Contributions of the Thalamus to Striatal Pathology and Motor Dysfunction in Huntington's Disease

Gabriel Crevier-Sorbo

Integrated Program in Neuroscience

McGill University, Montreal

A thesis submitted to McGill University in partial fulfillment of the requirements of the degree of Master of Neuroscience

Table of Contents

This work is dedicated to my mother for her untiring support and encouragement.

Author's Contributions

Experimental work for the paper was completed by myself under the supervision of Dr. Abbas Sadikot. Conceptualization and planning were done with the help of the senior authors, Dr. Sadikot and Dr. Vladimir Rymar. Practical work including animal behavioural experiments and procedures, tissue collection and processing, quantification and behavioural analysis were done by myself, except for tissue processing and unbiased stereology for the cholinergic ablation experiment which was done by Raphael Crevier-Sorbo, under my supervision and the senior authors. Manuscript writing was done by myself and Dr. Sadikot, with further review by other co-authors.

Acknowledgements

This work is the culmination of seven years at the Cone Laboratory starting from my first summer internship in 2013. I would like to thank Dr. Abbas Sadikot for giving me the opportunity to wade into the scientific world. By supervising me throughout this degree, he shouldered the task of making me into a thorough, logical, patient and uncompromising scientist. These qualities reflect his own constant and careful approach which should be espoused by all physician-scientists. Lastly, his continued mentorship which began during my formative years in medicine have been and are very much appreciated.

I would also like to thank Dr. Vladimir Rymar for his constant guidance and technical assistance, and for making my coffee breaks always more stimulating and entertaining. Next, I would like to thank my thesis committee members, Drs Sadikot, Séguéla, and McKinney, for teaching me how to prepare for a scientific presentation and for their insight during this process. I would like to thank my brother, Raphael Crevier-Sorbo, whose hard work made part of this work possible as well as all the other Cone lab students with whom I have crossed paths with over the years for their camaraderie and friendship. I owe a special thanks to the animal handling facility staff for their help in maintaining the mouse colonies and providing operative advice. Lastly, the person whom I owe the most gratitude is my mother, Angela Sorbo, for always guiding, counselling and supporting me wherever my path has taken me.

Abstract

Huntington's disease (HD) is an autosomal dominant neurodegenerative disorder characterized by progressive loss of striatal medium spiny projection neurons (MSN), some striatal interneuron subpopulations, and degeneration of striatal afferent systems. HD manifestations include choreiform movements, dystonia, cognitive and emotional problems, often leading to premature death. Many genetic mouse models have been created to reproduce different aspects of this disease. The R6/2 model shows a rapidly progressive phenotype and reproduces much of the cell loss and protein deficits seen in HD patients. In order to determine how afferent striatal afferent systems impact the vulnerability of striatal neurons in HD, we determined whether a major source of striatal afferents arising from the thalamic parafascicular nucleus degenerates in the R6/2 model, whether thalamostriatal afferents provide a sustaining influence for striatal neurons in R6/2 mice, and whether motor symptoms are modified with lesions of the parafascicular nucleus. We found that the parafascicular nucleus degenerates in R6/2 mice and that striatal cholinergic interneurons are especially vulnerable to thalamostriatal deafferentation. We also demonstrated that both thalamostriatal and cholinergic degeneration play an important role in the development of dystonic pathology in R6/2 mice. Our results add to the growing body of evidence that dystonia in HD and other movement disorders is generated by multiple hits to the motor network. These findings suggest that new treatments aimed at preserving the thalamostriatal circuit may improve motor symptoms in HD.

Résumé

La maladie de Huntington est une maladie neurodégénérative autosomique dominante qui entraîne une perte sévère des neurones épineux moyens du noyau strié et une dégénérescence des systèmes afférents de ce dernier. Les symptômes de cette maladie incluent les mouvements choréiformes, la dystonie, les problèmes cognitifs et émotionnels, et entraine souvent une mort précoce. Plusieurs modèles murins ont été créées pour reproduire divers aspects de cette maladie. La souris R6/2 a un phénotype moteur qui progresse rapidement et reproduit les pertes cellulaires et les déficits protéiques de cette maladie. Pour mieux comprendre l'impact des afférences sur la dégénérescence des neurones du noyau strié dans la maladie de Huntington, nous avons caractérisé le noyau parafasciculaire (PF), une source importante d'afférences, dans la souris R6/2. Par la suite, nous avons étudié le soutien trophique qu'apporte les afférences thalamiques aux neurones du noyau strié ainsi que la progression phénotypique motrice dans la souris R6/2 à la suite d'une lésion du PF. Nous avons trouvé que le PF dégénère dans la souris R6/2 et que les neurones cholinergiques sont particulièrement vulnérables à la désafférentation thalamique. De plus, nous avons montré que la dégénérescence des afférences thalamostriées ou des neurones cholinergiques contribue au développement de la dystonie dans la souris R6/2. Ces expériences ont ajouté à la connaissance actuelle qui démontre que la dystonie dans la maladie de Huntington, entre autres, est générée à la suite d'impacts multiples au réseau moteur. Ces résultats suggèrent que de nouveaux traitements qui visent à préserver le circuit thalamostrié pourraient améliorer les symptômes moteurs de la maladie de Huntington.

Abbreviations

AAA	ATPase Associated with various intracellular Activities
Ache	Acetylcholinesterase
AMPA	α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid
ANOVA	Analysis of variance
BAC	Bacterial Artficial Chormosome
BDNF	Brain-derived neurotrophic factor
CHAT	Choline Acetyltransferase
CHAT +	Choline Acetyltransferase positive
CM-PF	Centromedian-parafascicular
DrD1	Dopamine receptor D1
DrD2	Dopamine receptor D2
DYT 1	Dystonia type 1
E	Embryonic day
GABA	Gamma-aminobutyric Acid
HD	Huntington's Disease
htt	huntingtin protein
LCI	Large cholinergic interneurons
LTD	Long-term depression
LTP	Long-term potentiation
M 1-5	Muscarinic receptor 1-5
mGLUR	Metabotropic glutamate receptor
mhtt	mutant huntingtin protein
MOR	Mu-opioid receptor
mRNA	Messenger Ribonucleic acid
MSNs	Medium spiny neurons
NGF	Nerve growth factor
NMDA	N-methyl D-aspartate
NMDAr	N-methyl D-aspartate receptor
NP-3	3-nitroproprionic acid
NT-3	Neurotrophin-3
NT4/5	Neurotrophin 4/5
P	Postnatal day
PF	Parafascicular
PV+	Parvalbumin positive
SEM	Standard error of the mean
SOD	Superoxide dismutase
SOP	Standard Operating Procedures
Trk	Tyrosine kinase receptor
VachT	Vesicular acetylcholine transporter
VA-VL	Ventranterior-ventrolateral
YAC	Yeast Artificial Chomosome

Chapter 1: Review of the Relevant Literature

Huntington's Disease (HD) is an autosomal dominant highly penetrant trinucleotide repeat disorder that is caused by an abnormal expansion of CAG repeats (generally >35) of the *huntingtin* gene (*mhtt*) on chromosome 4 with larger numbers of repeats leading to earlier age of onset and more severe symptoms (1993). Motor disturbances in this disorder are characterized by choreiform movements and dystonia (Louis et al., 1999; Vonsattel and DiFiglia, 1998). Early cognitive and emotional manifestations such as deficits in executive function, anxiety, obsessive compulsive disorder, depression and suicidal ideation may precede overt motor manifestations (Anderson et al., 2001; Epping and Paulsen, 2011; Fiedorowicz et al., 2011; Waldvogel et al., 2012). In late stages, hypokinetic symptoms including difficulty swallowing predominate leading to death (Paulsen, 2011; Vonsattel et al., 1985).

Neuronal loss and atrophy of striatal MSNs is well known but degeneration of striatal afferents as well as striatal interneuron sub-populations also occurs (Aylward et al., 2011; Cicchetti and Parent, 1996; Heinsen et al., 1996; Kassubek et al., 2005; Massouh et al., 2008; Matsui et al., 2014; Reiner et al., 1988; Reiner et al., 2013; Rosas et al., 2008; Rub et al., 2015; Smith et al., 2006; Spokes, 1980; Suzuki et al., 2001; Vonsattel and DiFiglia, 1998). Contextualizing these phenotypic and morphological changes requires knowledge of striatal microcircuit anatomy, and its connections with the rest of the brain, as well as knowledge of HD pathophysiology.

1.1 Organization of Afferents to Striatal Neurons and Their Intrinsic Connectivity

The basal ganglia are a group of subcortical nuclei composed of the dorsal striatum (caudate-putamen in primates), the ventral striatum or nucleus accumbens, the internal and external segments of the globus pallidus (GPi, GPe), the subthalamic nucleus (STN) and the substantia nigra. The main input structure of the basal ganglia is the dorsal striatum whereas the main output structure is the GPi/SNr in primates or entopeduncular nucleus/SNr in rodents (Chang

et al., 1981; DeVito and Anderson, 1982; DeVito et al., 1980; Gerfen, 1985; Parent, 1990; Parent and De Bellefeuille, 1982; Sidibé et al., 1997; Smith and Bolam, 1989; Van Der Kooy and Carter, 1981). The neostriatum receives glutamatergic inputs from the entire cerebral cortex, and thalamic subnuclei as well as a dense dopaminergic input from the substantia nigra *pars compacta* (SNc) (Kemp and Powell, 1971c; Parent, 1990; Parent et al., 1983; Sadikot et al., 1992a; Sadikot et al., 1992b; Szabo, 1980; Voorn et al., 2004). These afferent systems converge to innervate medium spiny neurons (MSNs), the projection neurons of the striatum (Calabresi et al., 2014; Cowan and Wilson, 1994; Huerta-Ocampo et al., 2014; Ramanathan et al., 2002a; Smith et al., 1998). Other striatal afferents include the dorsal raphe nuclei, the pedunculopontine nucleus and hypothalamic histaminergic neurons (Airaksinen and Panula, 1988; Bolam and Ellender, 2016; Kuhar et al., 1971; Nakano et al., 1990; Priestley et al., 1981; Steinbusch et al., 1980).

