Hiona, A., Pugh, T.D., Someya, S., Panzer, K., Wohlgemuth, S.E., Hofer, T., Seo, A.Y., Sullivan, R., Jobling, W.A., et al. (2005). Mitochondrial DNA Mutations, Oxidative Stress, and Apoptosis in Mammalian Aging. Science *309*, 481–484.

Kumar, A., Aguirre, J.D., Condos, T.E.C., Martinez-Torres, R.J., Chaugule, V.K., Toth, R., Sundaramoorthy, R., Mercier, P., Knebel, A., Spratt, D.E., et al. (2015). Disruption of the autoinhibited state primes the E3 ligase parkin for activation and catalysis. EMBO J. *34*, 2506–2521.

Van Laar, V.S., Arnold, B., Cassady, S.J., Chu, C.T., Burton, E.A., and Berman, S.B. (2011). Bioenergetics of neurons inhibit the translocation response of Parkin following rapid mitochondrial depolarization. Hum. Mol. Genet. *20*, 927–940.

Lagace, D.C., Whitman, M.C., Noonan, M.A., Ables, J.L., DeCarolis, N.A., Arguello, A.A., Donovan, M.H., Fischer, S.J., Farnbauch, L.A., Beech, R.D., et al. (2007). Dynamic contribution of nestin-expressing stem cells to adult neurogenesis. J. Neurosci. *27*, 12623–12629.

Langston, J.W., Ballard, P., Tetrud, J.W., and Irwin, I. (1983). Chronic Parkinsonism in humans due to a product of meperidine-analog synthesis. Science *219*, 979–980.

Lazarou, M., Jin, S.M., Kane, L.A., and Youle, R.J. (2012). Role of PINK1 Binding to the TOM Complex and Alternate Intracellular Membranes in Recruitment and Activation of the E3 Ligase Parkin. Dev. Cell *22*, 320–333.

Lazarou, M., Sliter, D.A., Kane, L.A., Sarraf, S.A., Wang, C., Burman, J.L., Sideris, D.P., Fogel, A.I., and Youle, R.J. (2015). The ubiquitin kinase PINK1 recruits autophagy receptors to induce mitophagy. Nature *524*, 309–314.

Lebovitz, R.M., Zhang, H., Vogel, H., Cartwright, J., Dionne, L., Lu, N., Huang, S., and Matzuk, M.M. (1996). Neurodegeneration, myocardial injury, and perinatal death in mitochondrial superoxide dismutase-deficient mice. Proc. Natl. Acad. Sci. U. S. A. *93*, 9782–9787.

Lee, H., and Thuret, S. (2018). Adult Human Hippocampal Neurogenesis: Controversy and

Evidence. Trends Mol. Med. *24*, 521–522.

Lee, Y., Stevens, D.A., Kang, S.-U., Jiang, H., Lee, Y.-I., Ko, H.S., Scarffe, L.A., Umanah, G.E., Kang, H., Ham, S., et al. (2017). PINK1 Primes Parkin-Mediated Ubiquitination of PARIS in Dopaminergic Neuronal Survival. Cell Rep. *18*, 918–932.

Lendahl, U., Zimmerman, L.B., and McKay, R.D. (1990). CNS stem cells express a new class of intermediate filament protein. Cell *60*, 585–595.

Lesnik, C., Golani-Armon, A., and Arava, Y. (2015). Localized translation near the mitochondrial outer membrane: An update. RNA Biol. *12*, 801–809.

Leuner, K., Kurz, C., Guidetti, G., Orgogozo, J.M., and Müller, W.E. (2010). Improved mitochondrial function in brain aging and Alzheimer disease - the new mechanism of action of the old metabolic enhancer piracetam. Front. Neurosci. *4*, 44.

Li, J., Qi, W., Chen, G., Feng, D., Liu, J., Ma, B., Zhou, C., Mu, C., Zhang, W., Chen, Q., et al. (2015). Mitochondrial outer-membrane E3 ligase MUL1 ubiquitinates ULK1 and regulates selenite-induced mitophagy. Autophagy *11*, 1216–1229.

Lim, K.-L., Ng, X.-H., Grace, L.G.-Y., and Yao, T.-P. (2012). Mitochondrial dynamics and Parkinson's disease: focus on parkin. Antioxid. Redox Signal. *16*, 935–949.

Lin, W., and Kang, U.J. (2008). Characterization of PINK1 processing, stability, and subcellular localization. J. Neurochem. *106*, 464–474.

Lin, J., Puigserver, P., Donovan, J., Tarr, P., and Spiegelman, B.M. (2002). Peroxisome Proliferator-activated Receptor γ Coactivator 1β (PGC-1β), A Novel PGC-1-related Transcription Coactivator Associated with Host Cell Factor. J. Biol. Chem. *277*, 1645–1648.

Liu, L., Feng, D., Chen, G., Chen, M., Zheng, Q., Song, P., Ma, Q., Zhu, C., Wang, R., Qi, W., et al. (2012a). Mitochondrial outer-membrane protein FUNDC1 mediates hypoxia-induced mitophagy in mammalian cells. Nat. Cell Biol. *14*, 177–185.

Liu, S., Sawada, T., Lee, S., Yu, W., Silverio, G., Alapatt, P., Millan, I., Shen, A., Saxton, W., Kanao, T., et al. (2012b). Parkinson's Disease–Associated Kinase PINK1 Regulates Miro Protein Level and Axonal Transport of Mitochondria. PLoS Genet. *8*, e1002537.

Lois, C., and Alvarez-Buylla, A. (1993). Proliferating subventricular zone cells in the adult mammalian forebrain can differentiate into neurons and glia. Proc. Natl. Acad. Sci. U. S. A. *90*, 2074–2077.

Lois, C., and Alvarez-Buylla, A. (1994). Long-distance neuronal migration in the adult mammalian brain. Science *264*, 1145–1148.

Lois, C., García-Verdugo, J.M., and Alvarez-Buylla, A. (1996). Chain migration of neuronal precursors. Science *271*, 978–981.

Losón, O.C., Song, Z., Chen, H., and Chan, D.C. (2013). Fis1, Mff, MiD49, and MiD51 mediate Drp1 recruitment in mitochondrial fission. Mol. Biol. Cell *24*, 659–667.

Lugert, S., Basak, O., Knuckles, P., Haussler, U., Fabel, K., Götz, M., Haas, C.A., Kempermann, G., Taylor, V., and Giachino, C. (2010). Quiescent and Active Hippocampal Neural Stem Cells

with Distinct Morphologies Respond Selectively to Physiological and Pathological Stimuli and Aging. Cell Stem Cell *6*, 445–456.

Lugert, S., Vogt, M., Tchorz, J.S., Müller, M., Giachino, C., and Taylor, V. (2012). Homeostatic neurogenesis in the adult hippocampus does not involve amplification of Ascl1high intermediate progenitors. Nat. Commun. *3*, 670.

Lutz, A.K., Exner, N., Fett, M.E., Schlehe, J.S., Kloos, K., Lämmermann, K., Brunner, B., Kurz-Drexler, A., Vogel, F., Reichert, A.S., et al. (2009). Loss of Parkin or PINK1 Function Increases Drp1-dependent Mitochondrial Fragmentation. J. Biol. Chem. *284*, 22938–22951.

Lyublinskaya, O.G., Borisov, Y.G., Pugovkina, N.A., Smirnova, I.S., Obidina, J. V., Ivanova, J.S., Zenin, V. V., Shatrova, A.N., Borodkina, A. V., Aksenov, N.D., et al. (2015). Reactive oxygen species are required for human mesenchymal stem cells to initiate proliferation after the quiescence exit. Oxid. Med. Cell. Longev. *2015*, 502105.

MacAskill, A.F., Rinholm, J.E., Twelvetrees, A.E., Arancibia-Carcamo, I.L., Muir, J., Fransson, A., Aspenstrom, P., Attwell, D., and Kittler, J.T. (2009). Miro1 Is a Calcium Sensor for Glutamate Receptor-Dependent Localization of Mitochondria at Synapses. Neuron *61*, 541–555.

Madeo, G., Schirinzi, T., Martella, G., Latagliata, E.C., Puglisi, F., Shen, J., Valente, E.M., Federici, M., Mercuri, N.B., Puglisi-Allegra, S., et al. (2014). PINK1 heterozygous mutations induce subtle alterations in dopamine-dependent synaptic plasticity. Mov. Disord. *29*, 41–53.

Magrinelli, F., Picelli, A., Tocco, P., Federico, A., Roncari, L., Smania, N., Zanette, G., and Tamburin, S. (2016). Pathophysiology of Motor Dysfunction in Parkinson's Disease as the Rationale for Drug Treatment and Rehabilitation. Parkinsons. Dis. *2016*, 9832839.

Mandal, S., Lindgren, A.G., Srivastava, A.S., Clark, A.T., and Banerjee, U. (2011). Mitochondrial function controls proliferation and early differentiation potential of embryonic stem cells. Stem Cells *29*, 486–495.

Marongiu, R., Spencer, B., Crews, L., Adame, A., Patrick, C., Trejo, M., Dallapiccola, B., Valente, E.M., and Masliah, E. (2009). Mutant Pink1 induces mitochondrial dysfunction in a neuronal cell model of Parkinson's disease by disturbing calcium flux. J. Neurochem. *108*, 1561–1574.

Marxreiter, F., Nuber, S., Kandasamy, M., Klucken, J., Aigner, R., Burgmayer, R., Couillard-Despres, S., Riess, O., Winkler, J., and Winner, B. (2009). Changes in adult olfactory bulb neurogenesis in mice expressing the A30P mutant form of alpha-synuclein. Eur. J. Neurosci. *29*, 879–890.

Marxreiter, F., Ettle, B., May, V.E.L., Esmer, H., Patrick, C., Kragh, C.L., Klucken, J., Winner, B., Riess, O., Winkler, J., et al. (2013a). Glial A30P alpha-synuclein pathology segregates neurogenesis from anxiety-related behavior in conditional transgenic mice. Neurobiol. Dis. *59*, 38–51.

Marxreiter, F., Regensburger, M., and Winkler, J. (2013b). Adult neurogenesis in Parkinson's disease. Cell. Mol. Life Sci. *70*, 459–473.

Matheoud, D., Sugiura, A., Bellemare-Pelletier, A., Laplante, A., Rondeau, C., Chemali, M., Fazel, A., Bergeron, J.J., Trudeau, L.-E., Burelle, Y., et al. (2016). Parkinson's Disease-Related

Proteins PINK1 and Parkin Repress Mitochondrial Antigen Presentation. Cell *166*, 314–327.

Mathews, K.J., Allen, K.M., Boerrigter, D., Ball, H., Shannon Weickert, C., and Double, K.L. (2017). Evidence for reduced neurogenesis in the aging human hippocampus despite stable stem cell markers. Aging Cell *16*, 1195–1199.

Matsuda, N., Sato, S., Shiba, K., Okatsu, K., Saisho, K., Gautier, C.A., Sou, Y.-S., Saiki, S., Kawajiri, S., Sato, F., et al. (2010). PINK1 stabilized by mitochondrial depolarization recruits Parkin to damaged mitochondria and activates latent Parkin for mitophagy. J. Cell Biol. *189*, 211–221.

Matsumine, H., Saito, M., Shimoda-Matsubayashi, S., Tanaka, H., Ishikawa, A., Nakagawa-Hattori, Y., Yokochi, M., Kobayashi, T., Igarashi, S., Takano, H., et al. (1997). Localization of a gene for an autosomal recessive form of juvenile Parkinsonism to chromosome 6q25.2-27. Am. J. Hum. Genet. *60*, 588–596.

May, V.E.L., Nuber, S., Marxreiter, F., Riess, O., Winner, B., and Winkler, J. (2012). Impaired olfactory bulb neurogenesis depends on the presence of human wild-type alpha-synuclein. Neuroscience *222*, 343–355.

McLelland, G.-L., Soubannier, V., Chen, C.X., McBride, H.M., and Fon, E.A. (2014). Parkin and PINK1 function in a vesicular trafficking pathway regulating mitochondrial quality control. EMBO J. *33*, 282–295.

Mcwilliams, T.G., Prescott, A.R., Montava-Garriga, L., Muqit, M.M.K., Brooks, S.P., and Ganley, I.G. (2018). Basal Mitophagy Occurs Independently of PINK1 in Mouse Tissues of High Metabolic Demand. Cell Metab. *27*, 439-449.e5.

McWilliams, T.G., and Muqit, M.M. (2017). PINK1 and Parkin: emerging themes in mitochondrial homeostasis. Curr. Opin. Cell Biol. *45*, 83–91.

McWilliams, T.G., Prescott, A.R., Allen, G.F.G., Tamjar, J., Munson, M.J., Thomson, C., Muqit, M.M.K., and Ganley, I.G. (2016). mito-QC illuminates mitophagy and mitochondrial architecture in vivo. J. Cell Biol. *214*, 333–345.

Meeusen, S., DeVay, R., Block, J., Cassidy-Stone, A., Wayson, S., McCaffery, J.M., and Nunnari, J. (2006). Mitochondrial Inner-Membrane Fusion and Crista Maintenance Requires the Dynamin-Related GTPase Mgm1. Cell *127*, 383–395.