MSNs are GABAergic and make up approximately 95% of all striatal neurons. Classically, MSNs are divided into direct and indirect pathways (Albin et al., 1989; Bolam et al., 2000; Smith et al., 1998; Surmeier et al., 2007). These two populations are intermixed in the striatum and are differentiated by their expression of substance P/dopamine receptor D1 (DrD1) or enkephalin/DrD2 (Albin et al., 1989; Bolam et al., 2000; Gerfen et al., 1990; Gerfen, 1992; Smith et al., 1998; Surmeier et al., 2007). Each MSN innervates a different target, with direct pathway neurons projecting heavily to the GPi/SNr and the indirect pathway projecting mainly to the GPe (Bolam et al., 2000; Gerfen et al., 1990; Gerfen, 1992; Surmeier et al., 2007).

Another histochemical compartmentalization of the striatum that overlays the direct and indirect pathways are the patch (also known as striosome) and matrix compartments. This division was originally defined as islands of mu-opioid receptor (MOR) positive patches in a sea of acetylcholinesterase positive matrix (Gerfen, 1984; Herkenham and Pert, 1981). The patches make

up approximately 10-20% of the striatal volume in mammals (Johnston et al., 1990; Nakamura et al., 2009). Interestingly, the patch and matrix MSNs confine their axonal and dendritic processes to their own compartments (Bolam et al., 1988; Graybiel et al., 1986; Kawaguchi et al., 1989; Lopez-Huerta et al., 2015; Penny et al., 1988). Given that patch and matrix neurons are also direct and indirect pathway MSNs, it is no surprise that the patch/matrix compartments project heavily to the pallidal segments and SNr (Fujiyama et al., 2011; Kawaguchi et al., 1990; Levesque and Parent, 2005). There is some evidence of a direct connection from the patch MSNs to the SNc in rodents (Fujiyama et al., 2011; McGregor et al., 2019; Watabe-Uchida et al., 2012) and cats (Jimenez-Castellanos and Graybiel, 1989), but this has been difficult to reproduce in monkeys (Levesque and Parent, 2005; Parent and Parent, 2016). Patches are preferentially innervated by the limbic cortex but also receive synapses from other cortical areas (Eblen and Graybiel, 1995; Jimenez-Castellanos and Graybiel, 1987; Kincaid and Wilson, 1996). Matrix neurons receive synapses from most cortical areas (Donoghue and Herkenham, 1986; Flaherty and Graybiel, 1994; Kincaid and Wilson, 1996; Malach and Graybiel, 1986). The centromedian-parafascicular nucleus (CM-PF) or just parafascicular nucleus (PF) in rodents, is the main source of thalamostriatal afferents to the striatum, and projects almost exclusively to the matrix component (Guo et al., 2015; Lacey et al., 2007; Raju et al., 2006; Sadikot et al., 1992a; Sadikot et al., 1992b). Interestingly, PF afferents to the striatum project preferentially on MSN dendrites while other glutamatergic afferents have predominantly axospinous synapses (Dube et al., 1988; Kemp and Powell, 1971a, b; Lacey et al., 2007; Raju et al., 2006; Sadikot et al., 1992a; Sidibe and Smith, 1999; Smith et al., 2014b). Both direct and indirect pathway MSNs of the matrix compartment receive PF innervation (Doig et al., 2010; Ellender et al., 2013; Huerta-Ocampo et al., 2014; Lei et al., 2013; Wall et al., 2013). Other thalamic nuclei that project to the striatum include the central

lateral, paracentral, central medial, mediodorsal and lateral posterior nuclei, the ventroposterior medial nucleus, the pulvinar, and the VA-VL complex. However, these nuclei appear to organize their projections along functional territories rather than a specific patch or matrix predominance (Alloway et al., 2014; Beckstead, 1984; Berendse and Groenewegen, 1990, 1991; Fenelon et al., 1991; Francois et al., 1991; Groenewegen and Berendse, 1994; Guo et al., 2015; McFarland and Haber, 2000, 2001; Smith and Parent, 1986).

Interneurons make up five percent of all striatal neurons and can be GABAergic characterized by somatostatin, parvalbumin, or calretinin expression, or they can be cholinergic (Aosaki et al., 1995; Bolam et al., 2000; Kawaguchi, 1993). Striatal interneurons synapse locally with MSNs across patch and matrix borders and onto both direct and indirect pathway neurons (Bernacer et al., 2012; Chesselet and Graybiel, 1986; Cowan et al., 1990; Kawaguchi, 1992; Penny et al., 1988; Prensa et al., 1999; Rushlow et al., 1996; Walker et al., 1993; Walker and Graybiel, 1993). Parvalbumin interneurons receive synapses from cerebral cortex, and thalamic nuclei including the PF, and the ventralanterior-ventrolateral (VA-VL) nucleus. They also receive synapses from local MSNs and cholinergic interneurons (Bracci et al., 2002; Centonze et al., 2003; Koos and Tepper, 2002; Nakano et al.; Ramanathan et al., 2002b; Rudkin and Sadikot, 1999). Some evidence suggests that PV+ interneurons preferentially form synapses with direct pathway MSNs (Gittis et al., 2010). Somatostatin positive interneurons receive cortical afferents. However, there is some controversy as to whether they receive thalamic afferents with some suggesting that they do not receive thalamic inputs in rats (Kachidian et al., 1996), and others indicating they do receive inputs from the thalamus in monkeys (Sidibe and Smith, 1999). Calretinin positive interneurons receive both glutamatergic and non-glutamatergic synapses but their specific

afferents are not known in rodents (Bennett and Bolam, 1993); see (Petryszyn et al., 2017) for review.

Cholinergic interneurons make up 1-2% of all striatal neurons. They exert their effects on other striatal neurons through muscarinic (M 1-5) and nicotinic receptors (α4, β2) (Goldberg and Wilson, 2017). M1-type muscarinic receptors can directly depolarize cells while M2-type receptors can hyperpolarize cells. Cholinergic interneurons synapse onto most MSNs, cholinergic interneurons and parvalbumin positive interneurons, as well as onto the presynaptic axons of nigrostriatal and corticostriatal afferents (Gonzales and Smith, 2015). Pre-synaptically, the action of acetylcholine on nicotinic receptors promotes increased dopamine, glutamate and acetylcholine release and can lead to long-term potentiation of corticostriatal and nigrostriatal synapses (Calabresi et al., 2000; Calabresi et al., 1998; Ding et al., 2006; Pisani et al., 2007; Rice and Cragg, 2004; Threlfell et al., 2012; Zhang et al., 2002). On the other hand, inhibition of muscarinic receptors in slice preparations abolished long-term depression following low frequency stimulation of MSNs suggesting that these receptors play an important role in depotentiating MSNs following low frequency stimulation in slice preparations (Picconi et al., 2006).

Cholinergic interneurons activity is modulated by glutamatergic afferents from the PF in rodents and monkeys (Bennett and Bolam, 1994; Bolam et al., 1984; Lapper et al., 1992; Sidibe and Smith, 1999), and a smaller proportion of synapses from the secondary motor cortex and central lateral nucleus of the thalamus (Guo et al., 2015). Cholinergic interneurons co-express DrD2 and DrD1-like receptors and receive dopaminergic synapses (Pisani et al., 2007; Pisani et al., 2000). Lastly, inhibitory inputs to cholinergic interneurons include MSNs (Bolam et al., 1986), as well as somatostatin and cholinergic (M2-receptor mediated inhibition) interneurons (Gonzales and Smith, 2015; Holley et al., 2015).

In summary, the neostriatal microcircuit is complex. It receives inputs from the entire cerebral cortex, several thalamic nuclei, the substantia nigra as well as serotonergic and other pontine nuclei, and histaminergic populations. It has four main interneuron populations and two broad classes of projection neurons which are further divided into patch and matrix compartments. Subsequent sections will build on these concepts to explore how striatal afferent systems may affect cell survival and loss in the striatum and symptom progression in HD.

1.2 Possible Role of Afferents in Survival of Striatal Neurons in Development and Disease

The disease process in HD is complicated by loss of striatal afferents systems (Rub et al., 2016; Rub et al., 2015) and loss of striatal cell groups (Gomez-Tortosa et al., 2001; Massouh et al., 2008; Myers et al., 1985; Reiner et al., 1988; Reiner et al., 2013; Vonsattel and DiFiglia, 1998; Vonsattel et al., 1985). This generates several important questions: what is the effect of deafferentation from afferent degeneration on striatal neurons, does deafferentation promote striatal degeneration, and if so what mediates the protective role of afferents. In order to explore these questions, previous *in utero* experiments on developing striatal populations as well as lesion studies in adult animals are reviewed in an attempt summarize how afferents may affect survival of striatal cells.

The progenitor cells of the mammalian telencephalon develop in two germinal zones within the lateral ganglionic eminence: the dorsal zone which produces the glutamatergic cortical neurons and the ventral zone which produces GABAergic neurons of the basal ganglia, cortical GABAergic interneurons, striatal cholinergic and somatostatinergic interneurons as well as septal cholinergic interneurons (Chen et al., 2010; Marín et al., 2000; Olsson et al., 1998; Rymar and Sadikot, 2007; Rymar et al., 2004; Sadikot and Sasseville, 1997). There is a large body of evidence that amino acid neurotransmitter systems, especially glutamatergic, GABAergic and dopaminergic systems,

and their associated trophic factors promote proliferation, differentiation, migration and survival of developing neuronal populations. This pro-survival effect has been shown in the progenitor cells of the cerebral cortex, hippocampus and striatum (Behar et al., 1999; Canudas et al., 2005; Chakraborty et al., 2017; Hetman and Kharebava, 2006; Krammer, 1980; Leveille et al., 2010; Linden, 1994; Luk et al., 2003; Luk and Sadikot, 2001; Ohtani et al., 2003; Popolo et al., 2004; Reiprich et al., 2005; Sadikot et al., 1998; Suzuki et al., 2006) for review see (Jansson and Akerman, 2014). Dopaminergic and serotonergic afferents are also important for striatal and cortical progenitor cell development as they project to the lateral ganglionic eminence during the peak of striatal proliferation (Ohtani et al., 2003; Popolo et al., 2004; Vitalis and Parnavelas, 2003; Wallace and Lauder, 1983). Afferents contribute to many aspects of lateral ganglion cell development including proliferation, survival and differentiation (Brezun and Daszuta, 1999; Dooley et al., 1997; Vitalis and Parnavelas, 2003).

Glutamate is the main excitatory neurotransmitter in the brain and exerts a depolarising effect on neurons and neural progenitors expressing them through two main types of receptors: the ionotropic receptors (AMPA-KA or NMDA receptors) responsible for fast synaptic transmission or the metabotropic receptors (mGLUR 1-8) (Jansson and Akerman, 2014; Suzuki et al., 2006). AMPA/KA receptors provide important proliferative cues to neurons of the dorsal zone which become the cortical neurons (Brazel et al., 2005; LoTurco et al., 1995). However, AMPA receptor blockade *in utero* does not lead to a reduction of mature striatal cells (Luk et al., 2003; Sadikot et al., 1998). On the other hand, NMDA receptors exert proliferative effects on developing striatal neurons, and *in utero* exposure to NMDA receptor blockade leads to a reduction in the number of striatal cells and total striatal volume (Luk et al., 2003; Sadikot et al., 1998). In the dorsal germinal zone, NMDA blockade has the opposite effect and stimulates neural precursor production and

leads to a greater density of adult dentate gyrus neurons (Cameron et al., 1995). Finally, mGLUR 5 blockade was found to negatively affect the survival of striatal precursor cells but did not have the same effect on cortical neural precursors (Gandhi et al., 2008). In summary, these findings suggest a differential regulation of cortical and striatal progenitor proliferation and survival in response to stimulation of specific ionotropic and metabotropic glutamate receptor subtypes.

The striatum is composed of GABAergic cells and is devoid of glutamatergic cells (Acklin and van der Kooy, 1993; Fishell et al., 1993; Halliday and Cepko, 1992; Krushel et al., 1993). The striatal precursors receive glutamatergic inputs from thalamostriatal afferents on E 11-13 in rats; just before the phase of neuronal proliferation (E15-18) (Métin and Godement, 1996; Polleux et al., 1996). The other main source of glutamatergic afferents, the cerebral cortex, only begins to innervate the striatum after the proliferative phase on post-natal day (P) 3-4 and until P14 when adult levels of corticostriatal afferents are established (McGeorge and Faull, 1987, 1989; Sohur et al., 2014). PF lesions in neonatal rats, before major cortical innervation, led to an increase in activated caspace-3, and a 30% reduction in the number of neostriatal neurons in the post-apoptotic period (Sadikot et al., 2005). This suggests that thalamostriatal inputs may contribute to proliferative and antiapoptotic effects of glutamate receptors in the early phase of striatal development at least until cortical afferents reach these targets.

Neurotrophins are another class of molecules which strongly influence neural development. They are co-released with monoamine neurotransmitters from afferent connections and locally from developing lateral ganglionic eminence cells (Abiru et al., 1996a; Baydyuk et al., 2013; Obrietan et al., 2002). They act in concert with neurotransmitters to promote proliferation, differentiation and survival of developing cortical and striatal neurons (Abiru et al., 1996b; Baydyuk et al., 2013; Causing et al., 1997; Ghosh et al., 1994; Jung and Bennett, 1996;

Kirschenbaum and Goldman, 1995; Maisonpierre et al., 1990; Mamounas et al., 1995; Obrietan et al., 2002). There are several different types of neurotrophins. The main ones in the developing forebrain are brain derived neurotrophic factor (BDNF), nerve growth factor (NGF), neurotrophin-3 (NT3) and neurotrophin 4/5 (NT4/5) (Bartkowska et al., 2010; Chao, 2003). These exert their effects by binding to specific tyrosine kinase (trk) receptors. NGF binds to trkA, BDNF and NT4/5 bind to the trkB, and NT3 binds to the trkC receptors. Another receptor, p75, binds neurotrophins in a less specific manner and is a part of the tumor necrosis factor receptor family (Bartkowska et al., 2010; Baydyuk and Xu, 2014; Chao, 2003; Zuccato and Cattaneo, 2009). Neurotrophins, particularly BDNF and NT-3, are transported across synaptic membranes in an anterograde fashion from thalamic and nigral afferents and contribute to high neurotrophin concentrations found in developing cortical and striatal progenitor zones. They promote afferent axonal sprouting and survival of their afferent neural targets (Baydyuk et al., 2013; Baydyuk and Xu, 2014; Causing et al., 1997; Dehay et al., 2001; Fawcett et al., 1998; Jung and Bennett, 1996). Striatal progenitor cells including striatal cholinergic cells are responsive to BDNF, NT3 and NT4 but only become responsive to NGF later in development (Fryer et al., 1996; Jung and Bennett, 1996). BDNF promotes the post-differentiation survival of striatal cholinergic neurons as well as striatal MSN progenitor cells (Abiru et al., 1996b). Forebrain knock-out of BDNF during embryogenesis leads to a decreased survival and atrophy of cortical and striatal cells in adulthood (Baquet et al., 2004). Developmental knock out of genes which are involved in the transcription of BDNF like *huntingtin* also causes striatal atrophy and cortical neuron pathology (Baydyuk and Xu, 2014; Cattaneo et al., 2005; Dragatsis et al., 2000). In the postnatal period, thalamostriatal inputs to striatal neurons undergoing apoptosis are essential to their survival, and this response is mediated in part by both glutamate and BDNF (Sadikot et al., 2005).

In the adult mammalian brain, less is known about how striatal afferents modulate the survival of target neurons. Studies in cats and in rats have shown that hemidecortication in adult animals leads to atrophy of the ipsilateral striatum and thalamus (Kolb et al., 1992; Loopuijt et al., 1997a; Loopuijt et al., 1998; Loopuijt et al., 1997b; Schmanke et al., 1998; Villablance et al., 1986). Both anterograde degeneration as well as retrograde degeneration from axonal disruption may explain these findings. In the striatum, anterograde degeneration likely predominates as it does not send output to the cerebral cortex.

Decreased glutamatergic stimulation is a proposed mechanism of anterograde striatal degeneration following cortical deafferentation. In keeping with this, NMDA receptor blockade in injury models leads to increased striatal neuron loss including cholinergic interneurons. These injury models include exposure to the mitochondrial toxin 3-nitroproprionic acid (NP-3), ischemia models and mild traumatic brain injury (Hardingham, 2009; Hetman and Kharebava, 2006; Ikonomidou et al., 2000). Synaptic NMDA receptor activation leads to a host of downstream cellular processes that are pro-survival including: increased anti-apoptotic gene expression, reduced pro-apoptotic gene expression, and increased oxidant scavenging systems (Hardingham, 2009; Hardingham and Bading, 2010; Hetman and Kharebava, 2006; Ikonomidou et al., 2000).

Mature striatal cells produce no endogenous BDNF and thus also rely entirely on anterograde signaling from afferents (Baydyuk and Xu, 2014; Maisonpierre et al., 1990). Loss of BDNF-mediated trkB stimulation is therefore another possible mechanism for the subsequent striatal atrophy observed after hemidecortication. In keeping with this, BDNF and other neurotrophins promote the survival of cortical and striatal projection neurons, striatal cholinergic interneurons and medial septal cholinergic cells both *in vitro* and *in vivo* in response to ischemia, axotomy and other insults (Abiru et al., 1996b; Baquet et al., 2004; Baydyuk and Xu, 2014; Ghosh

et al., 1994; Hammond et al., 1999; Morse et al., 1993; Shulga et al., 2009; Xu et al., 2000; Zuccato et al., 2001). Overall this evidence suggests that the striatum is dependent on glutamatergic afferents for sustaining NMDA receptor stimulation through synaptic glutamate release as well as BDNF-mediated trkB receptor activation (Baquet et al., 2004; Baydyuk and Xu, 2014; Hardingham, 2009; Hardingham and Bading, 2010; Hetman and Kharebava, 2006; Ikonomidou et al., 2000; Lu et al., 2001; Morse et al., 1993).

Afferent systems also degenerate in other neurodegenerative diseases such as Alzheimer's Disease (AD), progressive supranuclear palsy (PSP) and Parkinson's Disease (PD), and may contribute to significantly to the neurodegenerative process. For example in AD the medial septal cholinergic neurons atrophy and undergo cell loss (Coyle et al., 1983). This atrophy precedes the characteristic entorhinal or hippocampal atrophy (Mesulam et al., 2004; Schmitz et al., 2016) and predicts downstream hippocampal atrophy (Schmitz et al., 2016) and cognitive decline (Teipel et al., 2018). In PD and PSP, brainstem atrophy of large cholinergic cell groups that provide afferents to dopaminergic midbrain cells precedes dopaminergic cell loss (Hirsch et al., 1987). Furthermore, noradrenergic inputs to dopaminergic neurons protect vulnerable dopaminergic neurons from toxic injury, and noradrenergic cell loss may also precede dopaminergic cell loss in PD (Del Tredici and Braak, 2013; Hassani et al., 2020).

The posterior intralaminar nuclei provide important glutamatergic inputs to the striatum and are severely affected in PD, PSP and HD (Halliday, 2009; Heinsen et al., 1996). In PD, loss of the CM-PF complex occurs early in the progression of motor symptoms (Brooks and Halliday, 2009), is associated with a loss of dendritic spines on MSNs (Zaja-Milatovic et al., 2005) and correlates with worse alpha synuclein deposition (Halliday, 2009). CM-PF loss is also present in presymptomatic MPTP-treated monkeys suggesting that degeneration occurs early in the disease

process (Villalba et al., 2014). Furthermore, work on toxin-induced models of PD suggests that glutamatergic afferents to the substantia nigra are protective during the degenerative process (Bezard et al., 1997). Contrary to these findings, evidence in MPTP treated rats suggests that posterior intralaminar lesions prevent some neurochemical changes due to dopaminergic denervation (Kerkerian-Le Goff et al., 2009). However, the neurochemical effects of PF lesion seen in rats were not reproduced in MPTP-treated monkeys (Lanciego et al., 2008). As a result of this contradictory evidence, CM-PF neuromodulation is likely to be of little benefit in PD (Lanciego et al., 2008; Smith et al., 2014b). In HD, the CM-PF undergoes significant degeneration (Heinsen et al., 1996). Work in mouse models suggests that thalamostriatal deafferentation occurs early in the disease process (Deng et al., 2013; Deng et al., 2014) and is associated with concurrent pathological changes in striatal cholinergic interneurons (Deng and Reiner, 2016). Overall, these findings suggest that the CM-PF is an early and important target of cell loss in several neurodegenerative diseases. The concurrent pathological changes observed in thalamostriatal afferents as a result of that cell loss are likely to have important ramifications for disease progression and the development of motor symptoms in PD, PSP, and HD.