Meirhaeghe, A., Crowley, V., Lenaghan, C., Lelliott, C., Green, K., Stewart, A., Hart, K., Schinner, S., Sethi, J.K., Yeo, G., et al. (2003). Characterization of the human, mouse and rat PGC1 beta (peroxisome-proliferator-activated receptor-gamma co-activator 1 beta) gene in vitro and in vivo. Biochem. J. *373*, 155–165.

Meissner, C., Lorenz, H., Weihofen, A., Selkoe, D.J., and Lemberg, M.K. (2011). The mitochondrial intramembrane protease PARL cleaves human Pink1 to regulate Pink1 trafficking. J. Neurochem. *117*, 856–867.

Ming, G.-L., and Song, H. (2011). Adult neurogenesis in the mammalian brain: significant answers and significant questions. Neuron *70*, 687–702.

Mirzadeh, Z., Merkle, F.T., Soriano-Navarro, M., Garcia-Verdugo, J.M., and Alvarez-Buylla, A.

(2008). Neural Stem Cells Confer Unique Pinwheel Architecture to the Ventricular Surface in Neurogenic Regions of the Adult Brain. Cell Stem Cell *3*, 265–278.

Montessuit, S., Somasekharan, S.P., Terrones, O., Lucken-Ardjomande, S., Herzig, S., Schwarzenbacher, R., Manstein, D.J., Bossy-Wetzel, E., Basañez, G., Meda, P., et al. (2010). Membrane Remodeling Induced by the Dynamin-Related Protein Drp1 Stimulates Bax Oligomerization. Cell *142*, 889–901.

Mouton-Liger, F., Jacoupy, M., Corvol, J.-C., and Corti, O. (2017). PINK1/Parkin-Dependent Mitochondrial Surveillance: From Pleiotropy to Parkinson's Disease. Front. Mol. Neurosci. *10*, 120.

Nakashiba, T., Cushman, J.D., Pelkey, K.A., Renaudineau, S., Buhl, D.L., McHugh, T.J., Barrera, V.R., Chittajallu, R., Iwamoto, K.S., McBain, C.J., et al. (2012). Young Dentate Granule Cells Mediate Pattern Separation, whereas Old Granule Cells Facilitate Pattern Completion. Cell *149*, 188–201.

Narendra, D., Tanaka, A., Suen, D.-F., and Youle, R.J. (2008). Parkin is recruited selectively to impaired mitochondria and promotes their autophagy. J. Cell Biol. *183*, 795–803.

Narendra, D.P., Jin, S.M., Tanaka, A., Suen, D.-F., Gautier, C.A., Shen, J., Cookson, M.R., and Youle, R.J. (2010). PINK1 Is Selectively Stabilized on Impaired Mitochondria to Activate Parkin. PLoS Biol. *8*, e1000298.

Nicklas, W.J., Vyas, I., and Heikkila, R.E. (1985). Inhibition of NADH-linked oxidation in brain mitochondria by 1-methyl-4-phenyl-pyridine, a metabolite of the neurotoxin, 1-methyl-4-phenyl-1,2,5,6-tetrahydropyridine. Life Sci. *36*, 2503–2508.

Nicola, Z., Fabel, K., and Kempermann, G. (2015). Development of the adult neurogenic niche in the hippocampus of mice. Front. Neuroanat. *9*, 53.

Novak, I., Kirkin, V., McEwan, D.G., Zhang, J., Wild, P., Rozenknop, A., Rogov, V., Löhr, F., Popovic, D., Occhipinti, A., et al. (2010). Nix is a selective autophagy receptor for mitochondrial clearance. EMBO Rep. *11*, 45–51.

Nuber, S., Petrasch-Parwez, E., Winner, B., Winkler, J., Hörsten, S. von, Schmidt, T., Boy, J., Kuhn, M., Nguyen, H.P., Teismann, P., et al. (2008). Neurodegeneration and Motor Dysfunction in a Conditional Model of Parkinson's Disease. J. Neurosci. *28*, 2471–2484.

O'Keeffe, G.C., Tyers, P., Aarsland, D., Dalley, J.W., Barker, R.A., and Caldwell, M.A. (2009). Dopamine-induced proliferation of adult neural precursor cells in the mammalian subventricular zone is mediated through EGF. Proc. Natl. Acad. Sci. U. S. A. *106*, 8754–8759.

Obernier, K., and Alvarez-Buylla, A. (2019). Neural stem cells: origin, heterogeneity and regulation in the adult mammalian brain. Development *146*, dev156059.

Okatsu, K., Oka, T., Iguchi, M., Imamura, K., Kosako, H., Tani, N., Kimura, M., Go, E., Koyano, F., Funayama, M., et al. (2012). PINK1 autophosphorylation upon membrane potential dissipation is essential for Parkin recruitment to damaged mitochondria. Nat. Commun. *3*, 1016.

Okatsu, K., Uno, M., Koyano, F., Go, E., Kimura, M., Oka, T., Tanaka, K., and Matsuda, N. (2013). A Dimeric PINK1-containing Complex on Depolarized Mitochondria Stimulates Parkin

Recruitment. J. Biol. Chem. *288*, 36372–36384.

Okatsu, K., Kimura, M., Oka, T., Tanaka, K., and Matsuda, N. (2015). Unconventional PINK1 localization to the outer membrane of depolarized mitochondria drives Parkin recruitment. J. Cell Sci. *128*, 964–978.

Oliveira, S.L.B., Pillat, M.M., Cheffer, A., Lameu, C., Schwindt, T.T., and Ulrich, H. (2013). Functions of neurotrophins and growth factors in neurogenesis and brain repair. Cytom. Part A *83 A*, 76–89.

Ordureau, A., Sarraf, S.A., Duda, D.M., Heo, J.-M., Jedrychowski, M.P., Sviderskiy, V.O., Olszewski, J.L., Koerber, J.T., Xie, T., Beausoleil, S.A., et al. (2014). Quantitative Proteomics Reveal a Feedforward Mechanism for Mitochondrial PARKIN Translocation and Ubiquitin Chain Synthesis. Mol. Cell *56*, 360–375.

Owusu-Ansah, E., and Banerjee, U. (2009). Reactive oxygen species prime Drosophila haematopoietic progenitors for differentiation. Nature *461*, 537–541.

Paisán-Ruíz, C., Jain, S., Evans, E.W., Gilks, W.P., Simón, J., van der Brug, M., de Munain, A.L., Aparicio, S., Gil, A.M., Khan, N., et al. (2004). Cloning of the Gene Containing Mutations that Cause PARK8-Linked Parkinson's Disease. Neuron *44*, 595–600.

Palacino, J.J., Sagi, D., Goldberg, M.S., Krauss, S., Motz, C., Wacker, M., Klose, J., and Shen, J. (2004). Mitochondrial Dysfunction and Oxidative Damage in *parkin* -deficient Mice. J. Biol. Chem. *279*, 18614–18622.

Palmer, T.D., Takahashi, J., and Gage, F.H. (1997). The Adult Rat Hippocampus Contains Primordial Neural Stem Cells. Mol. Cell. Neurosci. *8*, 389–404.

Panopoulos, A.D., Yanes, O., Ruiz, S., Kida, Y.S., Diep, D., Tautenhahn, R., Herrerías, A., Batchelder, E.M., Plongthongkum, N., Lutz, M., et al. (2012). The metabolome of induced pluripotent stem cells reveals metabolic changes occurring in somatic cell reprogramming. Cell Res. *22*, 168–177.

Park, J.-S., Davis, R.L., and Sue, C.M. (2018). Mitochondrial Dysfunction in Parkinson's Disease: New Mechanistic Insights and Therapeutic Perspectives. Curr. Neurol. Neurosci. Rep. *18*, 21.

Park, J., Lee, S.B., Lee, S., Kim, Y., Song, S., Kim, S., Bae, E., Kim, J., Shong, M., Kim, J.-M., et al. (2006). Mitochondrial dysfunction in Drosophila PINK1 mutants is complemented by parkin. Nature *441*, 1157–1161.

Parker, W.D., Boyson, S.J., and Parks, J.K. (1989). Abnormalities of the electron transport chain in idiopathic parkinson's disease. Ann. Neurol. *26*, 719–723.

Parker, W.D., Parks, J.K., Swerdlow, R.H., and Swerdlow, R.H. (2008). Complex I deficiency in Parkinson's disease frontal cortex. Brain Res. *1189*, 215–218.

Pastrana, E., Silva-Vargas, V., and Doetsch, F. (2011). Eyes wide open: a critical review of sphere-formation as an assay for stem cells. Cell Stem Cell *8*, 486–498.

Patten, D.A., Wong, J., Khacho, M., Soubannier, V., Mailloux, R.J., Pilon-Larose, K.,

MacLaurin, J.G., Park, D.S., McBride, H.M., Trinkle-Mulcahy, L., et al. (2014). OPA1-dependent cristae modulation is essential for cellular adaptation to metabolic demand. EMBO J. *33*, 2676–2691.

Paul, M.K., Bisht, B., Darmawan, D.O., Chiou, R., Ha, V.L., Wallace, W.D., Chon, A.T., Hegab, A.E., Grogan, T., Elashoff, D.A., et al. (2014). Dynamic Changes in Intracellular ROS Levels Regulate Airway Basal Stem Cell Homeostasis through Nrf2-Dependent Notch Signaling. Cell Stem Cell *15*, 199–214.

Pereira, S.L., Grãos, M., Rodrigues, A.S., Anjo, S.I., Carvalho, R.A., Oliveira, P.J., Arenas, E., and Ramalho-Santos, J. (2013). Inhibition of Mitochondrial Complex III Blocks Neuronal Differentiation and Maintains Embryonic Stem Cell Pluripotency. PLoS One *8*, e82095.

Perez, F.A., and Palmiter, R.D. (2005). Parkin-deficient mice are not a robust model of parkinsonism. Proc. Natl. Acad. Sci. *102*, 2174–2179.

Periquet, M., Corti, O., Jacquier, S., and Brice, A. (2005). Proteomic analysis of parkin knockout mice: alterations in energy metabolism, protein handling and synaptic function. J. Neurochem. *95*, 1259–1276.

Pesah, Y., Pham, T., Burgess, H., Middlebrooks, B., Verstreken, P., Zhou, Y., Harding, M., Bellen, H., and Mardon, G. (2004). Drosophila parkin mutants have decreased mass and cell size and increased sensitivity to oxygen radical stress. Development *131*, 2183–2194.

Petit, A., Kawarai, T., Paitel, E., Sanjo, N., Maj, M., Scheid, M., Chen, F., Gu, Y., Hasegawa, H., Salehi-Rad, S., et al. (2005). Wild-type PINK1 Prevents Basal and Induced Neuronal Apoptosis, a Protective Effect Abrogated by Parkinson Disease-related Mutations. J. Biol. Chem. *280*, 34025–34032.

Piccoli, C., Ria, R., Scrima, R., Cela, O., D'Aprile, A., Boffoli, D., Falzetti, F., Tabilio, A., and Capitanio, N. (2005). Characterization of Mitochondrial and Extra-mitochondrial Oxygen Consuming Reactions in Human Hematopoietic Stem Cells. J. Biol. Chem. *280*, 26467–26476.

Pickrell, A.M., and Youle, R.J. (2015). The Roles of PINK1, Parkin, and Mitochondrial Fidelity in Parkinson's Disease. Neuron *85*, 257–273.

Pickrell, A.M., Huang, C.-H., Kennedy, S.R., Ordureau, A., Sideris, D.P., Hoekstra, J.G., Harper, J.W., and Youle, R.J. (2015). Endogenous Parkin Preserves Dopaminergic Substantia Nigral Neurons following Mitochondrial DNA Mutagenic Stress. Neuron *87*, 371–381.

Polymeropoulos, M.H., Lavedan, C., Leroy, E., Ide, S.E., Dehejia, A., Dutra, A., Pike, B., Root, H., Rubenstein, J., Boyer, R., et al. (1997). Mutation in the alpha-synuclein gene identified in families with Parkinson's disease. Science *276*, 2045–2047.

Poole, A.C., Thomas, R.E., Andrews, L.A., McBride, H.M., Whitworth, A.J., and Pallanck, L.J. (2008). The PINK1/Parkin pathway regulates mitochondrial morphology. Proc. Natl. Acad. Sci. *105*, 1638–1643.

Poole, A.C., Thomas, R.E., Yu, S., Vincow, E.S., and Pallanck, L. (2010). The Mitochondrial Fusion-Promoting Factor Mitofusin Is a Substrate of the PINK1/Parkin Pathway. PLoS One *5*, e10054.

Pridgeon, J.W., Olzmann, J.A., Chin, L.-S., and Li, L. (2007). PINK1 protects against oxidative stress by phosphorylating mitochondrial chaperone TRAP1. PLoS Biol. *5*, e172.

Prigione, A., Fauler, B., Lurz, R., Lehrach, H., and Adjaye, J. (2010). The Senescence-Related Mitochondrial/Oxidative Stress Pathway is Repressed in Human Induced Pluripotent Stem Cells. Stem Cells *28*, 721–733.