In summary, experiments in both developing and mature striatal populations suggest that afferents are important for the survival of vulnerable striatal cells. Mechanisms mediating this protective role include: glutamatergic mGLUR 5 and NMDA receptors, serotonergic and dopaminergic innervation, and from neurotrophins. Furthermore, research suggests that deafferentation may play a key role in downstream pathology in many neurodegenerative diseases. More specifically, the deafferentation of vulnerable striatal neurons may lead to dysfunction and cell loss of those neurons in HD.

1.3 Proposed Pathophysiology of Huntington's Disease

Huntingtin (htt) protein is water soluble and ubiquitously expressed but is enriched in the brain and testis. It is found anywhere within neurons from the nucleus to the cytoplasm to clathrin-coated vesicles (DiFiglia et al., 1995; Ferrante et al., 1997; Velier et al., 1998). It is necessary for life as evidenced by the preservation of its homologue across diverse phyla and interacts with over 50 proteins; reviewed by (Reiner et al., 2011). Abnormal expansion of CAG repeats (generally >35) of exon 1 of the *huntingtin* gene (*mhtt*) on chromosome 4 leads to HD (1993). At the protein level, two possible pathophysiologic processes may be at play; the first being that *mhtt* may have gained a cytotoxic function and or that normal *htt* function has been impaired by the polyglutamine expansion. Initial studies suggested that people heterozygous or homozygous for the mutation have the same illness course (Myers et al., 1989; Wexler et al., 1987) but more recent studies have shown that homozygous individuals have an earlier onset and display more symptoms then heterozygous counterparts (Squitieri et al., 2003). On the other hand, transgenic models such as R6/2 animals have severe deficits despite having two intact copies of the *htt* gene (Mangiarini et al., 1996), humans and mice with only one functioning copy of the *htt* gene are asymptomatic and individuals heterozygous for a mutated gene express both good and bad copies (Ambrose et al., 1994; Duyao et al., 1995a). This seemingly contradictory evidence suggests that both processes likely contribute to the pathogenesis of the disease.

Early experiments using glutamate analogs or glutamate receptor agonists to generate lesional models of Huntington's disease has led to the proposal of a hyper-excitatory neurotransmitter hypothesis of HD pathophysiology (Beal et al., 1991; Beal et al., 1986; Coyle and Schwarcz, 1976; Ferrante et al., 1993). These lesion models recreated the striatal cell loss of HD and produced a dystonic "clasping" phenotype. *In vitro* experiments then showed that over

stimulation of NMDA receptors led to imbalance of calcium homeostasis in neurons leading to the activation of apoptotic pathways, including mitochondrial membrane destabilization and eventually cell death (Milnerwood and Raymond, 2007, 2010; Raymond et al., 2011; Sepers and Raymond, 2014; Zeron et al., 2002). The interaction between *mhtt* protein and NMDA receptor subunits has been linked to pathological intracellular calcium signaling *in vitro* in some HD models (Ali and Levine, 2006; Song et al., 2003; VanDongen, 2008). Indeed, Stack et al. described increased survival following cortical and nigral lesions in R6/2 mice as well as increased neuronal volume in the striatum following cortical lesions (Stack et al., 2007). Other evidence suggests that NMDA receptor mediated toxicity is a more nuanced story as blockade of NMDA receptors leads to increased striatal apoptosis in the NP-3 model—a lesional model of Huntington's disease (Ikonomidou et al., 2000). Furthermore, it is likely that extra-synaptic NMDAr are responsible for the induction of apoptotic pathways while synaptic activation is protective and induces anti-apoptotic proteins, promotes anti-oxidant generation, and leads to cell survival (Hardingham, 2009; Hardingham and Bading, 2010; Hetman and Kharebava, 2006). In keeping with the former, NMDA receptors have been shown to be downregulated in the striatum of HD patients suggesting a loss of their neurotrophic role even in presymptomatic stages of the disease (Albin et al., 1990a; Albin et al., 1990b; Young et al., 1988).

Another proposed mechanism of cell loss and degeneration in HD involves neurotrophic factor reduction, particularly BDNF, which has been shown to play an important role in the function, and survival of mature neurons (Baydyuk and Xu, 2014). This protein and its receptor, trk B has been found to be reduced in multiple brain areas including the striatum of HD models and in the CSF of HD patients (Baydyuk and Xu, 2014; Nguyen et al., 2016; Samadi et al., 2013; Zuccato and Cattaneo, 2007, 2009; Zuccato et al., 2001; Zuccato et al., 2005; Zuccato et al., 2008).

At the cellular level, normal *htt* positively regulates the transcription of BDNF among other genes through its interaction with repressor proteins; a function which is lost in the mutated protein (Reilly, 2001; Zuccato et al., 2007; Zuccato et al., 2003). Moreover, mice who have conditional forebrain BDNF knockout have similar delayed cortical and striatal atrophy as well as a dystonic phenotype and decreased survival seen in mouse models of HD (Baquet et al., 2004). Many others have demonstrated that inducing the transcription of BDNF protein in the striatum and striatal afferents or exogenous administration of BDNF rescues HD models both phenotypically and at the neuronal level further linking BDNF dysfunction to HD pathogenesis (Bemelmans et al., 1999; Giampà et al., 2013; Silva et al., 2015; Xie et al., 2010; Zuccato et al., 2005). BDNF helps promote survival among neurons during embryogenesis and axonal sprouting of early afferent systems (Mamounas et al., 1995) so BDNF reduction which was previously noted in HD models (Baydyuk and Xu, 2014; Zuccato and Cattaneo, 2007, 2009) may lead to inappropriate wiring of the brain. In keeping with this notion, recent evidence in the Q140 mouse model demonstrates an early deficit in thalamostriatal synapses long before neuronal atrophy, loss of cortical synapses or behavioural changes are seen (Deng and Reiner, 2016; Deng et al., 2013; Deng et al., 2014).

Other pathogenic mechanisms which likely contribute to neurodegeneration in HD include: cytoskeletal abnormalities (Fernandez-Nogales et al., 2016), slowed and inappropriate axonal transport (Goldstein, 2012; Smith et al., 2014a; Weiss and Littleton, 2016), prion-like effects of Huntington aggregates (Cicchetti et al., 2014; Cicchetti et al., 2009; Pearce et al., 2015) and, importantly, excessive immune activation and microglia mediated release of inflammatory factors (Crotti et al., 2014; Franciosi et al., 2012; Giampa et al., 2009; Pavese et al., 2006; Pearce et al., 2015; Politis et al., 2011; Stack et al., 2006; Tai et al., 2007). The multiple proposed pathogenic mechanisms likely work in synchrony to create a perfect storm, and lead to impressive striatal but

also cortical, thalamic and cerebellar cell loss which is associated with the debilitating symptoms of this disease.

1.4 Animal Models of Huntington's Disease

Before the advent of genetic models of HD or indeed the discovery of the *htt* gene, striatal lesion models in both rodents and non-human primates had been used to model the disease as these could create striatal lesions. Various substances have been used to create these lesions including: kainic acid, quinolinic acid and 3-nitroproprionic acid (NP-3) (Beal et al., 1991; Beal et al., 1986; Coyle and Schwarcz, 1976; Ferrante et al., 1993). The main limitation of these models is that they do not reproduce the progressive neurodegenerative process which occurs in HD and that pathology is restricted to the lesioned areas, usually the striatum. In HD, pathological changes can be found throughout the brain and also in other organ systems including, but not limited to, the immune and endocrine systems (Crotti et al., 2014; Saleh et al., 2009; Vonsattel and DiFiglia, 1998).

With the advent of modern genetics, Huntington's disease was found to be a highly penetrant, monogenetic disorder (1993). This characteristic has allowed for the development of a plethora of animal models to study the neuropathological changes of this disease. These include fly and worm models as well as rodent, porcine and non-human primate models (Jackson et al., 1998; Menalled et al., 2009; Yang et al., 2010; Yang et al., 2008) see (Pouladi et al., 2013) for review. Rodent models are the most studied as they allow for large scale breeding experiments, and complex phenotypical characterization while providing *in situ* evidence into the neuropathology brought on by *mhtt* (Baydyuk and Xu, 2014; Hickey et al., 2008; Menalled et al., 2009; Menalled, 2005; Menalled et al., 2002b; Petrasch-Parwez et al., 2007; Samadi et al., 2013; Tallaksen-Greene et al., 2005; Yu-Taeger et al., 2012).

There exist two broad categories of genetic mouse models: transgenic mouse models and knock-in models. To create the transgenic models, the N-terminal portion of the human *mhtt* gene was randomly inserted into the mouse genome (R6 and N-171 lines) (Mangiarini et al., 1996; Schilling et al., 1999). In the R6/1 and R6/2 lines, the untranslated human *mhtt* exon 1 sequence is under control of the human *htt* promoter (Mangiarini et al., 1996) while in the N-171 model, a truncated cDNA *mhtt* sequence corresponding to the first 802 base pairs of exon 1 and containing 82 CAG repeats was inserted into the mouse genome under control of the mouse prion protein promoter (Schilling et al., 1999). The second type of genetic HD mouse models are the knock-in models in which one or both of the normal mouse *htt* genes have been replaced with either: exon 1 of the human *mhtt* gene added to a normal mouse *htt* gene (Hdh and Q140 models) often referred to as "chimeric" models or the entire human *mhtt* gene including promoter and intron regions (YAC, BACHD lines) (Gray et al., 2008; Hickey et al., 2008; Hodgson et al., 1999; Menalled et al., 2002a; Pouladi et al., 2013; Slow et al., 2003; Wheeler et al., 2000). These models have been summarized in **Table 1.0** below.

Each of these models has different advantages and disadvantages. While the full length knock-in lines (YAC and BACHD) may have a good construct validity, they have a milder phenotype then expected for the large number of repeats. These mice have normal survival and exhibit only mild neurodegenerative changes such as synaptic deficits, striatal and cortical atrophy, mild striatal cell loss as well as protein deficits including *mhtt* aggregation (Gray et al., 2008; Hickey et al., 2008; Hodgson et al., 1999; Menalled et al., 2002a; Pouladi et al., 2013; Pouladi et al., 2012; Slow et al., 2003; Teo et al., 2016; Van Raamsdonk et al., 2005; Wheeler et al., 2000).

Table 1.0: Genetic and Phenotypical Characteristics of HD Mouse Lines

	Transgenic Mouse Lines		**Knock in Mouse Lines**			
	R6	**N-171**	**Hdh**	**HD Q**	**YAC128**	**BACHD**
Repeat Number	R6/1: 75-90 R6/2: 115-125	82	Several strains available. Most common: Hdh150	Several strains available. Most common: Q140 or Q175	100-128	97-98
Construct Type	Human htt promoter; Human mhtt exon 1	Mouse prion protein promoter; Human mhtt exon 1	Mouse htt promoter; CAG expansion in exon 1 of mouse htt	Mouse htt promoter; Chimeric human mhtt exon1//mouse htt	Full length Human mhtt genomic construct including promoter	Full length Human mhtt genomic construct including promoter
Motor Phenotype	R6/2: 6-9 wk – rotarod deficits, clasping, decreased grip strength, hypoactivity, weight loss R6/1: 16-20 wk milder phenotype	16-20 wk: rotarod deficits, clasping, hypoactivity	20-40 wks: rotarod deficits, clasping, hypoactivity, weight loss	1 mo: hyperactivity, 4mo: hypoactivity. 12 mo rotarod deficits	> 12 mo: rotarod deficits	24 wks: rotarod deficits
Terminal Timepoint	R6/2: 13-15wk R6/1: 40-50wk	24-30 wk	Hdh150: ~52wk	unaffected	unaffected	unaffected

On the other hand, the transgenic and chimeric knock-in mouse lines, particularly the R6/2 model, reproduce severe juvenile HD, developing an early and severe dystonic phenotype, choreiform movements, seizures, cognitive impairment, decreased spontaneous locomotion, and decreased survival (Bailey and Johnson, 2005, 2006; Carter et al., 1999; Dedeoglu et al., 2002; Hickey et al., 2005; Hockly et al., 2002; Menalled et al., 2009; Phillips et al., 2005; Sawiak et al., 2016; Sawiak et al., 2009; Stack et al., 2005; Wood et al., 2010; Wood et al., 2011; Wood et al., 2008; Wood and Morton, 2015; Wood et al., 2007). The R6/2 line, in particular, has prominent striatal and cortical deficits including mhtt aggregates, protein derangements, decreased trophic factor mRNA, abnormal trophic factor receptor activation, striatal MSN loss, striatal volume reduction, and reduction of MSN soma size (Baydyuk and Xu, 2014; Nguyen et al., 2016; Rattray

et al., 2013; Samadi et al., 2013). Furthermore, this model has a stereotyped phenotypical progression which is advantageous when assessing concurrent behavioural and pathological changes at discreet timepoints (Menalled et al., 2009; Samadi et al., 2013). In our experiments, ovarian transplanted female mice were crossbred with B6CBA males to generate R6/2 mice as R6/2 females are infertile. The mice used in these experiments had approximately 120 CAG repeats and this was verified for each experimental animal.

1.5 Proposed Pathophysiology of Dystonia in HD and Other Movement Disorders

Dystonia is defined as sustained or intermittent muscle contractions causing twisting or repetitive movements and abnormal postures (Klein and Fahn, 2013; Marsden, 1976). It is the second most common motor symptom in HD after chorea and is present in 95% of patients (Louis et al., 1999). Dystonia also occurs in over a dozen primary dystonic syndromes, as a complication of Parkinson's treatment, after chronic neuroleptic use, and in other neurodegenerative diseases (D'Abreu et al., 2011; Goto et al., 2005; Kanovsky et al., 2015; Marálek, 2000; Marsden, 1976; Phukan et al., 2011; Steele et al., 1964; Tinazzi et al., 2003; Tolosa and Compta, 2006; van Harten et al., 1997; Wilson and Hess, 2013).

Several primary dystonia syndromes and their causative genetic mutations have been elucidated. These genes are involved in a myriad of cellular processes including: nuclear membrane integrity (Goodchild et al.), processing of secretory proteins and synaptic vesicle machinery (Granata and Warner, 2010; Hewett et al., 2007), TATAbox binding genes responsible for broad regulation of gene transcription (Herzfeld et al., 2013; Sako et al., 2011), detoxification and stress response proteins (Lee et al., 2004), cytoskeletal protein misfolding (Esapa et al., 2007; Klein et al., 2002), Na/K ATPases (de Carvalho Aguiar et al., 2004), co-factor production for dopamine synthesis (Ichinose et al., 1994), BDNF polymorphisms (Cramer et al., 2010; Sako et

al.) and calcium channel signaling (Breakefield et al., 2008; Hess and Jinnah, 2015; Kanovsky et al., 2015). It is not surprising, given the high prevalence of dystonia in HD, that many these cellular processes are also affected in HD (Anborgh et al., 2005; Fernandez-Nogales et al., 2016; Goldstein, 2012; Hardingham, 2009; Hardingham and Bading, 2010; Hetman and Kharebava, 2006; Ikonomidou et al., 2000; Mao et al., 2005; Raymond et al., 2011; Ribeiro et al., 2010; Rong et al., 2003; Smith et al., 2014a; Vanhoutte and Bading, 2003; Weiss and Littleton, 2016; Zuccato et al., 2001; Zuccato et al., 2003).

At the circuit level, *in vivo* imaging and neuronal recordings in dystonia patients demonstrate that the putamen is hyperactive, while the GPi is hypoactive and abnormally locked to cortical beta oscillations (Galardi et al., 1996; Neumann et al., 2015; Zeuner et al., 2015). Other studies show increased pallidal activation in response to repeated tasks (Lenz et al., 1998). Deep brain stimulation of the globus pallidus improves symptoms further suggesting that abnormal basal ganglia plasticity is important to its pathogenesis (Mink, 2018; Ostrem and Starr, 2008). *In vivo* imaging also suggests that sensory and motor integration in the cerebellum is abnormal in various dystonia syndromes and can normalize after treatment (Avanzino and Fiorio, 2014; Dresel et al., 2014; Hutchinson et al., 2000; Koch et al., 2014; Lehericy et al., 2013; Niethammer et al., 2011). On the other hand, patients with DYT1, the most common primary dystonia, have no major atrophy of the basal ganglia, substantia nigra, thalamus or cerebellum at autopsy although detailed stereological analyses are lacking (de Carvalho Aguiar and Ozelius, 2002; Furukawa et al., 2000; Hedreen et al., 1988; Holton et al., 2008; Kanovsky et al., 2015). This possibly reflects a difference in pathogenic process between progressive neurodegenerative diseases with dystonia compared to some of the primary dystonic syndromes.

Lesional pathology such as strokes or tumours can also cause dystonia. Dystonia occurring after an acquired lesion is referred to as secondary dystonia and can occur many months after discreet lesions in the basal ganglia (Bhatia and Marsden, 1994; Demierre and Rondot, 1983; Marsden et al., 1985). Cerebellar lesions occasionally also present as dystonia rather than ataxia or dysmetria (LeDoux and Brady, 2003; Lee and Marsden, 1994; Marsden et al., 1985; Meissner et al., 1987; Rantamäki et al., 2001; Rumbach et al., 1995; Zadro et al., 2008). Lastly, thalamic lesions have been linked to hemidystonia, hand dystonia, and cervical dystonia. In most of those cases, the lesions affected the paramedian or posterolateral thalamus but in a handful of cases dystonia began after midbrain infarct or ventroanterior/ventrolateral thalamotomy (Lee and Marsden, 1994; Marsden et al., 1985; Meissner et al., 1987; Rantamäki et al., 2001). Indeed, neurodegeneration in all three of these brain regions occurs in HD (GOEBEL et al., 1978; Heinsen et al., 1996; Rees et al., 2014; Reiner et al., 1988; Rub et al., 2013; Rub et al., 2015; Vonsattel and DiFiglia, 1998; Waldvogel et al., 2012) and is at least conceptually similar to what occurs in secondary dystonia syndromes.

1.6 Animal Models of Dystonia

Many genetic syndromes causing primary dystonia and various genetic mutations have been linked to them. However, few are highly penetrant and many are seen in only a handful of individuals (Breakefield et al., 2008; Kanovsky et al., 2015). This has led to much difficulty in developing animal models to study these syndromes as they often lack a robust dystonic phenotype upon which interventions can be tested (Breakefield et al., 2008). The first models arose spontaneously from animal colonies around the world although their phenotype may be more ataxic then dystonic (Duchen, 1976; Duchen et al., 1964; Messer and Strominger, 1980). With the advent of modern genetics, mouse models displaying a more convincing dystonic phenotype have

been created. The most commonly described dystonic phenotype in these models is “clasping” behaviour in which rodents have sustained or repetitive limb contractions towards the midline after being lifted in the air by their tail. This has good face validity as a surrogate for dystonia and is commonly accepted as such (Gernert et al., 2000; Jardim et al., 2001; Lalonde and Strazielle, 2011; Menalled et al., 2009; Neychev et al., 2011; Pappas et al., 2015; Samadi et al., 2013; Wilson and Hess, 2013). Furthermore, it is easily and reliably elicited in R6/2 and worsens throughout their lifespan making it is a useful behavioural phenotype to study in those animals.

DYT 1 is the most common primary dystonia and is caused by a loss of function mutation in exon 5 of the TOR1A gene coding for the Torsin A protein (Kanovsky et al., 2015; Ozelius et al., 1997). Transgenic mice with mutations found in primary dystonic syndromes have good construct validity. Transgenic mice which express human or mouse mutated TorsinA are mildly hyperactive and have a dystonic clasping phenotype (Bao et al., 2010; Hewett et al., 2010; Hewett et al., 2007; Lange et al., 2011; Richter and Richter, 2014; Shashidharan et al., 2005; Song et al., 2012; Uluğ et al., 2011). They also have decreased fractional anisotropy in cerebellothalamocortical and thalamostriatal connections suggesting disrupted network connectivity affecting those circuits (Ramdhani et al., 2014; Yang et al., 2014).