Pryde, K.R., Smith, H.L., Chau, K.-Y., and Schapira, A.H.V. (2016). PINK1 disables the anti-fission machinery to segregate damaged mitochondria for mitophagy. J. Cell Biol. *213*, 163–171.

Puigserver, P., Wu, Z., Park, C.W., Graves, R., Wright, M., and Spiegelman, B.M. (1998). A cold-inducible coactivator of nuclear receptors linked to adaptive thermogenesis. Cell *92*, 829–839.

Rakovic, A., Shurkewitsch, K., Seibler, P., Grünewald, A., Zanon, A., Hagenah, J., Krainc, D., and Klein, C. (2013). Phosphatase and tensin homolog (PTEN)-induced putative kinase 1 (PINK1)-dependent ubiquitination of endogenous Parkin attenuates mitophagy: study in human primary fibroblasts and induced pluripotent stem cell-derived neurons. J. Biol. Chem. *288*, 2223–2237.

Rambold, A.S., Kostelecky, B., Elia, N., and Lippincott-Schwartz, J. (2011). Tubular network formation protects mitochondria from autophagosomal degradation during nutrient starvation. Proc. Natl. Acad. Sci. *108*, 10190–10195.

Ramirez, A., Heimbach, A., Gründemann, J., Stiller, B., Hampshire, D., Cid, L.P., Goebel, I., Mubaidin, A.F., Wriekat, A.-L., Roeper, J., et al. (2006). Hereditary parkinsonism with dementia is caused by mutations in ATP13A2, encoding a lysosomal type 5 P-type ATPase. Nat. Genet. *38*, 1184–1191.

Ramsay, R.R., Dadgar, J., Trevor, A., and Singer, T.P. (1986). Energy-driven uptake of N-methyl-4-phenylpyridine by brain mitochondria mediates the neurotoxicity of MPTP. Life Sci. *39*, 581–588.

Rasool, S., Soya, N., Truong, L., Croteau, N., Lukacs, G.L., and Trempe, J. (2018). PINK1 autophosphorylation is required for ubiquitin recognition. EMBO Rep. *19*, e44981.

Regensburger, M., Prots, I., and Winner, B. (2014). Adult Hippocampal Neurogenesis in Parkinson's Disease: Impact on Neuronal Survival and Plasticity. Neural Plast. *2014*, 454696.

Rikka, S., Quinsay, M.N., Thomas, R.L., Kubli, D.A., Zhang, X., Murphy, A.N., and Gustafsson, Å.B. (2011). Bnip3 impairs mitochondrial bioenergetics and stimulates mitochondrial turnover. Cell Death Differ. *18*, 721–731.

Ryall, J.G., Dell'Orso, S., Derfoul, A., Juan, A., Zare, H., Feng, X., Clermont, D., Koulnis, M., Gutierrez-Cruz, G., Fulco, M., et al. (2015). The NAD+-Dependent SIRT1 Deacetylase Translates a Metabolic Switch into Regulatory Epigenetics in Skeletal Muscle Stem Cells. Cell Stem Cell *16*, 171–183.

Sanai, N., Tramontin, A.D., Quiñones-Hinojosa, A., Barbaro, N.M., Gupta, N., Kunwar, S., Lawton, M.T., McDermott, M.W., Parsa, A.T., Manuel-García Verdugo, J., et al. (2004). Unique astrocyte ribbon in adult human brain contains neural stem cells but lacks chain migration. Nature *427*, 740–744.

Sanai, N., Nguyen, T., Ihrie, R.A., Mirzadeh, Z., Tsai, H.-H., Wong, M., Gupta, N., Berger, M.S., Huang, E., Garcia-Verdugo, J.-M., et al. (2011). Corridors of migrating neurons in the human brain and their decline during infancy. Nature *478*, 382–386.

Sánchez-Aragó, M., García-Bermúdez, J., Martínez-Reyes, I., Santacatterina, F., and Cuezva, J.M. (2013). Degradation of IF1 controls energy metabolism during osteogenic differentiation of stem cells. EMBO Rep. *14*, 638–644.

Saotome, M., Safiulina, D., Szabadkai, G., Das, S., Fransson, A., Aspenstrom, P., Rizzuto, R., and Hajnoczky, G. (2008). Bidirectional Ca2+-dependent control of mitochondrial dynamics by the Miro GTPase. Proc. Natl. Acad. Sci. *105*, 20728–20733.

Sarraf, S.A., Raman, M., Guarani-Pereira, V., Sowa, M.E., Huttlin, E.L., Gygi, S.P., and Harper, J.W. (2013). Landscape of the PARKIN-dependent ubiquitylome in response to mitochondrial depolarization. Nature *496*, 372–376.

Sauvé, V., Sung, G., Soya, N., Kozlov, G., Blaimschein, N., Miotto, L.S., Trempe, J.-F., Lukacs, G.L., and Gehring, K. (2018). Mechanism of parkin activation by phosphorylation. Nat. Struct. Mol. Biol. *25*, 623–630.

Sawamoto, K., Wichterle, H., Gonzalez-Perez, O., Cholfin, J.A., Yamada, M., Spassky, N., Murcia, N.S., Garcia-Verdugo, J.M., Marin, O., Rubenstein, J.L.R., et al. (2006). New Neurons Follow the Flow of Cerebrospinal Fluid in the Adult Brain. Science *311*, 629–632.

Saxe, M.D., Battaglia, F., Wang, J.-W., Malleret, G., David, D.J., Monckton, J.E., Garcia, A.D.R., Sofroniew, M. V., Kandel, E.R., Santarelli, L., et al. (2006). Ablation of hippocampal neurogenesis impairs contextual fear conditioning and synaptic plasticity in the dentate gyrus. Proc. Natl. Acad. Sci. *103*, 17501–17506.

Schapira, A.H., Cooper, J.M., Dexter, D., Jenner, P., Clark, J.B., and Marsden, C.D. (1989). Mitochondrial complex I deficiency in Parkinson's disease. Lancet (London, England) *1*, 1269.

Schapira, A.H., Cooper, J.M., Dexter, D., Clark, J.B., Jenner, P., and Marsden, C.D. (1990). Mitochondrial complex I deficiency in Parkinson's disease. J. Neurochem. *54*, 823–827.

Schapira, A.H. V., Emre, M., Jenner, P., and Poewe, W. (2009). Levodopa in the treatment of Parkinson's disease. Eur. J. Neurol. *16*, 982–989.

Schapira, A.H.V., Chaudhuri, K.R., and Jenner, P. (2017). Non-motor features of Parkinson disease. Nat. Rev. Neurosci. *18*, 435–450.

Schell, J.C., Wisidagama, D.R., Bensard, C., Zhao, H., Wei, P., Tanner, J., Flores, A., Mohlman, J., Sorensen, L.K., Earl, C.S., et al. (2017). Control of intestinal stem cell function and proliferation by mitochondrial pyruvate metabolism. Nat. Cell Biol. *19*, 1027–1036.

Schmidt, O., Pfanner, N., and Meisinger, C. (2010). Mitochondrial protein import: from proteomics to functional mechanisms. Nat. Rev. Mol. Cell Biol. *11*, 655–667.

Scholzen, T., and Gerdes, J. (2000). The Ki-67 protein: From the known and the unknown. J. Cell. Physiol. *182*, 311–322.

Seib, D.R.M., and Martin-Villalba, A. (2014). Neurogenesis in the Normal Ageing

Hippocampus: A Mini-Review. Gerontology *61*, 327–335.

Sekine, S., and Youle, R.J. (2018). PINK1 import regulation; a fine system to convey mitochondrial stress to the cytosol. BMC Biol. *16*, 2.

Semerci, F., and Maletic-Savatic, M. (2016). Transgenic mouse models for studying adult neurogenesis. Front. Biol. (Beijing). *11*, 151–167.

Seri, B., García-Verdugo, J.M., McEwen, B.S., and Alvarez-Buylla, A. (2001). Astrocytes give rise to new neurons in the adult mammalian hippocampus. J. Neurosci. *21*, 7153–7160.

Seri, B., García-Verdugo, J.M., Collado-Morente, L., McEwen, B.S., and Alvarez-Buylla, A. (2004). Cell types, lineage, and architecture of the germinal zone in the adult dentate gyrus. J. Comp. Neurol. *478*, 359–378.

Shen, Q., Wang, Y., Kokovay, E., Lin, G., Chuang, S.-M., Goderie, S.K., Roysam, B., and Temple, S. (2008). Adult SVZ stem cells lie in a vascular niche: a quantitative analysis of niche cell-cell interactions. Cell Stem Cell *3*, 289–300.

Shiba-Fukushima, K., Imai, Y., Yoshida, S., Ishihama, Y., Kanao, T., Sato, S., and Hattori, N. (2012). PINK1-mediated phosphorylation of the Parkin ubiquitin-like domain primes mitochondrial translocation of Parkin and regulates mitophagy. Sci. Rep. *2*, 1002.

Shimura, H., Hattori, N., Kubo, S., Mizuno, Y., Asakawa, S., Minoshima, S., Shimizu, N., Iwai, K., Chiba, T., Tanaka, K., et al. (2000). Familial Parkinson disease gene product, parkin, is a ubiquitin-protein ligase. Nat. Genet. *25*, 302–305.

Shin, J.-H., Ko, H.S., Kang, H., Lee, Y., Lee, Y.-I., Pletinkova, O., Troconso, J.C., Dawson, V.L., and Dawson, T.M. (2011). PARIS (ZNF746) Repression of PGC-1α Contributes to Neurodegeneration in Parkinson's Disease. Cell *144*, 689–702.

Shin, J., Berg, D.A., Zhu, Y., Shin, J.Y., Song, J., Bonaguidi, M.A., Enikolopov, G., Nauen, D.W., Christian, K.M., Ming, G., et al. (2015). Single-Cell RNA-Seq with Waterfall Reveals Molecular Cascades underlying Adult Neurogenesis. Cell Stem Cell *17*, 360–372.

Shohayeb, B., Diab, M., Ahmed, M., and Ng, D.C.H. (2018). Factors that influence adult neurogenesis as potential therapy. Transl. Neurodegener. *7*, 4.

Shults, C.W. (2006). Lewy bodies. Proc. Natl. Acad. Sci. *103*, 1661–1668.

Silvestri, L., Caputo, V., Bellacchio, E., Atorino, L., Dallapiccola, B., Valente, E.M., and Casari, G. (2005). Mitochondrial import and enzymatic activity of PINK1 mutants associated to recessive parkinsonism. Hum. Mol. Genet. *14*, 3477–3492.

Sim, C.H., Gabriel, K., Mills, R.D., Culvenor, J.G., and Cheng, H.-C. (2012). Analysis of the regulatory and catalytic domains of PTEN-induced kinase-1 (PINK1). Hum. Mutat. *33*, 1408–1422.

Singleton, A.B., Farrer, M., Johnson, J., Singleton, A., Hague, S., Kachergus, J., Hulihan, M., Peuralinna, T., Dutra, A., Nussbaum, R., et al. (2003). α-Synuclein Locus Triplication Causes Parkinson's Disease. Science *302*, 841.

Smirnova, E., Griparic, L., Shurland, D.-L., and Bliek, A.M. van der (2001). Dynamin-related

Protein Drp1 Is Required for Mitochondrial Division in Mammalian Cells. Mol. Biol. Cell *12*, 2245–2256.

Song, Z., Ghochani, M., McCaffery, J.M., Frey, T.G., and Chan, D.C. (2009). Mitofusins and OPA1 mediate sequential steps in mitochondrial membrane fusion. Mol. Biol. Cell *20*, 3525–3532.

Sorrells, S.F., Paredes, M.F., Cebrian-Silla, A., Sandoval, K., Qi, D., Kelley, K.W., James, D., Mayer, S., Chang, J., Auguste, K.I., et al. (2018). Human hippocampal neurogenesis drops sharply in children to undetectable levels in adults. Nature *555*, 377–381.

Sorrentino, V., Menzies, K.J., and Auwerx, J. (2018). Repairing Mitochondrial Dysfunction in Disease. Annu. Rev. Pharmacol. Toxicol. *58*, 353–389.

Soubannier, V., McLelland, G.-L., Zunino, R., Braschi, E., Rippstein, P., Fon, E.A., and McBride, H.M. (2012). A Vesicular Transport Pathway Shuttles Cargo from Mitochondria to Lysosomes. Curr. Biol. *22*, 135–141.

Spalding, K.L., Bergmann, O., Alkass, K., Bernard, S., Salehpour, M., Huttner, H.B., Boström, E., Westerlund, I., Vial, C., Buchholz, B.A., et al. (2013). Dynamics of Hippocampal Neurogenesis in Adult Humans. Cell *153*, 1219–1227.

Spillantini, M.G., Schmidt, M.L., Lee, V.M.-Y., Trojanowski, J.Q., Jakes, R., and Goedert, M. (1997). α-Synuclein in Lewy bodies. Nature *388*, 839–840.