Striatal microcircuitry in dystonia has been interrogated by several groups. Dystonia models have abnormal cholinergic interneuron excitation to thalamostriatal stimulation, paradoxical D2 receptor excitation of cholinergic neurons, decreased intrastriatal dopamine release secondary to altered cholinergic tone, inability of striatal neurons to undergo long-term depression and increased corticostriatal synaptic potentiation (Bao et al., 2010; Hewett et al., 2010; Hewett et al., 2007; Lange et al., 2011; Martella et al., 2014; Pisani et al., 2006; Richter and Richter, 2014; Sciamanna et al., 2012a; Sciamanna et al., 2012b; Shashidharan et al., 2005; Song et al., 2012;

Uluğ et al., 2011). The protein mutated in DYT 1, TorsinA, is enriched in normal striatal cholinergic neurons (Konakova et al., 2001). Other work indicates that thalamostriatal afferents also produce abnormal responses in DYT1 cholinergic neurons (Sciamanna et al., 2012b). These synaptic deficits may be a product of abnormal brain development as selective Torsin A forebrain knock out models have an early post-natal loss of cholinergic striatal interneurons and decreased striatal acetylcholine release and a clasping phenotype (Pappas et al., 2015). Although DYT1 models have a good construct validity, they do not capture the breadth of motor pathology in dystonic syndromes and so may not provide a universal mechanistic understanding for this movement disorder.

Many other models with a prominent dystonic clasping phenotype have been developed and recently reviewed (Eskow Jaunarajs et al., 2015; Hess and Jinnah, 2015; Lalonde and Strazielle, 2011; Lehericy et al., 2013; Neychev et al., 2011; Wilson and Hess, 2013). Some examples include : NP-3 striatal lesion models, transgenic HD models including R6/2, Hdh mice, BDNF forebrain knock out models, and mice with mutations in various cerebellar proteins (Baquet et al., 2004; Fernagut et al., 2002; Gernert et al., 2000; Hess and Jinnah, 2015; Jardim et al., 2001; Lalonde and Strazielle, 2007, 2011; Menalled et al., 2009; Neychev et al., 2011; Pappas et al., 2015; Samadi et al., 2013; Wilson and Hess, 2013). These studies echo the pathophysiologic changes described in DYT 1 models such as: abnormal network connectivity, striatal synaptic plasticity, abnormal cholinergic responses to thalamostriatal afferents, dysregulated BDNF signaling and cerebellar pathology (Baquet et al., 2004; Nguyen et al., 2016; Picconi et al., 2006; Samadi et al., 2013; Tanimura et al., 2016).

In summary, knock-in DYT 1 mice, HD models, and many other mouse models provide converging lines of evidence that aberrant cerebellar, thalamic and basal ganglia connectivity as

well as intrinsic striatal microcircuit pathology play a role in the development of a dystonic phenotype. These parts of the motor circuit namely the basal ganglia, thalamus and cerebellum have also been shown to be abnormal on *in vivo* imaging and in intraoperative recordings in patients suffering from dystonia (Avanzino and Fiorio, 2014; Blood et al., 2012; Dresel et al., 2014; Galardi et al., 1996; Hutchinson et al., 2000; Koch et al., 2014; Lehericy et al., 2013; Mink, 2018; Neumann et al., 2015; Niethammer et al., 2011; Ostrem and Starr, 2008; Semenova et al., 2016).

1.7 Hypotheses and Experimental Plan

Afferent systems provide trophic support to both mature and immature striatal neurons through glutamatergic stimulation and anterograde trophic factor release (Baydyuk and Xu, 2014; Sadikot et al., 2005). Previous work has demonstrated that PF lesions in neonatal rats led to an increase in activated caspace-3, decreased BDNF on immunoblots and a 30% reduction in the number of neostriatal neurons in the post-apoptotic period (Sadikot et al., 2005). These findings are in keeping with the neurotrophic hypothesis of neural development whereby cell death due to limited concentrations of trophic factors at targets allows for the survival of the appropriate number of neurons (Luk et al., 2003; Luk and Sadikot, 2001, 2004; Sadikot et al., 1998). In the mature striatum, the neurotrophic effects of TS afferents are less well studied although no striatal cell loss has been reported in wildtype rodents undergoing PF lesions (Bacci et al., 2002; Bradfield and Balleine, 2017; Bradfield et al., 2013; Brown et al., 2010). However, this resistance to deafferentation may not apply to mature striatal neurons undergoing degeneration which may once again be dependent on afferents for survival (Hardingham, 2009; Hetman and Kharebava, 2006; Ikonomidou et al., 2000).

The CM-PF in primates or the parafascicular (PF) in rodents is a major source of glutamatergic afferents to the matrix sub-compartment of the neostriatal mosaic (Fujiyama et al., 2006; Raju et al., 2006; Sadikot et al., 1992b). The PF also provides input to two major striatal interneuron subtypes implicated in HD: the cholinergic and PV positive interneurons (Lapper and Bolam, 1992; Rudkin and Sadikot, 1999; Sidibe and Smith, 1999). Ultrastructural studies in the heterozygous Q140 mouse model of HD suggest that early pathology of the thalamostriatal (TS) projection occurs prior to corticostriatal degeneration (Deng and Reiner, 2016; Deng et al., 2013). This early TS pathology is coexistent with degenerative changes in striatal cholinergic interneurons (Deng and Reiner, 2016).

Early pathological changes in TS circuitry may have important implications for motor symptoms of HD. In particular, the pathogenesis of dystonia, a common symptom of HD (Louis et al., 2000; Louis et al., 1999), has been linked to thalamostriatal and striatal cholinergic interneuron dysfunction in various dystonia and HD models (Deng and Reiner, 2016; Heinsen et al., 1996; Pappas et al., 2015; Picconi et al., 2006; Tanimura et al., 2016).

The R6/2 model, a transgenic mouse with approximately 125 CAG repeats in the N-terminal portion of the *mhtt* gene (Mangiarini et al., 1996) is a well-studied model of HD (Menalled et al., 2009; Samadi et al., 2013). This model was used to test our hypotheses on the effects of thalamostriatal deafferentation in HD because of its progressive and stereotypical motor phenotype and because it reproduces many of the morphological and biochemical changes of HD including striatal degeneration (Menalled et al., 2009; Samadi et al., 2013).

First Hypothesis

The PF of R6/2 mice undergoes degeneration as was demonstrated in *post-mortem* studies of the CM-PF in HD (Heinsen et al., 1996). To test this hypothesis, we quantified the number of

PF neurons in in R6/2 and WT mice at different timepoints throughout the R6/2 lifespan using unbiased cell counting techniques on immunohistochemically stained brain slices.

Second Hypothesis

Thalamostriatal (TS) inputs provide trophic support to vulnerable striatal neurons in R6/2 mice. To determine whether thalamic inputs to the striatum play a role in survival of striatal neurons, we assessed the effects of early TS lesions on striatal neurons in the R6/2 model by performing an unbiased stereological analysis of neuronal subpopulations at different timepoints after lesioning.

Third Hypothesis

Thalamostriatal-cholinergic interactions contribute to motor learning and action selection in the basal ganglia and dysfunction of either of these pathways will affect the motor phenotype of R6/2 mice. To test this hypothesis, a battery of behavioural assays which interrogated different aspects of motor function was performed in R6/2 and WT mice undergoing early unilateral PF lesions or early unilateral striatal cholinergic ablation using a cell-specific immunotoxin. The results of the experiments outlined in this section are presented in the following chapter.

Chapter 2: Thalamostriatal Degeneration Contributes to Cholinergic Interneuron Dysfunction and Dystonia in a Mouse Model of Huntington's Disease (2020). Acta Neur Comm.

2.1 Abstract

Huntington's disease (HD) is an autosomal dominant trinucleotide repeat disorder characterized by choreiform movements, dystonia and striatal neuronal loss. Amongst multiple cellular processes, abnormal neurotransmitter signalling and decreased trophic support from glutamatergic cortical afferents are major mechanisms underlying striatal degeneration. Recent work suggests that the thalamostriatal (TS) system, another major source of glutamatergic input, is abnormal in HD although its phenotypical significance is unknown. We hypothesized that TS dysfunction plays an important role in generating motor symptoms and contributes to degeneration of striatal neuronal subtypes. Our results using the R6/2 mouse model of HD indicate that neurons of the parafascicular nucleus (PF), the main source of TS afferents, degenerate at an early stage. PF lesions performed prior to motor dysfunction or striatal degeneration result in an accelerated dystonic phenotype and are associated with premature loss of cholinergic interneurons. The progressive loss of striatal medium spiny neurons and parvalbumin-positive interneurons observed in R6/2 mice is unaltered by PF lesions. Early striatal cholinergic ablation using a mitochondrial immunotoxin provides evidence for increased cholinergic vulnerability to cellular energy failure in R6/2 mice, and worsens the dystonic phenotype. The TS system therefore contributes to trophic support of striatal interneuron subtypes in the presence of neurodegenerative stress, and TS deafferentation may be a novel cell non-autonomous mechanism contributing to the pathogenesis of HD. Furthermore, behavioural experiments demonstrate that the TS system and striatal cholinergic interneurons are key motor-network structures involved in the pathogenesis of dystonia. This work suggests that treatments aimed at rescuing the TS system may preserve important elements of striatal structure and function and provide symptomatic relief in HD.

2.2 Introduction

Huntington's disease (HD) is a progressive autosomal dominant neurodegenerative disorder characterized by choreiform movements, dystonia and psychiatric symptoms (Louis et al., 1999; Vonsattel and DiFiglia, 1998). HD is caused by an abnormal expansion of CAG trinucleotides in exon 1 of the *huntingtin* gene (*mhtt*) with larger numbers of repeats leading to earlier age of onset and more severe symptoms (1993). Despite ubiquitous expression of *mhtt*, medium spiny projection neurons (MSNs) of the striatum are a major target for degeneration (Vonsattel and DiFiglia, 1998). Interneurons, which comprise a small proportion of all striatal neurons, were initially thought to be spared in HD (Ferrante et al., 1987a). However recent work suggests that the density of GABAergic parvalbumin (PV) positive (Reiner et al., 2013) and cholinergic interneuron subtypes are reduced in HD (Massouh et al., 2008) with relative sparing of other interneuron groups (Cicchetti and Parent, 1996; Massouh et al., 2008; Reiner et al., 2013). Multiple pathophysiological mechanisms may explain the predilection for striatal neuronal loss including: hyperexcitability, loss of afferent-derived trophic support, immune cell activation, and diverse intracellular signaling abnormalities (Crotti et al., 2014; Dau et al., 2014; Fernandes et al., 2007; Gladding et al., 2012; Li et al., 2003; Nguyen et al., 2016; Stack et al., 2007; Zhang et al., 2008b; Zuccato et al., 2005).

Loss of afferent-mediated trophic support contributes to neuronal loss in common neurodegenerative diseases, including Alzheimer's disease (Coyle et al., 1983; Teipel et al., 2018) and Parkinson's disease (Braak et al., 2003; Halliday et al., 1990). Trophic support from the major glutamatergic striatal afferent systems may also play an important role in HD. *In vivo* imaging and autopsy studies suggest that the cerebral cortex atrophies in prodromal HD along with the striatum, and this atrophy is severe by late stages (Rosas et al., 2008; Vonsattel and DiFiglia, 1998).

Recent MRI studies indicate that the thalamus also undergoes significant atrophy in early disease (Kassubek et al., 2005). *Post-mortem* analysis indicates that the posterior intralaminar thalamus, or centromedian-parafascicular (CM-PF) complex, is an important target for degeneration in HD (Heinsen et al., 1996).

The CM-PF in primates or the parafascicular (PF) in rodents is a major source of glutamatergic afferents to the striatum, specifically targeting MSNs of the matrix sub-compartment of the neostriatal mosaic (Fujiyama et al., 2006; Raju et al., 2006; Sadikot et al., 1992b). The PF also provides dense input to two major striatal interneuron subtypes implicated in HD: the cholinergic and PV positive interneurons (Lapper and Bolam, 1992; Rudkin and Sadikot, 1999; Sidibe and Smith, 1999). Recent ultrastructural studies in the heterozygous Q140 mouse model of HD suggest early pathology in the thalamostriatal (TS) projection prior to corticostriatal degeneration (Deng and Reiner, 2016; Deng et al., 2013). Further experiments show coexistent ultrastructural pathology of striatal cholinergic interneurons at early time-points in Q140 mice (Deng and Reiner, 2016).

In order to determine whether thalamic inputs to the striatum play a critical role in survival of striatal neurons and in development of motor dysfunction in HD, we assessed the structural and functional effects of early PF lesions in an animal model of HD. We used the R6/2 model, a transgenic mouse with approximately 125 CAG repeats in the N-terminal portion of the *mhtt* gene (Mangiarini et al., 1996). The R6/2 mouse is a well-studied model and reproduces many of the motor and morphological features of HD (Samadi et al., 2013). Our results provide evidence for early degeneration of PF neurons prior to striatal neuron loss in the R6/2 model. Early lesions of the TS in R6/2 mice result in an acceleration of clasping movements suggesting worsened dystonic behaviour. PF lesions do not accelerate the time course of progressive loss of spontaneous

locomotion in an open field during the R6/2 lifespan. PF lesioned mice regardless of genotype show decreased exploration using the contralateral forelimb. Morphological analysis indicates that PF lesions do not alter the extent of degeneration of striatal projection neurons and PV neurons in R6/2 mice. In contrast, TS lesions in R6/2 mice lead to early degeneration of striatal cholinergic neurons. Finally, early unilateral striatal cholinergic ablation in R6/2 mice using cell-specific immunotoxins also leads to an increase in clasping suggesting an important link between TS inputs to cholinergic neurons and dystonia in HD.

2.3 Materials and Methods

2.3.1 Animals

The behavioural experiments were performed using R6/2 mice and WT littermate mice from a colony maintained at the Facility for Neurological Disease Models of the Montreal Neurological Institute. Ovarian transplanted R6/2 females were obtained from a line maintained at The Jackson Laboratory and were crossed with males of the C6CBA background. CAG repeat lengths were sequenced and found to be between 119-125 for R6/2 mice and normal for WT littermates.

2.3.2 Surgery and Lesion Verification

All surgical procedures were performed in accordance with the Standard Operating Procedures (SOPs) for stereotaxic mouse surgery at McGill University. Twenty-eight day-old mice were anaesthetized using a ketamine and xylazine (Rompun, Bayer, USA) cocktail**.** Stereotactic lesions were made at coordinates corresponding to the PF (Bregma -2.20 mm, -3.3mm below the cortical surface, and 0.6 mm lateral to midline) (Franklin and Paxinos, 2008). A loop-shaped retractable leucotome (Sadikot et al., 2005) was inserted to the level of the PF, deployed to a radius

of 0.5 mm, rotated twice, closed and then retracted. Sham-lesioned animals underwent the same procedure except the leucotome was inserted 2.5 mm beneath the cortical surface but not deployed. Lesions were verified on Nissl stain or Nissl-NeuN using the 4X objective and the extent of each lesion was analyzed on images captured on tiled images (StereoInvestigator (v10, Microbrightfield, USA). Mice with lesions that either crossed the midline or with large lesions extending beyond the PF into the ventral thalamus were excluded.

2.3.3 Saporin Injection and Verification of Striatal Effects

Use of anti-ChAT conjugated saporin toxins are well-described for selectively ablating cholinergic interneurons in the rodent striatum (Aoki et al., 2015). Using the same stereotactic techniques mentioned above, 28-day-old R6/2 and WT mice underwent unilateral, striatal injections with either anti-ChAT-saporin or anti-Rabbit IgG-saporin (ATS BIO, USA). The total volume and concentration of either saporin construct was the same (0.7 µL of 0.6 µg/µL solution). The approximate center of mass of the neostriatum was targeted (0.65 mm from Bregma, 2.6 mm from the cortical surface and 2.15 mm lateral to midline)(Franklin and Paxinos, 2008). The toxin was infused at a rate of 0.1 µL /minute using an automated system (Pump 11 Elite, Harvard Apparatus, USA) through a 5 µL syringe (Hamilton 700 series, USA). Histological sections were immunostained for ChAT protein and counterstained with cresyl violet allowing visualization of the needle tract, confirming injection placement in the neostriatum, and allowing unbiased stereological analysis of striatal cholinergic cell morphology.

2.3.4 Behavioural Studies

All behavioural testing was performed during the first five hours of the light phase in a standard 12-h light–dark cycle (Wahlsten, 2011). Tests were performed at 4, 6, 9 and 11 wks ± 1

day (**Appendix, Experimental Timeline**), with the open field and cylinder test at day 1, and the clasping test at day 2 (Wahlsten, 2011).

Spontaneous Locomotion in Open Field

Mice were placed in a four arena 50X50cm open field with infrared backlighting for one-hour (Samadi et al., 2013), and movements were videotaped using an overhead camera (Bailoo et al., 2010) and later analyzed using VideoTrack (Viewpoint, Montreal, Canada). Spontaneous voluntary locomotor activity was categorized as follows: inactivity or non-ambulatory movements (<1 cm/second), moderate speed (between 1 and 5 cm/second) or fast speed (>5 cm/second).

Vertical exploratory behaviour

Mice were placed in a plexiglass cylinder (diameter 20cm, height 30 cm) with two mirrors positioned behind the cylinder in order to ensure a 360-degree view of the animal's forelimb wall touches. The session was video-recorded and the number of vertical contacts on the cylinder wall with the right paw, left paw or both paws simultaneously were scored on frame by frame analysis with the viewer blind to operative status and genotype.

Clasping Score

A tail suspension or clasping test was used to assess the development of dystonic forelimb contractions previously documented in the R6/2 mouse (Samadi et al., 2013; Stack et al., 2007). Mice were suspended by the tail at a height of at least 30 cm, for three trials lasting 30-seconds each, while limb movements were videotaped. Clasping was defined as a retraction of a limb toward the body. In order to provide a semi-quantitative index of abnormal involuntary movements, clasping at each limb was graded as: none=0, mild=0.25, moderate= 0.5, severe = 0.75 by an observer blind to genotype. Clasping was rated as: "none" if the mouse did not retract

the limb towards the midline and "mild" if partial retraction of a limb occurred toward the midline but did not reach the midline, and the contraction was not sustained. "Moderate" clasping was a high-amplitude limb retraction to or beyond the midline that was not sustained, or partial limb retraction that was sustained for ≥5 consecutive seconds. "Severe" clasping was a high-amplitude limb retraction to or beyond the midline sustained for ≥5 seconds. The score for the forelimbs and hindlimbs was summed making the maximum score 3. The average value of all three clasping trials was analyzed.

2.3.5 Tissue Processing

R6/2 mice and WT mice were deeply anesthetized and perfused transcardially with 0.9% heparinized saline followed by 4% paraformaldehyde in phosphate buffer (4% PFA) (0.1M, pH 7.4) both at 4°C. Brains were removed, fixed in 4% PFA for 24 h then transferred to a phosphate buffered 30% sucrose solution for 24-48 hours. Brains were sectioned at 40 μm in the coronal plane with a freezing microtome. Free-floating sections were collected serially in six vials containing phosphate-buffered saline (PBS, 0.1M, pH 7.4). One set of sections was mounted out of distilled water onto glass slides, stained in 0.1% cresyl violet (Nissl stain) and coverslipped using Permount (Fisher Scientific, Whitby, ON, Canada). The remaining vials were immediately placed in buffered anti-freeze solution and stored at -20°C.

2.3.6 Immunohistochemistry

The following primary antibodies were used in these experiments: mouse anti- NeuN (Millipore, Etobicoke, Canada; MAB377, 1:1000), Rabbit anti-μ-opioid receptor (Immunostar, Hudson, USA; #24216; 1:8000), mouse anti-parvalbumin (Swant, Fribourg, Switzerland; #235; 1:5000) and rabbit anti-ChAT (Millipore; AB143; 1:600). Sections were removed from antifreeze,

rinsed six times in PBS, and then incubated for one hour in a blocking solution (10% bovine serum albumin (BSA), 0.3 % Triton-X, 0.1M PBS, pH 7.4). Next the sections were incubated in primary antibody in PBS containing 0.1% Triton-X and either 2% BSA or 5% NGS for 24-48 hours at 4°C. After washes in PBS, sections were incubated in the following biotinylated secondary antibodies: horse anti-mouse IgG (Vector Laboratories, Burlingame, California, USA; BA-2000; 1:200), goat anti-rabbit IgG (Vector Laboratories; BA-1000; 1:200). Sections were washed once more in PBS and then incubated for 1 hour in 1:100 ABC elite kit (PK6100, Vector Laboratories). Antibody binding was revealed using 0.05% 3,3'-diaminobenzidsine (D5905, Sigma-Aldrich, Oakville, ON, Canada) in TBS (pH 7.6) and hydrogen peroxide (0.01%). All slices were then mounted out of distilled water onto slides, counterstained with 0.1% cresyl violet and coverslipped using Permount (SP15, Fisher Scientific).