Spinazzi, M., and De Strooper, B. (2016). PARL: The mitochondrial rhomboid protease. Semin. Cell Dev. Biol. *60*, 19–28.

van Spronsen, M., Mikhaylova, M., Lipka, J., Schlager, M.A., van den Heuvel, D.J., Kuijpers, M., Wulf, P.S., Keijzer, N., Demmers, J., Kapitein, L.C., et al. (2013). TRAK/Milton Motor-Adaptor Proteins Steer Mitochondrial Trafficking to Axons and Dendrites. Neuron *77*, 485–502.

Sterky, F.H., Lee, S., Wibom, R., Olson, L., and Larsson, N.-G. (2011). Impaired mitochondrial transport and Parkin-independent degeneration of respiratory chain-deficient dopamine neurons in vivo. Proc. Natl. Acad. Sci. *108*, 12937–12942.

von Stockum, S., Marchesan, E., and Ziviani, E. (2018). Mitochondrial quality control beyond PINK1/Parkin. Oncotarget *9*, 12550–12551.

Stowers, R.S., Megeath, L.J., Górska-Andrzejak, J., Meinertzhagen, I.A., and Schwarz, T.L. (2002). Axonal transport of mitochondria to synapses depends on milton, a novel Drosophila protein. Neuron *36*, 1063–1077.

Suh, H., Consiglio, A., Ray, J., Sawai, T., D'Amour, K.A., and Gage, F.H. (2007). In Vivo Fate Analysis Reveals the Multipotent and Self-Renewal Capacities of Sox2+ Neural Stem Cells in the Adult Hippocampus. Cell Stem Cell *1*, 515–528.

Sui, Y., Horne, M.K., and Stanić, D. (2012). Reduced Proliferation in the Adult Mouse Subventricular Zone Increases Survival of Olfactory Bulb Interneurons. PLoS One *7*, e31549.

Sun, G.J., Zhou, Y., Stadel, R.P., Moss, J., Yong, J.H.A., Ito, S., Kawasaki, N.K., Phan, A.T., Oh, J.H., Modak, N., et al. (2015). Tangential migration of neuronal precursors of glutamatergic

neurons in the adult mammalian brain. Proc. Natl. Acad. Sci. *112*, 9484–9489.

Sun, J., Sun, J., Ming, G., and Song, H. (2011). Epigenetic regulation of neurogenesis in the adult mammalian brain. Eur. J. Neurosci. *33*, 1087–1093.

Sun, N., Malide, D., Liu, J., Rovira, I.I., Combs, C.A., and Finkel, T. (2017). A fluorescence-based imaging method to measure in vitro and in vivo mitophagy using mt-Keima. Nat. Protoc. *12*, 1576–1587.

Szargel, R., Shani, V., Abd Elghani, F., Mekies, L.N., Liani, E., Rott, R., and Engelender, S. (2016). The PINK1, synphilin-1 and SIAH-1 complex constitutes a novel mitophagy pathway. Hum. Mol. Genet. *25*, 3476–3490.

Takatori, S., Ito, G., and Iwatsubo, T. (2008). Cytoplasmic localization and proteasomal degradation of N-terminally cleaved form of PINK1. Neurosci. Lett. *430*, 13–17.

Takubo, K., Nagamatsu, G., Kobayashi, C.I., Nakamura-Ishizu, A., Kobayashi, H., Ikeda, E., Goda, N., Rahimi, Y., Johnson, R.S., Soga, T., et al. (2013). Regulation of Glycolysis by Pdk Functions as a Metabolic Checkpoint for Cell Cycle Quiescence in Hematopoietic Stem Cells. Cell Stem Cell *12*, 49–61.

Tanaka, A., Cleland, M.M., Xu, S., Narendra, D.P., Suen, D.-F., Karbowski, M., and Youle, R.J. (2010). Proteasome and p97 mediate mitophagy and degradation of mitofusins induced by Parkin. J. Cell Biol. *191*, 1367–1380.

Tavazoie, M., Van der Veken, L., Silva-Vargas, V., Louissaint, M., Colonna, L., Zaidi, B., Garcia-Verdugo, J.M., and Doetsch, F. (2008). A Specialized Vascular Niche for Adult Neural Stem Cells. Cell Stem Cell *3*, 279–288.

Taylor, E.B., and Rutter, J. (2011). Mitochondrial quality control by the ubiquitin–proteasome system: Figure 1. Biochem. Soc. Trans. *39*, 1509–1513.

Tondera, D., Grandemange, S., Jourdain, A., Karbowski, M., Mattenberger, Y., Herzig, S., Da Cruz, S., Clerc, P., Raschke, I., Merkwirth, C., et al. (2009). SLP-2 is required for stress-induced mitochondrial hyperfusion. EMBO J. *28*, 1589–1600.

Tormos, K.V., Anso, E., Hamanaka, R.B., Eisenbart, J., Joseph, J., Kalyanaraman, B., and Chandel, N.S. (2011). Mitochondrial Complex III ROS Regulate Adipocyte Differentiation. Cell Metab. *14*, 537–544.

Trempe, J.-F., and Fon, E.A. (2013). Structure and Function of Parkin, PINK1, and DJ-1, the Three Musketeers of Neuroprotection. Front. Neurol. *4*, 38.

Trempe, J.-F., Sauve, V., Grenier, K., Seirafi, M., Tang, M.Y., Menade, M., Al-Abdul-Wahid, S., Krett, J., Wong, K., Kozlov, G., et al. (2013). Structure of Parkin Reveals Mechanisms for Ubiquitin Ligase Activation. Science *340*, 1451–1455.

Tronel, S., Belnoue, L., Grosjean, N., Revest, J.-M., Piazza, P.-V., Koehl, M., and Abrous, D.N. (2012). Adult-born neurons are necessary for extended contextual discrimination. Hippocampus *22*, 292–298.

Truban, D., Hou, X., Caulfield, T.R., Fiesel, F.C., and Springer, W. (2017). PINK1, Parkin, and

Mitochondrial Quality Control: What can we Learn about Parkinson's Disease Pathobiology? J. Parkinsons. Dis. *7*, 13–29.

Unoki, M., and Nakamura, Y. (2001). Growth-suppressive effects of BPOZ and EGR2, two genes involved in the PTEN signaling pathway. Oncogene *20*, 4457–4465.

Vahsen, N., Candé, C., Brière, J.-J., Bénit, P., Joza, N., Larochette, N., Mastroberardino, P.G., Pequignot, M.O., Casares, N., Lazar, V., et al. (2004). AIF deficiency compromises oxidative phosphorylation. EMBO J. *23*, 4679–4689.

Valente, E.M., Abou-Sleiman, P.M., Caputo, V., Muqit, M.M.K., Harvey, K., Gispert, S., Ali, Z., Del Turco, D., Bentivoglio, A.R., Healy, D.G., et al. (2004). Hereditary Early-Onset Parkinson's Disease Caused by Mutations in PINK1. Science *304*, 1158–1160.

Varanita, T., Soriano, M.E., Romanello, V., Zaglia, T., Quintana-Cabrera, R., Semenzato, M., Menabò, R., Costa, V., Civiletto, G., Pesce, P., et al. (2015). The Opa1-dependent mitochondrial cristae remodeling pathway controls atrophic, apoptotic, and ischemic tissue damage. Cell Metab. *21*, 834–844.

Varum, S., Rodrigues, A.S., Moura, M.B., Momcilovic, O., Easley, C.A., Ramalho-Santos, J., Van Houten, B., and Schatten, G. (2011). Energy Metabolism in Human Pluripotent Stem Cells and Their Differentiated Counterparts. PLoS One *6*, e20914.

Vilar, M., and Mira, H. (2016). Regulation of Neurogenesis by Neurotrophins during Adulthood: Expected and Unexpected Roles. Front. Neurosci. *10*, 26.

Vincow, E.S., Merrihew, G., Thomas, R.E., Shulman, N.J., Beyer, R.P., MacCoss, M.J., and Pallanck, L.J. (2013). The PINK1-Parkin pathway promotes both mitophagy and selective respiratory chain turnover in vivo. Proc. Natl. Acad. Sci. U. S. A. *110*, 6400–6405.

Virbasius, J. V, and Scarpulla, R.C. (1994). Activation of the human mitochondrial transcription factor A gene by nuclear respiratory factors: a potential regulatory link between nuclear and mitochondrial gene expression in organelle biogenesis. Proc. Natl. Acad. Sci. U. S. A. *91*, 1309–1313.

Vives-Bauza, C., Zhou, C., Huang, Y., Cui, M., de Vries, R.L.A., Kim, J., May, J., Tocilescu, M.A., Liu, W., Ko, H.S., et al. (2010). PINK1-dependent recruitment of Parkin to mitochondria in mitophagy. Proc. Natl. Acad. Sci. *107*, 378–383.

Vyas, I., Heikkila, R.E., and Nicklas, W.J. (1986). Studies on the neurotoxicity of 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine: inhibition of NAD-linked substrate oxidation by its metabolite, 1-methyl-4-phenylpyridinium. J. Neurochem. *46*, 1501–1507.

Waegemans, T., Wilsher, C.R., Danniau, A., Ferris, S.H., Kurz, A., and Winblad, B. (2002). Clinical Efficacy of Piracetam in Cognitive Impairment: A Meta-Analysis. Dement. Geriatr. Cogn. Disord. *13*, 217–224.

Wang, X., and Schwarz, T.L. (2009). The Mechanism of Ca2+-Dependent Regulation of Kinesin-Mediated Mitochondrial Motility. Cell *136*, 163–174.

Wang, C., Liu, F., Liu, Y.-Y., Zhao, C.-H., You, Y., Wang, L., Zhang, J., Wei, B., Ma, T., Zhang, Q., et al. (2011a). Identification and characterization of neuroblasts in the subventricular

zone and rostral migratory stream of the adult human brain. Cell Res. *21*, 1534–1550.

Wang, H., Song, P., Du, L., Tian, W., Yue, W., Liu, M., Li, D., Wang, B., Zhu, Y., Cao, C., et al. (2011b). Parkin Ubiquitinates Drp1 for Proteasome-dependent Degradation. J. Biol. Chem. *286*, 11649–11658.

Wang, X., Winter, D., Ashrafi, G., Schlehe, J., Wong, Y.L., Selkoe, D., Rice, S., Steen, J., LaVoie, M.J., and Schwarz, T.L. (2011c). PINK1 and Parkin Target Miro for Phosphorylation and Degradation to Arrest Mitochondrial Motility. Cell *147*, 893–906.

Wauer, T., and Komander, D. (2013). Structure of the human Parkin ligase domain in an autoinhibited state. EMBO J. *32*, 2099–2112.

Weihofen, A., Thomas, K.J., Ostaszewski, B.L., Cookson, M.R., and Selkoe, D.J. (2009). Pink1 Forms a Multiprotein Complex with Miro and Milton, Linking Pink1 Function to Mitochondrial Trafficking †. Biochemistry *48*, 2045–2052.

Weisiger, R.A., and Fridovich, I. (1973). Superoxide dismutase. Organelle specificity. J. Biol. Chem. *248*, 3582–3592.

Wellen, K.E., Hatzivassiliou, G., Sachdeva, U.M., Bui, T. V., Cross, J.R., and Thompson, C.B. (2009). ATP-Citrate Lyase Links Cellular Metabolism to Histone Acetylation. Science *324*, 1076–1080.

Wenz, L.-S., Opaliński, Ł., Wiedemann, N., and Becker, T. (2015). Cooperation of protein machineries in mitochondrial protein sorting. Biochim. Biophys. Acta - Mol. Cell Res. *1853*, 1119–1129.

Whitworth, A.J., Theodore, D.A., Greene, J.C., Benes, H., Wes, P.D., and Pallanck, L.J. (2005). Increased glutathione S-transferase activity rescues dopaminergic neuron loss in a Drosophila model of Parkinson's disease. Proc. Natl. Acad. Sci. *102*, 8024–8029.

Williams, J.A., Zhao, K., Jin, S., and Ding, W.-X. (2017). New methods for monitoring mitochondrial biogenesis and mitophagy in vitro and in vivo. Exp. Biol. Med. (Maywood). *242*, 781–787.

Winkler, J., Ehret, R., Büttner, T., Dillmann, U., Fogel, W., Sabolek, M., Winkelmann, J., and Kassubek, J. (2011). Parkinson's disease risk score: Moving to a premotor diagnosis. J. Neurol. *258*, 311–315.

Winner, B., and Winkler, J. (2015). Adult neurogenesis in neurodegenerative diseases. Cold Spring Harb. Perspect. Biol. *7*, a021287.

Winner, B., Lie, D.C., Rockenstein, E., Aigner, R., Aigner, L., Masliah, E., Kuhn, H.G., and Winkler, J. (2004). Human wild-type alpha-synuclein impairs neurogenesis. J. Neuropathol. Exp. Neurol. *63*, 1155–1166.

Winner, B., Geyer, M., Couillard-Despres, S., Aigner, R., Bogdahn, U., Aigner, L., Kuhn, G., and Winkler, J. (2006). Striatal deafferentation increases dopaminergic neurogenesis in the adult olfactory bulb. Exp. Neurol. *197*, 113–121.

Winner, B., Rockenstein, E., Lie, D.C., Aigner, R., Mante, M., Bogdahn, U., Couillard-Despres,

S., Masliah, E., and Winkler, J. (2008). Mutant alpha-synuclein exacerbates age-related decrease of neurogenesis. Neurobiol. Aging *29*, 913–925.

Winner, B., Melrose, H.L., Zhao, C., Hinkle, K.M., Yue, M., Kent, C., Braithwaite, A.T., Ogholikhan, S., Aigner, R., Winkler, J., et al. (2011). Adult neurogenesis and neurite outgrowth are impaired in LRRK2 G2019S mice. Neurobiol. Dis. *41*, 706–716.

Wood-Kaczmar, A., Gandhi, S., Yao, Z., Abramov, A.S.Y., Miljan, E.A., Keen, G., Stanyer, L., Hargreaves, I., Klupsch, K., Deas, E., et al. (2008). PINK1 Is Necessary for Long Term Survival and Mitochondrial Function in Human Dopaminergic Neurons. PLoS One *3*, e2455.

Wu, Z., Puigserver, P., Andersson, U., Zhang, C., Adelmant, G., Mootha, V., Troy, A., Cinti, S., Lowell, B., Scarpulla, R.C., et al. (1999). Mechanisms Controlling Mitochondrial Biogenesis and Respiration through the Thermogenic Coactivator PGC-1. Cell *98*, 115–124.

Xu, S., Peng, G., Wang, Y., Fang, S., and Karbowski, M. (2011). The AAA-ATPase p97 is essential for outer mitochondrial membrane protein turnover. Mol. Biol. Cell *22*, 291–300.

Yamada, M., Onodera, M., Mizuno, Y., and Mochizuki, H. (2004). Neurogenesis in olfactory bulb identified by retroviral labeling in normal and 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine-treated adult mice. Neuroscience *124*, 173–181.

Yamano, K., and Youle, R.J. (2013). PINK1 is degraded through the N-end rule pathway. Autophagy *9*, 1758–1769.

Yanes, O., Clark, J., Wong, D.M., Patti, G.J., Sánchez-Ruiz, A., Benton, H.P., Trauger, S.A., Desponts, C., Ding, S., and Siuzdak, G. (2010). Metabolic oxidation regulates embryonic stem cell differentiation. Nat. Chem. Biol. *6*, 411–417.

Yang, Y., Gehrke, S., Imai, Y., Huang, Z., Ouyang, Y., Wang, J.-W., Yang, L., Beal, M.F., Vogel, H., and Lu, B. (2006). Mitochondrial pathology and muscle and dopaminergic neuron degeneration caused by inactivation of Drosophila Pink1 is rescued by Parkin. Proc. Natl. Acad. Sci. *103*, 10793–10798.

Yang, Y., Ouyang, Y., Yang, L., Beal, M.F., McQuibban, A., Vogel, H., and Lu, B. (2008). Pink1 regulates mitochondrial dynamics through interaction with the fission/fusion machinery. Proc. Natl. Acad. Sci. U. S. A. *105*, 7070–7075.

Yogalingam, G., Hwang, S., Ferreira, J.C.B., and Mochly-Rosen, D. (2013). Glyceraldehyde-3-phosphate Dehydrogenase (GAPDH) Phosphorylation by Protein Kinase Cδ (PKCδ) Inhibits Mitochondria Elimination by Lysosomal-like Structures following Ischemia and Reoxygenation-induced Injury. J. Biol. Chem. *288*, 18947–18960.

Zelko, I.N., Mariani, T.J., and Folz, R.J. (2002). Superoxide dismutase multigene family: A comparison of the CuZn-SOD (SOD1), Mn-SOD (SOD2), and EC-SOD (SOD3) gene structures, evolution, and expression. Free Radic. Biol. Med. *33*, 337–349.

Zhang, H., Ryu, D., Wu, Y., Gariani, K., Wang, X., Luan, P., D'Amico, D., Ropelle, E.R., Lutolf, M.P., Aebersold, R., et al. (2016). NAD+ repletion improves mitochondrial and stem cell function and enhances life span in mice. Science *352*, 1436–1443.

Zhang, H., Menzies, K.J., and Auwerx, J. (2018). The role of mitochondria in stem cell fate and

aging. Development *145*, dev143420.

Zhang, J., Khvorostov, I., Hong, J.S., Oktay, Y., Vergnes, L., Nuebel, E., Wahjudi, P.N., Setoguchi, K., Wang, G., Do, A., et al. (2011). UCP2 regulates energy metabolism and differentiation potential of human pluripotent stem cells. EMBO J. *30*, 4860–4873.

Zhang, J., Nuebel, E., Daley, G.Q., Koehler, C.M., and Teitell, M.A. (2012). Metabolic Regulation in Pluripotent Stem Cells during Reprogramming and Self-Renewal. Cell Stem Cell *11*, 589–595.

Zhou, C., Huang, Y., Shao, Y., May, J., Prou, D., Perier, C., Dauer, W., Schon, E.A., and Przedborski, S. (2008). The kinase domain of mitochondrial PINK1 faces the cytoplasm. Proc. Natl. Acad. Sci. *105*, 12022–12027.

Zhu, S., Li, W., Zhou, H., Wei, W., Ambasudhan, R., Lin, T., Kim, J., Zhang, K., and Ding, S. (2010). Reprogramming of Human Primary Somatic Cells by OCT4 and Chemical Compounds. Cell Stem Cell *7*, 651–655.

Ziegler, A.N., Levison, S.W., and Wood, T.L. (2015). Insulin and IGF receptor signalling in neural-stem-cell homeostasis. Nat. Rev. Endocrinol. *11*, 161–170.

Zimprich, A., Biskup, S., Leitner, P., Lichtner, P., Farrer, M., Lincoln, S., Kachergus, J., Hulihan, M., Uitti, R.J., Calne, D.B., et al. (2004). Mutations in LRRK2 Cause Autosomal-Dominant Parkinsonism with Pleomorphic Pathology. Neuron *44*, 601–607.

Ziviani, E., Tao, R.N., and Whitworth, A.J. (2010). Drosophila Parkin requires PINK1 for mitochondrial translocation and ubiquitinates Mitofusin. Proc. Natl. Acad. Sci. *107*, 5018–5023.

Zywitza, V., Misios, A., Bunatyan, L., Willnow, T.E., and Rajewsky, N. (2018). Single-Cell Transcriptomics Characterizes Cell Types in the Subventricular Zone and Uncovers Molecular Defects Impairing Adult Neurogenesis. CellReports *25*, 2457-2469.e8.

APPENDICES

Appendix I: Supplemental Data

Genotype	Sox2+	Sox2+/Nestin+	Sox2+/Nestin+/GFAP+	Sox2+/Nestin+/GFAP-
WT	55242	49590	26838	22752
	76572	72846	29952	42894
	63126	60408	25560	34848
Average WT	64980	60948	27450	33498
PINK1 KO	65226	63189	22596	40593
	55206	51390	19710	31680
	52326	35748	21294	17604
Average PINK1 KO	57586	50109	21200	29959

Table S1: Cell counts involved with comparing the SVZ neural progenitor cell population between WT and PINK1 KO mice.

Genotype	EdU+	Dcx+
WT	7830	42408
	8424	38628
	7470	43740
Average WT	7908	41592
PINK1 KO	8550	40626
	8946	39276
	10170	37800
Average PINK1 KO	9222	39234

Table S2: Cell counts involved with comparing the total number of proliferating cells and Type A cells in the SVZ between WT and PINK1 KO mice.

Genotype	Sox2+	Sox2+/Nestin+	Sox2+/Nestin+/GFAP+	Sox2+/Nestin+/GFAP-
WT	12402	5787	2601	3186
	16299	7173	3807	3366
	13905	5535	2097	3438
	15507	7452	3276	4176
Average WT	14528	6487	2945	3542
PINK1 KO	14391	7515	3951	3564
	15507	7326	4383	2943
	14022	6039	2979	3060
	14103	6426	3708	2718
Average PINK1 KO	14506	6827	3755	3071

Table S3: Cell counts involved with comparing the SGZ neural progenitor cell population between WT and PINK1 KO mice.

Genotype	Ki67+	Dcx+
WT	1707	12878
	2169	14004
	1521	12654
	1611	11385
Average WT	1752	12730
PINK1 KO	1491	9702
	1386	11007
	1185	11061
	1188	11370
Average PINK1 KO	1313	10785

Table S4: Cell counts involved with comparing the total number of proliferating cells and newborn neurons in the SGZ between WT and PINK1 KO mice.

Genotype	Sox2+	Sox2+/Nestin+	Sox2+/Nestin+/GFAP+	Sox2+/Nestin+/GFAP-
WT	56502	51516	19530	31986
	54810	48852	23580	25272
	67554	47142	25668	21474
Average WT	59622	49170	22926	26244
Parkin KO	72702	50724	27324	29268
	63486	59364	27072	32292
	52326	49356	26352	23004
Average PINK KO	62838	53148	26916	28188
SODPAR	66276	62874	29574	33300
	53766	51246	16596	34650
	60354	57510	23058	34452
Average SODPAR	60132	57210	23076	34134

Table S5: Quantitative data involved with comparing the SVZ neural progenitor cell population between WT, Parkin KO, and SODPAR mice.

Genotype	EdU+	Dcx+
WT	10584	26496
	7362	19242
	11160	35802
Average WT	9702	27180
PINK1 KO	7830	32508
	7650	35712
	7668	20646
Average PINK1 KO	7716	29622
SODPAR	10857	30093
	8172	22860
	10458	18396
Average SODPAR	9829	23783

Table S6: Cell counts involved with comparing the total number of proliferating cells and Type A cells in the SVZ between WT and PINK1 KO mice.

Genotype	Sox2+	Sox2+/Nestin+	Sox2+/Nestin+/GFAP+	Sox2+/Nestin+/GFAP-
WT	18468	9162	3159	6003
	16056	6390	2745	3645
	11898	5463	3060	2403
Average WT	15474	7005	2988	4017
Parkin KO	17478	8595	3726	4869
	18324	8838	4158	4680
	11754	5085	2880	2205
Average Parkin KO	15852	7506	3588	3918
SODPAR	14274	5958	2322	3636
	15651	6336	3600	2736
	18330	11200	5900	4770
Average SODPAR	16085	7831	3941	3714

Table S7: Quantitative data involved with comparing the SGZ neural progenitor cell population between WT, Parkin KO, and SODPAR mice.

Genotype	Ki67+	Dcx+
WT	1440	11061
	1539	12384
	1656	10413
	1269	10620
Average WT	1476	11120
PINK1 KO	1368	9630
	1557	9414
	1089	7893
Average PINK1 KO	1338	8979
SODPAR	1764	9225
	1395	9396
	972	8253
	1251	7299
Average SODPAR	1346	8543

Table S8: Cell counts involved with comparing the total number of proliferating cells and newborn neurons in the SGZ between WT, Parkin KO, and SODPAR mice.

Appendix II: Permissions to Reprint Published Figures

SPRINGER NATURE LICENSE TERMS AND CONDITIONS

May 27, 2019

This Agreement between Joseph Daniel Bastasic ("You") and Springer Nature ("Springer Nature") consists of your license details and the terms and conditions provided by Springer Nature and Copyright Clearance Center.

License Number	4597291098145
License date	May 27, 2019
Licensed Content Publisher	Springer Nature
Licensed Content Publication	Nature Reviews Endocrinology
Licensed Content Title	Insulin and IGF receptor signalling in neural-stem-cell homeostasis
Licensed Content Author	Amber N. Ziegler, Steven W. Levison, Teresa L. Wood
Licensed Content Date	Dec 2, 2014
Licensed Content Volume	11
Licensed Content Issue	3
Type of Use	Thesis/Dissertation
Requestor type	academic/university or research institute
Format	print and electronic
Portion	figures/tables/illustrations
Number of figures/tables/illustrations	1
High-res required	no
Will you be translating?	no
Circulation/distribution	<501
Author of this Springer Nature content	no
Title	The Impact of Parkinson's Disease on Mammalian Adult Neurogenesis
Institution name	University of Ottawa
Expected presentation date	Aug 2019
Portions	Figure 1
Requestor Location	University of Ottawa 451 Smyth Rd Ottawa, ON K1H 8M5 Canada Attn: Joseph Daniel Bastasic

Total **0.00 CAD**

Terms and Conditions

Springer Nature Terms and Conditions for RightsLink Permissions

Springer Nature Customer Service Centre GmbH (the Licensor) hereby grants you a non-exclusive, world-wide licence to reproduce the material and for the purpose and requirements specified in the attached copy of your order form, and for no other use, subject to the conditions below:

1. The Licensor warrants that it has, to the best of its knowledge, the rights to license reuse of this material. However, you should ensure that the material you are requesting is original to the Licensor and does not carry the copyright of another entity (as credited in the published version).

   If the credit line on any part of the material you have requested indicates that it was reprinted or adapted with permission from another source, then you should also seek permission from that source to reuse the material.

2. Where **print only** permission has been granted for a fee, separate permission must be obtained for any additional electronic re-use.

3. Permission granted **free of charge** for material in print is also usually granted for any electronic version of that work, provided that the material is incidental to your work as a whole and that the electronic version is essentially equivalent to, or substitutes for, the print version.

4. A licence for 'post on a website' is valid for 12 months from the licence date. This licence does not cover use of full text articles on websites.

5. Where **'reuse in a dissertation/thesis'** has been selected the following terms apply: Print rights of the final author's accepted manuscript (for clarity, NOT the published version) for up to 100 copies, electronic rights for use only on a personal website or institutional repository as defined by the Sherpa guideline (www.sherpa.ac.uk/romeo/).

6. Permission granted for books and journals is granted for the lifetime of the first edition and does not apply to second and subsequent editions (except where the first edition permission was granted free of charge or for signatories to the STM Permissions Guidelines http://www.stm-assoc.org/copyright-legal-affairs/permissions/permissions-guidelines/), and does not apply for editions in other languages unless additional translation rights have been granted separately in the licence.

7. Rights for additional components such as custom editions and derivatives require additional permission and may be subject to an additional fee. Please apply to Journalpermissions@springernature.com/bookpermissions@springernature.com for these rights.

8. The Licensor's permission must be acknowledged next to the licensed material in print. In electronic form, this acknowledgement must be visible at the same time as the figures/tables/illustrations or abstract, and must be hyperlinked to the journal/book's homepage. Our required acknowledgement format is in the Appendix below.

9. Use of the material for incidental promotional use, minor editing privileges (this does not include cropping, adapting, omitting material or any other changes that affect the meaning, intention or moral rights of the author) and copies for the disabled are permitted under this licence.

10. Minor adaptations of single figures (changes of format, colour and style) do not require the Licensor's approval. However, the adaptation should be credited as shown in Appendix below.

Appendix — Acknowledgements:

For Journal Content:
Reprinted by permission from [**the Licensor**]: [**Journal Publisher** (e.g. Nature/Springer/Palgrave)] [**JOURNAL NAME**] [**REFERENCE CITATION** (Article name, Author(s) Name), [**COPYRIGHT**] (year of publication)
For **Advance Online Publication papers:**
Reprinted by permission from [**the Licensor**]: [**Journal Publisher** (e.g. Nature/Springer/Palgrave)] [**JOURNAL NAME**] [**REFERENCE CITATION** (Article name, Author(s) Name), [**COPYRIGHT**] (year of publication), advance online publication, day month year (doi: 10.1038/sj.[JOURNAL ACRONYM].)
For Adaptations/Translations:
Adapted/Translated by permission from [**the Licensor**]: [**Journal Publisher** (e.g. Nature/Springer/Palgrave)] [**JOURNAL NAME**] [**REFERENCE CITATION** (Article name, Author(s) Name), [**COPYRIGHT**] (year of publication)
Note: For any republication from the British Journal of Cancer, the following credit line style applies:
Reprinted/adapted/translated by permission from [**the Licensor**]: on behalf of Cancer Research UK: : [**Journal Publisher** (e.g. Nature/Springer/Palgrave)] [**JOURNAL NAME**] [**REFERENCE CITATION** (Article name, Author(s) Name), [**COPYRIGHT]** (year of publication)
For **Advance Online Publication** papers:
Reprinted by permission from The [**the Licensor**]: on behalf of Cancer Research UK: [**Journal Publisher** (e.g. Nature/Springer/Palgrave)] [**JOURNAL NAME**] [**REFERENCE CITATION** (Article name, Author(s) Name), [**COPYRIGHT**] (year of publication), advance online publication, day month year (doi: 10.1038/sj.[JOURNAL ACRONYM])
For Book content:
Reprinted/adapted by permission from [**the Licensor**]: [**Book Publisher** (e.g. Palgrave Macmillan, Springer etc) [**Book Title**] by [**Book author**(s)] [**COPYRIGHT**] (year of publication)

Other Conditions:

Version 1.1

Questions? customercare@copyright.com or +1-855-239-3415 (toll free in the US) or +1-978-646-2777.

ELSEVIER LICENSE
TERMS AND CONDITIONS

May 27, 2019

This Agreement between Joseph Daniel Bastasic ("You") and Elsevier ("Elsevier") consists of your license details and the terms and conditions provided by Elsevier and Copyright Clearance Center.

License Number	4597311452173
License date	May 27, 2019
Licensed Content Publisher	Elsevier
Licensed Content Publication	Neuron
Licensed Content Title	Adult Neurogenesis in the Mammalian Brain: Significant Answers and Significant Questions
Licensed Content Author	Guo-li Ming,Hongjun Song
Licensed Content Date	May 26, 2011
Licensed Content Volume	70
Licensed Content Issue	4
Licensed Content Pages	16
Start Page	687
End Page	702
Type of Use	reuse in a thesis/dissertation
Portion	figures/tables/illustrations
Number of figures/tables/illustrations	2
Format	both print and electronic
Are you the author of this Elsevier article?	No
Will you be translating?	No
Original figure numbers	Parts of Figures 2 and 3
Title of your thesis/dissertation	The Impact of Parkinson's Disease on Mammalian Adult Neurogenesis
Expected completion date	Aug 2019
Estimated size (number of pages)	130
Requestor Location	University of Ottawa 451 Smyth Rd Ottawa, ON K1H 8M5 Canada Attn: Joseph Daniel Bastasic
Publisher Tax ID	GB 494 6272 12
Total	**0.00 CAD**
Terms and Conditions	

INTRODUCTION

1. The publisher for this copyrighted material is Elsevier. By clicking "accept" in connection with completing this licensing transaction, you agree that the following terms and conditions apply to this transaction (along with the Billing and Payment terms and conditions established by Copyright Clearance Center, Inc. ("CCC"), at the time that you opened your Rightslink account and that are available at any time at http://myaccount.copyright.com).

GENERAL TERMS

2. Elsevier hereby grants you permission to reproduce the aforementioned material subject to the terms and conditions indicated.

3. Acknowledgement: If any part of the material to be used (for example, figures) has appeared in our publication with credit or acknowledgement to another source, permission must also be sought from that source. If such permission is not obtained then that material may not be included in your publication/copies. Suitable acknowledgement to the source must be made, either as a footnote or in a reference list at the end of your publication, as follows:

"Reprinted from Publication title, Vol /edition number, Author(s), Title of article / title of chapter, Pages No., Copyright (Year), with permission from Elsevier [OR APPLICABLE SOCIETY COPYRIGHT OWNER]." Also Lancet special credit - "Reprinted from The Lancet, Vol. number, Author(s), Title of article, Pages No., Copyright (Year), with permission from Elsevier."

4. Reproduction of this material is confined to the purpose and/or media for which permission is hereby given.

5. Altering/Modifying Material: Not Permitted. However figures and illustrations may be altered/adapted minimally to serve your work. Any other abbreviations, additions, deletions and/or any other alterations shall be made only with prior written authorization of Elsevier Ltd. (Please contact Elsevier atpermissions@elsevier.com). No modifications can be made to any Lancet figures/tables and they must be reproduced in full.

6. If the permission fee for the requested use of our material is waived in this instance, please be advised that your future requests for Elsevier materials may attract a fee.

7. Reservation of Rights: Publisher reserves all rights not specifically granted in the combination of (i) the license details provided by you and accepted in the course of this licensing transaction, (ii) these terms and conditions and (iii) CCC's Billing and Payment terms and conditions.

8. License Contingent Upon Payment: While you may exercise the rights licensed immediately upon issuance of the license at the end of the licensing process for the transaction, provided that you have disclosed complete and accurate details of your proposed use, no license is finally effective unless and until full payment is received from you (either by publisher or by CCC) as provided in CCC's Billing and Payment terms and conditions. If full payment is not received on a timely basis, then any license preliminarily granted shall be deemed automatically revoked and shall be void as if never granted. Further, in the event that you breach any of these terms and conditions or any of CCC's Billing and Payment terms and conditions, the license is automatically revoked and shall be void as if never granted. Use of materials as described in a revoked license, as well as any use of the materials beyond the scope of an unrevoked license, may constitute copyright infringement and publisher reserves the right to take any and all action to protect its copyright in the materials.

9. Warranties: Publisher makes no representations or warranties with respect to the licensed material.

10. Indemnity: You hereby indemnify and agree to hold harmless publisher and CCC, and their respective officers, directors, employees and agents, from and against any and all claims arising out of your use of the licensed material other than as specifically authorized pursuant to this license.

11. No Transfer of License: This license is personal to you and may not be sublicensed, assigned, or transferred by you to any other person without publisher's written permission.

12. No Amendment Except in Writing: This license may not be amended except in a writing signed by both parties (or, in the case of publisher, by CCC on publisher's behalf).

13. Objection to Contrary Terms: Publisher hereby objects to any terms contained in any purchase order, acknowledgment, check endorsement or other writing prepared by you, which terms are inconsistent with these terms and conditions or CCC's Billing and Payment terms and conditions. These terms and conditions, together with CCC's Billing and Payment terms and conditions (which are incorporated herein), comprise the entire agreement between you and publisher (and CCC) concerning this licensing transaction. In the event of any conflict between your obligations established by these terms and conditions and those established by CCC's Billing and Payment terms and conditions, these terms and conditions shall control.

14. Revocation: Elsevier or Copyright Clearance Center may deny the permissions described in this License at their sole discretion, for any reason or no reason, with a full refund payable to you. Notice of such denial will be made using the contact information provided by you. Failure to receive such notice will not alter or invalidate the denial. In no event will Elsevier or Copyright Clearance Center be responsible or liable for any costs, expenses or damage incurred by you as a result of a denial of your permission request, other than a refund of the amount(s) paid by you to Elsevier and/or Copyright Clearance Center for denied permissions.

LIMITED LICENSE

The following terms and conditions apply only to specific license types:

15. **Translation**: This permission is granted for non-exclusive world **English** rights only unless your license was granted for translation rights. If you licensed translation rights you may only translate this content into the languages you requested. A professional translator must perform all translations and reproduce the content word for word preserving the integrity of the article.

16. **Posting licensed content on any Website**: The following terms and conditions apply as follows: Licensing material from an Elsevier journal: All content posted to the web site must maintain the copyright information line on the bottom of each image; A hyper-text must be included to the Homepage of the journal from which you are licensing at http://www.sciencedirect.com/science/journal/xxxxx or the Elsevier homepage for books at http://www.elsevier.com; Central Storage: This license does not include permission for a scanned version of the material to be stored in a central repository such as that provided by Heron/XanEdu.

Licensing material from an Elsevier book: A hyper-text link must be included to the Elsevier homepage at http://www.elsevier.com . All content posted to the web site must maintain the copyright information line on the bottom of each image.

Posting licensed content on Electronic reserve: In addition to the above the following clauses are applicable: The web site must be password-protected and made available only to bona fide students registered on a relevant course. This permission is granted for 1 year only. You may obtain a new license for future website posting.

17. **For journal authors:** the following clauses are applicable in addition to the above:

Preprints:

A preprint is an author's own write-up of research results and analysis, it has not been peer-reviewed, nor has it had any other value added to it by a publisher (such as formatting, copyright, technical enhancement etc.).

Authors can share their preprints anywhere at any time. Preprints should not be added to or enhanced in any way in order to appear more like, or to substitute for, the final versions of articles however authors can update their preprints on arXiv or RePEc with their Accepted Author Manuscript (see below).

If accepted for publication, we encourage authors to link from the preprint to their formal publication via its DOI. Millions of researchers have access to the formal publications on ScienceDirect, and so links will help users to find, access, cite and use the best available version. Please note that Cell Press, The Lancet and some society-owned have different preprint policies. Information on these policies is available on the journal homepage.

Accepted Author Manuscripts: An accepted author manuscript is the manuscript of an article that has been accepted for publication and which typically includes author-incorporated changes suggested during submission, peer review and editor-author communications.

Authors can share their accepted author manuscript:

- immediately
    - via their non-commercial person homepage or blog
    - by updating a preprint in arXiv or RePEc with the accepted manuscript
    - via their research institute or institutional repository for internal institutional uses or as part of an invitation-only research collaboration work-group
    - directly by providing copies to their students or to research collaborators for their personal use
    - for private scholarly sharing as part of an invitation-only work group on commercial sites with which Elsevier has an agreement
- After the embargo period
    - via non-commercial hosting platforms such as their institutional repository
    - via commercial sites with which Elsevier has an agreement

In all cases accepted manuscripts should:

- link to the formal publication via its DOI
- bear a CC-BY-NC-ND license - this is easy to do
- if aggregated with other manuscripts, for example in a repository or other site, be shared in alignment with our hosting policy not be added to or enhanced in any way to appear more like, or to substitute for, the published journal article.

Published journal article (JPA): A published journal article (PJA) is the definitive final record of published research that appears or will appear in the journal and embodies all value-adding publishing activities including peer review co-ordination, copy-editing, formatting, (if relevant) pagination and online enrichment.

Policies for sharing publishing journal articles differ for subscription and gold open access articles:

Subscription Articles: If you are an author, please share a link to your article rather than the full-text. Millions of researchers have access to the formal publications on ScienceDirect, and so links will help your users to find, access, cite, and use the best available version.

Theses and dissertations which contain embedded PJAs as part of the formal submission can be posted publicly by the awarding institution with DOI links back to the formal publications on ScienceDirect.

If you are affiliated with a library that subscribes to ScienceDirect you have additional private sharing rights for others' research accessed under that agreement. This includes use for classroom teaching and internal training at the institution (including use in course packs and courseware programs), and inclusion of the article for grant funding purposes.

Gold Open Access Articles: May be shared according to the author-selected end-user license and should contain a CrossMark logo, the end user license, and a DOI link to the formal publication on ScienceDirect.

Please refer to Elsevier's posting policy for further information.

18. **For book authors** the following clauses are applicable in addition to the above: Authors are permitted to place a brief summary of their work online only. You are not allowed to download and post the published electronic version of your chapter, nor may you scan the printed edition to create an electronic version. **Posting to a repository:** Authors are permitted to post a summary of their chapter only in their institution's repository.

19. **Thesis/Dissertation**: If your license is for use in a thesis/dissertation your thesis may be submitted to your institution in either print or electronic form. Should your thesis be published commercially, please reapply for permission. These requirements include permission for the Library and Archives of Canada to supply single copies, on demand, of the complete thesis and include permission for Proquest/UMI to supply single copies, on demand, of the complete thesis. Should your thesis be published commercially, please reapply for permission. Theses and dissertations which contain embedded PJAs as part of the formal submission can be posted publicly by the awarding institution with DOI links back to the formal publications on ScienceDirect.

Elsevier Open Access Terms and Conditions

You can publish open access with Elsevier in hundreds of open access journals or in nearly 2000 established subscription journals that support open access publishing. Permitted third party re-use of these open access articles is defined by the author's choice of Creative Commons user license. See our open access license policy for more information.

Terms & Conditions applicable to all Open Access articles published with Elsevier:

Any reuse of the article must not represent the author as endorsing the adaptation of the article nor should the article be modified in such a way as to damage the author's honour or reputation. If any changes have been made, such changes must be clearly indicated.

The author(s) must be appropriately credited and we ask that you include the end user license and a DOI link to the formal publication on ScienceDirect.

If any part of the material to be used (for example, figures) has appeared in our publication with credit or acknowledgement to another source it is the responsibility of the user to ensure their reuse complies with the terms and conditions determined by the rights holder.

Additional Terms & Conditions applicable to each Creative Commons user license:

CC BY: The CC-BY license allows users to copy, to create extracts, abstracts and new works from the Article, to alter and revise the Article and to make commercial use of the Article (including reuse and/or resale of the Article by commercial entities), provided the user gives appropriate credit (with a link to the formal publication through the relevant DOI), provides a link to the license, indicates if changes were made and the licensor is not represented as endorsing the use made of the work. The full details of the license are available at http://creativecommons.org/licenses/by/4.0.

CC BY NC SA: The CC BY-NC-SA license allows users to copy, to create extracts, abstracts and new works from the Article, to alter and revise the Article, provided this is not done for commercial purposes, and that the user gives appropriate credit (with a link to the formal publication through the relevant DOI), provides a link to the license, indicates if changes were made and the licensor is not represented as endorsing the use made of the work. Further, any new works must be made available on the same conditions. The full details of the license are available at http://creativecommons.org/licenses/by-nc-sa/4.0.

CC BY NC ND: The CC BY-NC-ND license allows users to copy and distribute the Article, provided this is not done for commercial purposes and further does not permit distribution of the Article if it is changed or edited in any way, and provided the user gives appropriate credit (with a link to the formal publication through the relevant DOI), provides a link to the license, and that the licensor is not represented as endorsing the use made of the work. The full details of the license are available at http://creativecommons.org/licenses/by-nc-nd/4.0. Any commercial reuse of Open Access articles published with a CC BY NC SA or CC BY NC ND license requires permission from Elsevier and will be subject to a fee.

Commercial reuse includes:

- Associating advertising with the full text of the Article
- Charging fees for document delivery or access
- Article aggregation
- Systematic distribution via e-mail lists or share buttons

Posting or linking by commercial companies for use by customers of those companies.

20. **Other Conditions**:

v1.9

Questions? customercare@copyright.com or +1-855-239-3415 (toll free in the US) or +1-978-646-2777.

ELSEVIER LICENSE
TERMS AND CONDITIONS

May 27, 2019

This Agreement between Joseph Daniel Bastasic ("You") and Elsevier ("Elsevier") consists of your license details and the terms and conditions provided by Elsevier and Copyright Clearance Center.

License Number	4597320211218
License date	May 27, 2019
Licensed Content Publisher	Elsevier
Licensed Content Publication	Cell Stem Cell
Licensed Content Title	Mitochondrial Dynamics Impacts Stem Cell Identity and Fate Decisions by Regulating a Nuclear Transcriptional Program
Licensed Content Author	Mireille Khacho,Alysen Clark,Devon S. Svoboda,Joelle Azzi,Jason G. MacLaurin,Cynthia Meghaizel,Hiromi Sesaki,Diane C. Lagace,Marc Germain,Mary-Ellen Harper,David S. Park,Ruth S. Slack
Licensed Content Date	Aug 4, 2016
Licensed Content Volume	19
Licensed Content Issue	2
Licensed Content Pages	16
Start Page	232
End Page	247
Type of Use	reuse in a thesis/dissertation
Intended publisher of new work	other
Portion	figures/tables/illustrations
Number of figures/tables/illustrations	1
Format	both print and electronic
Are you the author of this Elsevier article?	No
Will you be translating?	No
Original figure numbers	Figure 7Q
Title of your thesis/dissertation	The Impact of Parkinson’s Disease on Mammalian Adult Neurogenesis
Expected completion date	Aug 2019
Estimated size (number of pages)	130
Requestor Location	University of Ottawa 451 Smyth Rd

	Ottawa, ON K1H 8M5 Canada Attn: Joseph Daniel Bastasic
Publisher Tax ID	GB 494 6272 12
Total	**0.00 CAD**

Terms and Conditions

INTRODUCTION

1. The publisher for this copyrighted material is Elsevier. By clicking "accept" in connection with completing this licensing transaction, you agree that the following terms and conditions apply to this transaction (along with the Billing and Payment terms and conditions established by Copyright Clearance Center, Inc. ("CCC"), at the time that you opened your Rightslink account and that are available at any time at http://myaccount.copyright.com).

GENERAL TERMS

2. Elsevier hereby grants you permission to reproduce the aforementioned material subject to the terms and conditions indicated.

3. Acknowledgement: If any part of the material to be used (for example, figures) has appeared in our publication with credit or acknowledgement to another source, permission must also be sought from that source. If such permission is not obtained then that material may not be included in your publication/copies. Suitable acknowledgement to the source must be made, either as a footnote or in a reference list at the end of your publication, as follows:

"Reprinted from Publication title, Vol /edition number, Author(s), Title of article / title of chapter, Pages No., Copyright (Year), with permission from Elsevier [OR APPLICABLE SOCIETY COPYRIGHT OWNER]." Also Lancet special credit - "Reprinted from The Lancet, Vol. number, Author(s), Title of article, Pages No., Copyright (Year), with permission from Elsevier."

4. Reproduction of this material is confined to the purpose and/or media for which permission is hereby given.

5. Altering/Modifying Material: Not Permitted. However figures and illustrations may be altered/adapted minimally to serve your work. Any other abbreviations, additions, deletions and/or any other alterations shall be made only with prior written authorization of Elsevier Ltd. (Please contact Elsevier atpermissions@elsevier.com). No modifications can be made to any Lancet figures/tables and they must be reproduced in full.

6. If the permission fee for the requested use of our material is waived in this instance, please be advised that your future requests for Elsevier materials may attract a fee.

7. Reservation of Rights: Publisher reserves all rights not specifically granted in the combination of (i) the license details provided by you and accepted in the course of this licensing transaction, (ii) these terms and conditions and (iii) CCC's Billing and Payment terms and conditions.

8. License Contingent Upon Payment: While you may exercise the rights licensed immediately upon issuance of the license at the end of the licensing process for the transaction, provided that you have disclosed complete and accurate details of your proposed use, no license is finally effective unless and until full payment is received from you (either by publisher or by CCC) as provided in CCC's Billing and Payment terms and conditions. If full payment is not received on a timely basis, then any license preliminarily granted shall be deemed automatically revoked and shall be void as if never granted. Further, in the event that you breach any of these terms and conditions or any of CCC's Billing and Payment terms and conditions, the license is automatically revoked and shall be void as if never granted. Use of materials as described in a

revoked license, as well as any use of the materials beyond the scope of an unrevoked license, may constitute copyright infringement and publisher reserves the right to take any and all action to protect its copyright in the materials.

9. Warranties: Publisher makes no representations or warranties with respect to the licensed material.

10. Indemnity: You hereby indemnify and agree to hold harmless publisher and CCC, and their respective officers, directors, employees and agents, from and against any and all claims arising out of your use of the licensed material other than as specifically authorized pursuant to this license.

11. No Transfer of License: This license is personal to you and may not be sublicensed, assigned, or transferred by you to any other person without publisher's written permission.

12. No Amendment Except in Writing: This license may not be amended except in a writing signed by both parties (or, in the case of publisher, by CCC on publisher's behalf).

13. Objection to Contrary Terms: Publisher hereby objects to any terms contained in any purchase order, acknowledgment, check endorsement or other writing prepared by you, which terms are inconsistent with these terms and conditions or CCC's Billing and Payment terms and conditions. These terms and conditions, together with CCC's Billing and Payment terms and conditions (which are incorporated herein), comprise the entire agreement between you and publisher (and CCC) concerning this licensing transaction. In the event of any conflict between your obligations established by these terms and conditions and those established by CCC's Billing and Payment terms and conditions, these terms and conditions shall control.

14. Revocation: Elsevier or Copyright Clearance Center may deny the permissions described in this License at their sole discretion, for any reason or no reason, with a full refund payable to you. Notice of such denial will be made using the contact information provided by you. Failure to receive such notice will not alter or invalidate the denial. In no event will Elsevier or Copyright Clearance Center be responsible or liable for any costs, expenses or damage incurred by you as a result of a denial of your permission request, other than a refund of the amount(s) paid by you to Elsevier and/or Copyright Clearance Center for denied permissions.

LIMITED LICENSE

The following terms and conditions apply only to specific license types:

15. **Translation**: This permission is granted for non-exclusive world **English** rights only unless your license was granted for translation rights. If you licensed translation rights you may only translate this content into the languages you requested. A professional translator must perform all translations and reproduce the content word for word preserving the integrity of the article.

16. **Posting licensed content on any Website**: The following terms and conditions apply as follows: Licensing material from an Elsevier journal: All content posted to the web site must maintain the copyright information line on the bottom of each image; A hyper-text must be included to the Homepage of the journal from which you are licensing at http://www.sciencedirect.com/science/journal/xxxxx or the Elsevier homepage for books at http://www.elsevier.com; Central Storage: This license does not include permission for a scanned version of the material to be stored in a central repository such as that provided by Heron/XanEdu.

Licensing material from an Elsevier book: A hyper-text link must be included to the Elsevier homepage at http://www.elsevier.com . All content posted to the web site must maintain the copyright information line on the bottom of each image.

Posting licensed content on Electronic reserve: In addition to the above the following clauses are applicable: The web site must be password-protected and made available only to bona fide students registered on a relevant course. This permission is granted for 1 year only. You may obtain a new license for future website posting.

17. **For journal authors:** the following clauses are applicable in addition to the above:

Preprints:

A preprint is an author's own write-up of research results and analysis, it has not been peer-reviewed, nor has it had any other value added to it by a publisher (such as formatting, copyright, technical enhancement etc.).

Authors can share their preprints anywhere at any time. Preprints should not be added to or enhanced in any way in order to appear more like, or to substitute for, the final versions of articles however authors can update their preprints on arXiv or RePEc with their Accepted Author Manuscript (see below).

If accepted for publication, we encourage authors to link from the preprint to their formal publication via its DOI. Millions of researchers have access to the formal publications on ScienceDirect, and so links will help users to find, access, cite and use the best available version. Please note that Cell Press, The Lancet and some society-owned have different preprint policies. Information on these policies is available on the journal homepage.

Accepted Author Manuscripts: An accepted author manuscript is the manuscript of an article that has been accepted for publication and which typically includes author-incorporated changes suggested during submission, peer review and editor-author communications.

Authors can share their accepted author manuscript:

- immediately
    - via their non-commercial person homepage or blog
    - by updating a preprint in arXiv or RePEc with the accepted manuscript
    - via their research institute or institutional repository for internal institutional uses or as part of an invitation-only research collaboration work-group
    - directly by providing copies to their students or to research collaborators for their personal use
    - for private scholarly sharing as part of an invitation-only work group on commercial sites with which Elsevier has an agreement
- After the embargo period
    - via non-commercial hosting platforms such as their institutional repository
    - via commercial sites with which Elsevier has an agreement

In all cases accepted manuscripts should:

- link to the formal publication via its DOI
- bear a CC-BY-NC-ND license - this is easy to do
- if aggregated with other manuscripts, for example in a repository or other site, be shared in alignment with our hosting policy not be added to or enhanced in any way to appear more like, or to substitute for, the published journal article.

Published journal article (JPA): A published journal article (PJA) is the definitive final record of published research that appears or will appear in the journal and embodies all value-adding publishing activities including peer review co-ordination, copy-editing, formatting, (if relevant) pagination and online enrichment.

Policies for sharing publishing journal articles differ for subscription and gold open access articles:

Subscription Articles: If you are an author, please share a link to your article rather than the full-text. Millions of researchers have access to the formal publications on ScienceDirect, and so links will help your users to find, access, cite, and use the best available version.

Theses and dissertations which contain embedded PJAs as part of the formal submission can be posted publicly by the awarding institution with DOI links back to the formal publications on ScienceDirect.

If you are affiliated with a library that subscribes to ScienceDirect you have additional private sharing rights for others' research accessed under that agreement. This includes use for classroom teaching and internal training at the institution (including use in course packs and courseware programs), and inclusion of the article for grant funding purposes.

Gold Open Access Articles: May be shared according to the author-selected end-user license and should contain a CrossMark logo, the end user license, and a DOI link to the formal publication on ScienceDirect.

Please refer to Elsevier's posting policy for further information.

18. **For book authors** the following clauses are applicable in addition to the above: Authors are permitted to place a brief summary of their work online only. You are not allowed to download and post the published electronic version of your chapter, nor may you scan the printed edition to create an electronic version. **Posting to a repository:** Authors are permitted to post a summary of their chapter only in their institution's repository.

19. **Thesis/Dissertation**: If your license is for use in a thesis/dissertation your thesis may be submitted to your institution in either print or electronic form. Should your thesis be published commercially, please reapply for permission. These requirements include permission for the Library and Archives of Canada to supply single copies, on demand, of the complete thesis and include permission for Proquest/UMI to supply single copies, on demand, of the complete thesis. Should your thesis be published commercially, please reapply for permission. Theses and dissertations which contain embedded PJAs as part of the formal submission can be posted publicly by the awarding institution with DOI links back to the formal publications on ScienceDirect.

Elsevier Open Access Terms and Conditions

You can publish open access with Elsevier in hundreds of open access journals or in nearly 2000 established subscription journals that support open access publishing. Permitted third party re-use of these open access articles is defined by the author's choice of Creative Commons user license. See our open access license policy for more information.

Terms & Conditions applicable to all Open Access articles published with Elsevier:

Any reuse of the article must not represent the author as endorsing the adaptation of the article nor should the article be modified in such a way as to damage the author's honour or reputation. If any changes have been made, such changes must be clearly indicated.

The author(s) must be appropriately credited and we ask that you include the end user license and a DOI link to the formal publication on ScienceDirect.

If any part of the material to be used (for example, figures) has appeared in our publication with credit or acknowledgement to another source it is the responsibility of the user to ensure their reuse complies with the terms and conditions determined by the rights holder.

Additional Terms & Conditions applicable to each Creative Commons user license:

CC BY: The CC-BY license allows users to copy, to create extracts, abstracts and new works from the Article, to alter and revise the Article and to make commercial use of the Article (including reuse and/or resale of the Article by commercial entities), provided the user gives appropriate credit (with a link to the formal publication through the relevant DOI), provides a link to the license, indicates if changes were made and the

licensor is not represented as endorsing the use made of the work. The full details of the license are available at http://creativecommons.org/licenses/by/4.0.

CC BY NC SA: The CC BY-NC-SA license allows users to copy, to create extracts, abstracts and new works from the Article, to alter and revise the Article, provided this is not done for commercial purposes, and that the user gives appropriate credit (with a link to the formal publication through the relevant DOI), provides a link to the license, indicates if changes were made and the licensor is not represented as endorsing the use made of the work. Further, any new works must be made available on the same conditions. The full details of the license are available at http://creativecommons.org/licenses/by-nc-sa/4.0.

CC BY NC ND: The CC BY-NC-ND license allows users to copy and distribute the Article, provided this is not done for commercial purposes and further does not permit distribution of the Article if it is changed or edited in any way, and provided the user gives appropriate credit (with a link to the formal publication through the relevant DOI), provides a link to the license, and that the licensor is not represented as endorsing the use made of the work. The full details of the license are available at http://creativecommons.org/licenses/by-nc-nd/4.0. Any commercial reuse of Open Access articles published with a CC BY NC SA or CC BY NC ND license requires permission from Elsevier and will be subject to a fee.

Commercial reuse includes:

- Associating advertising with the full text of the Article
- Charging fees for document delivery or access
- Article aggregation
- Systematic distribution via e-mail lists or share buttons

Posting or linking by commercial companies for use by customers of those companies.

20. **Other Conditions**:

v1.9

Questions? customercare@copyright.com or +1-855-239-3415 (toll free in the US) or +1-978-646-2777.

Request Permission to Reuse a Published Figure for a Thesis / Dissertation

2 messages

Joseph Bastasic Mon, Mar 18, 2019 at 11:28 AM
To: publisher@iospress.nl

To Whom it May Concern,

My name is Joseph Bastasic and I am an MSc student at the University of Ottawa (Canada). I was wondering if it would be possible to obtain permission to use Figure 2 from the following paper in my thesis / dissertation (both in print and electronically)? I am not an author of this paper. The current title of my MSc thesis is "Studying the Impact of Parkinson's Disease on Mammalian Adult Neurogenesis."

Thank you,

Joseph Bastasic

Paper of Interest:
PINK1, Parkin, and Mitochondrial Quality Control: What can we Learn about Parkinson's Disease Pathobiology?

J Parkinsons Dis. 2017;7(1):13-29. doi: 10.3233/JPD-160989.

Truban D1, Hou X1, Caulfield TR1,2, Fiesel FC1,2, Springer W1,2.
Author information
1
Department of Neuroscience, Mayo Clinic, Jacksonville, FL, USA.
2
Mayo Clinic Graduate School of Biomedical Sciences, Jacksonville, FL, USA.

Carry Koolbergen <C.Koolbergen@iospress.nl> Fri, Mar 22, 2019 at 8:22 AM

To: Joseph Bastasic

J Parkinsons Dis. 2017;7(1):13-29. doi: 10.3233/JPD-160989.

Dear Joseph Bastasic,

We hereby grant you permission to reproduce the below mentioned material in **print and electronic format** at no charge subject to the following conditions:

1. Permission should also be granted by the original authors of the article in question.

2. If any part of the material to be used (for example, figures) has appeared in our publication with credit or acknowledgement to another source, permission must also be sought from that source. If such permission is not obtained then that material may not be included in your publication/copies.

3. Suitable acknowledgement to the source must be made, either as a footnote or in a reference list at the end of your publication, as follows:

"Reprinted from Publication title, Vol number, Author(s), Title of article, Pages No., Copyright (Year), with permission from IOS Press".

The publication is available at IOS Press through http://dx.doi.org/10.3233/JPD-160989

4. This permission is granted for non-exclusive world **English** rights only. For other languages please reapply separately for each one required.

5. Reproduction of this material is confined to the purpose for which permission is hereby given.

Yours sincerely

Carry Koolbergen (Mrs.)

Contracts, Rights & Permissions Coordinator

Not in the office on Wednesdays

IOS Press BV

Nieuwe Hemweg 6B

1013 BG Amsterdam

The Netherlands

Tel.: +31 (0)20 687 0022

Fax: +31 (0)20 687 0019

Email: c.koolbergen@iospress.nl / publisher@iospress.nl

URL: www.iospress.nl

Request Permission to Reuse a Published Figure for a Thesis / Dissertation

1 message

Joseph Bastasic Fri, Mar 22, 2019 at 1:20 PM
To: Truban.Dominika@mayo.edu

Dear Dr. Truban,

My name is Joseph Bastasic and I am an MSc student at the University of Ottawa (Canada). I was wondering if it would be possible to obtain permission to use Figure 2 from the following paper in my thesis / dissertation (both in print and electronically)? I have already contacted the publisher, but was advised to contact the original authors as well. The current title of my MSc thesis is "Studying the Impact of Parkinson's Disease on Mammalian Adult Neurogenesis."

Truban D, Hou X, Caulfield TR, Fiesel FC, Springer W. PINK1, Parkin, and Mitochondrial Quality Control: What can we Learn about Parkinson's Disease Pathobiology?. *J Parkinsons Dis*. 2017;7(1):13-29.

Thank you,

Joseph Bastasic

Request Permission to Reuse a Published Figure for a Thesis / Dissertation

1 message

Joseph Bastasic Fri, Mar 22, 2019 at 1:21 PM
To: Hou.Xu@mayo.edu

Dear Dr. Xu,

My name is Joseph Bastasic and I am an MSc student at the University of Ottawa (Canada). I was wondering if it would be possible to obtain permission to use Figure 2 from the following paper in my thesis / dissertation (both in print and electronically)? I have already contacted the publisher, but was advised to contact the original authors as well. The current title of my MSc thesis is "Studying the Impact of Parkinson's Disease on Mammalian Adult Neurogenesis."

Truban D, Hou X, Caulfield TR, Fiesel FC, Springer W. PINK1, Parkin, and Mitochondrial Quality Control: What can we Learn about Parkinson's Disease Pathobiology?. *J Parkinsons Dis*. 2017;7(1):13-29.

Thank you,

Joseph Bastasic

Request Permission to Reuse a Published Figure for a Thesis / Dissertation

3 messages

Joseph Bastasic Fri, Mar 22, 2019 at 1:23 PM
To: Caulfield.Thomas@mayo.edu

Dear Dr. Caulfield,

My name is Joseph Bastasic and I am an MSc student at the University of Ottawa (Canada). I was wondering if it would be possible to obtain permission to use Figure 2 from the following paper in my thesis / dissertation (both in print and electronically)? I have already contacted the publisher, but was advised to contact the original authors as well. The current title of my MSc thesis is "Studying the Impact of Parkinson's Disease on Mammalian Adult Neurogenesis."

Truban D, Hou X, Caulfield TR, Fiesel FC, Springer W. PINK1, Parkin, and Mitochondrial Quality Control: What can we Learn about Parkinson's Disease Pathobiology?. *J Parkinsons Dis*. 2017;7(1):13-29.

Thank you,

Joseph Bastasic

Caulfield, Thomas, Ph.D. <Caulfield.Thomas@mayo.edu> Fri, Mar 22, 2019 at 3:20 PM
To: Joseph Bastasic

I have no issue. Have you asked Dr. Springer?

Joseph Bastasic Sat, Mar 23, 2019 at 2:47 PM

To: "Caulfield, Thomas, Ph.D." <Caulfield.Thomas@mayo.edu>

Dear Dr. Caulfield,

I have asked Dr. Springer and he has informed me that he has no problems with me using the figure.

Thank you,

Joseph Bastasic

Request Permission to Reuse a Published Figure for a Thesis / Dissertation

1 message

Joseph Bastasic Fri, Mar 22, 2019 at 1:24 PM
To: Fiesel.Fabienne@mayo.edu

Dear Dr. Fiesel,

My name is Joseph Bastasic and I am an MSc student at the University of Ottawa (Canada). I was wondering if it would be possible to obtain permission to use Figure 2 from the following paper in my thesis / dissertation (both in print and electronically)? I have already contacted the publisher, but was advised to contact the original authors as well. The current title of my MSc thesis is "Studying the Impact of Parkinson's Disease on Mammalian Adult Neurogenesis."

Truban D, Hou X, Caulfield TR, Fiesel FC, Springer W. PINK1, Parkin, and Mitochondrial Quality Control: What can we Learn about Parkinson's Disease Pathobiology?. *J Parkinsons Dis*. 2017;7(1):13-29.

Thank you,

Joseph Bastasic

Request Permission to Reuse a Published Figure for a Thesis / Dissertation

2 messages

Joseph Bastasic Fri, Mar 22, 2019 at 1:26 PM
To: Springer.Wolfdieter@mayo.edu

Dear Dr. Springer,

Dear Dr. Truban,

My name is Joseph Bastasic and I am an MSc student at the University of Ottawa (Canada). I was wondering if it would be possible to obtain permission to use Figure 2 from the following paper in my thesis / dissertation (both in print and electronically)? I have already contacted the publisher, but was advised to contact the original authors as well. The current title of my MSc thesis is "Studying the Impact of Parkinson's Disease on Mammalian Adult Neurogenesis."

Truban D, Hou X, Caulfield TR, Fiesel FC, Springer W. PINK1, Parkin, and Mitochondrial Quality Control: What can we Learn about Parkinson's Disease Pathobiology?. *J Parkinsons Dis*. 2017;7(1):13-29.

Thank you,

Joseph Bastasic

Springer, Wolfdieter, Ph.D. <Springer.Wolfdieter@mayo.edu> Fri, Mar 22, 2019 at 4:10 PM
To: Joseph Bastasic

Hi Joseph,

Thanks for your interest.

Please feel free to use the figure.

Good luck with your thesis.

Regards,

Wolfdieter

Wolfdieter Springer, Ph.D. | Associate Professor | Mayo Clinic College of Medicine and Science | Translational Cell Biology of Parkinson's Disease

Office: 904-953-6129 | Secretary: 904-953-2439 | Lab: 904-953-6821 | Fax: 904-953-7117 | Email: Springer.Wolfdieter@mayo.edu

Mayo Clinic | Department of Neuroscience | 4500 San Pablo Road | Jacksonville, FL 32224 | mayoclinic.org