2.3.7 Unbiased Stereology

An unbiased stereological probe, the optical fractionator (Gundersen et al., 1999), was used to estimate the number of neurons in the areas of interest. The stereology apparatus consisted of a light microscope (BX40, Olympus, Japan) coupled with a video camera (DC200, DAGE, USA), motorized X–Y stage (BioPoint XYZ, LEP, USA), Z-axis indicator (MT12 microcator, Germany), and a computer running Stereo Investigator software (v11.06.2, Microbrightfield, USA). The neostriatum was delineated according to previously defined boundaries (Samadi et al., 2013) using the mouse brain atlas of Paxinos and Franklin (Franklin and Paxinos, 2008) and a 4X objective. Rostral and caudal limits were determined by the first and last coronal sections with visible caudate–putamen (Bregma 1.7 mm to -2.0 mm) (Franklin and Paxinos, 2008). Every sixth serial histological section within this zone was examined (240 μm intervals). The dorsal, medial and lateral limits of the neostriatum are well defined in the mouse brain atlas (Franklin and Paxinos,

2008). The ventral limit of the striatum at the post-commissural part is well delineated on Nissl stains. At the pre-commissural levels, we delimit the dorsal striatum from the nucleus accumbens with a line that extends from above the ventral most part of the lateral ventricle medially, to the tapered external capsule laterally, at an angle of 25–30◦ below the axial plane (Sadikot and Sasseville, 1997; Samadi et al., 2013). The PF was delineated using the same mouse brain atlas (Franklin and Paxinos, 2008) using a 10X objective. All sections with a clearly distinguishable PF were delineated (Bregma -2.0mm to -2.5 mm) (Franklin and Paxinos, 2008). Every other section within the PF reference range was examined (80 μm intervals).

Systematic random sampling of neurons was performed by randomly translating a grid onto the section of interest. At each intersection of grid lines an optical fractionator counting frame with exclusion lines was applied. A 150X150 μm grid size and a 60X60 μm counting frame was used for the PF neuron optical fractionator analysis (Gunderson CE (m=1) = 0.038 ± 0.001). A 300X300 μm grid size and a 25X25 μm counting frame was used for optical fractionator analysis in the neostriatum (Gunderson CE (m=1) = 0.029± 0.001). A 250X250 μm grid size and a 70X90 μm counting frame was used for the parvalbumin interneuron optical fractionator analysis (Gunderson CE (m=1) = 0.064± 0.001). A 175X175 μm grid size and a 70X90 μm counting frame was used for the cholinergic interneuron optical fractionator analysis (Gunderson CE (m=1) = 0.080 ± 0.003). All randomly assigned sample sites were then examined using a 100X objective (oil; numerical aperture, 1.3). Section thickness was assessed every ten counting sites using the Z-axis indicator (MT12 microcator, Germany). The top of the neuron was used as a unique identifier in all analyses. Neurons falling in the counting frame were counted only if they came into focus within a predetermined 8-μm-thick optical dissector positioned 1-μm above and below the surface of the mounted section as indicated by the Z-axis indicator. For the neostriatal mosaic analysis,

neurons were distinguished Nissl-stains based on cell diameter (>7 μm), and a lighter cytoplasm containing a dense nucleus (Samadi et al., 2013).

Neuron soma area and volume of the PF and striatum were estimated using four-ray nucleator probe (Moller et al., 1990) or the Cavalieri probe (Gundersen et al., 1999) respectively. For the Cavalieri probe, a grid of 40X40 μm squares was randomly translated over the delineated structures of interest and markers were placed at the intersection of grid lines that fell within the delineated structure. Estimates of the total number of neurons, soma area and Cavalieri volume were calculated by the Stereo Investigator software (v10, Microbrightfield, USA).

2.3.8 Statistical Analyses

Normality was assessed prior to performing comparative tests using the Shapiro-Wilks test. An analysis of variance (ANOVA) was performed on normal data using the aov function in R (Team, 2018). *Post hoc* analysis of normal data consisted of a two-tailed, paired or unpaired t-test based on whether the samples were dependent or independent respectively. *Post hoc* tests on normal data were corrected for multiple comparisons using *Tukey's Honestly Significant Difference* test (HSD). A non-parametric ANOVA was performed on non-normally distributed data or ordinal data using the art function from the 'ARTool' package (Wobbrock et al., 2011) in R. *Post hoc* analysis for non-parametric data included a two-tailed Mann-Whitney U-test or a Wilcoxon signed-rank test for independent and dependent samples respectively. Non-parametric *post hoc* tests were corrected for multiple comparisons using the *Bonferroni* correction. For behavioural tests, the main factors of the ANOVA were time as a within-subject factor, and genotype and lesion status as between-subject factors. The main ANOVA factors for morphological studies, genotype and lesion status were analyzed as independent groups. All data

are expressed as averages ± standard error of the mean (SEM). The SEM is represented graphically as error bars. P-values ≤0.05 were considered significant.

2.4 Results

2.4.1 The PF Degenerates in the R6/2 Model of HD

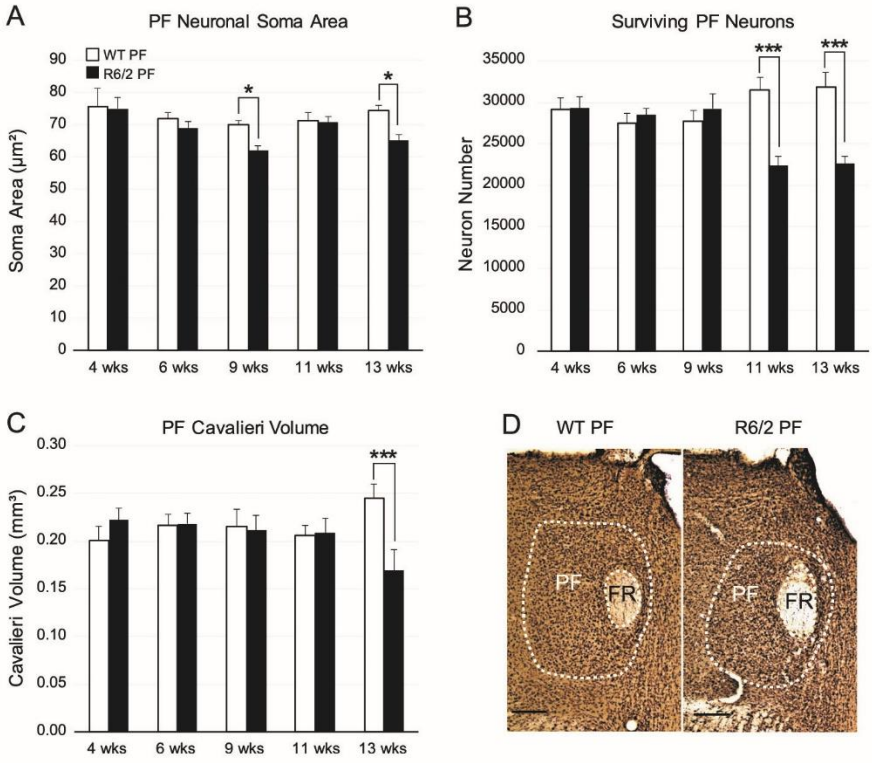


Fig. 1 Morphology of the PF nucleus over the R6/2 mouse lifespan compared to WT controls. (**a**) There is a significant decrease in neuronal area at 9 ($p = 0.03$) and 13 wks ($p = 0.01$) in R6/2 compared to WT mice, based on analysis with the nucleator, an unbiased stereology probe. (**b**) Loss of PF neurons in R6/2 compared to WT mice at 11 wks ($p = 0.0001$) and 13 wks ($p = 0.0002$) determined using the optical fractionator, an unbiased stereology probe. (**c**) Reduction in PF volume is noted at 13 wks compared to WT mice determined using the Cavalieri probe ($p = 0.0007$). (**d**) Photomicrographs of NeuN/Nissl stained coronal sections outlining the PF nucleus in WT and R6/2 mice at 13 wks. Scale bar: 250 µm. The data sets were analyzed using a two-way between subject ANOVA and a Tukey HSD post hoc test: *$p < 0.05$, *** $p < 0.001$. For all panels of Fig. 1, 4 week: WT ($n = 3$), R6/2 ($n = 4$); 6 week: WT (n = 4), R6/2 ($n = 5$); 9 week: WT ($n = 4$), R6/2 ($n = 4$), 11 week: WT ($n = 6$), R6/2 ($n = 4$); 13 week: WT ($n = 5$), R6/2 ($n = 4$). Abbreviations: FR = Fasciculus Retroflexus, PF = Parafascicular Nucleus

To determine if the PF is susceptible to degeneration in HD, we quantified neuron numbers and soma size in the PF throughout the lifespan of the R6/2 mouse model using unbiased stereology. Application of the nucleator probe demonstrated that soma size of PF neurons was reduced in R6/2 mice compared to WT at 9 and 13 weeks (wks) (Fig. 1a, w (43) = 0.954, p = 0.086,

F(GenotypeXTime)4,33 = 1.45, p = 0.24, F(Time)4,33 = 3.85, p = 0.01, F(Genotype)1,33 = 8.46, p = 0.006, post hoc comparison: 9 wks p = 0.03 and 13 wks p = 0.01). Analysis using the optical fractionator probe revealed a significant 29% decrease in the number of PF neurons in R6/2 mice at 11 wks com- pared to WT (Fig. 1b, w (43) = 0.972, p = 0.37, F(GenotypeXTime)4,33 = 7.65, p = 0.00018, post hoc: 11 wks p = 0.00014; 13 wks p = 0.00015). Neuronal degeneration progressed at later timepoints, and by 13 wks the Cavalieri volume estimate of the PF was 31% smaller in R6/2 mice compared to WT at 13 wks (Fig. 1c-e, w (43) = .948, p = 0.05, F(GenotypeXTime)4,33 = 3.34, p = 0.021; post hoc p = 0.0007). In summary, the main source of TS projections, the PF, shows an early reduction in neuronal size in R6/2 mice at 9 wks, followed by progressive neuronal loss at 11 and 13 wks of age.

2.4.2 The Effect of PF Lesions on Motor Behavior in R6/2 and WT Mice

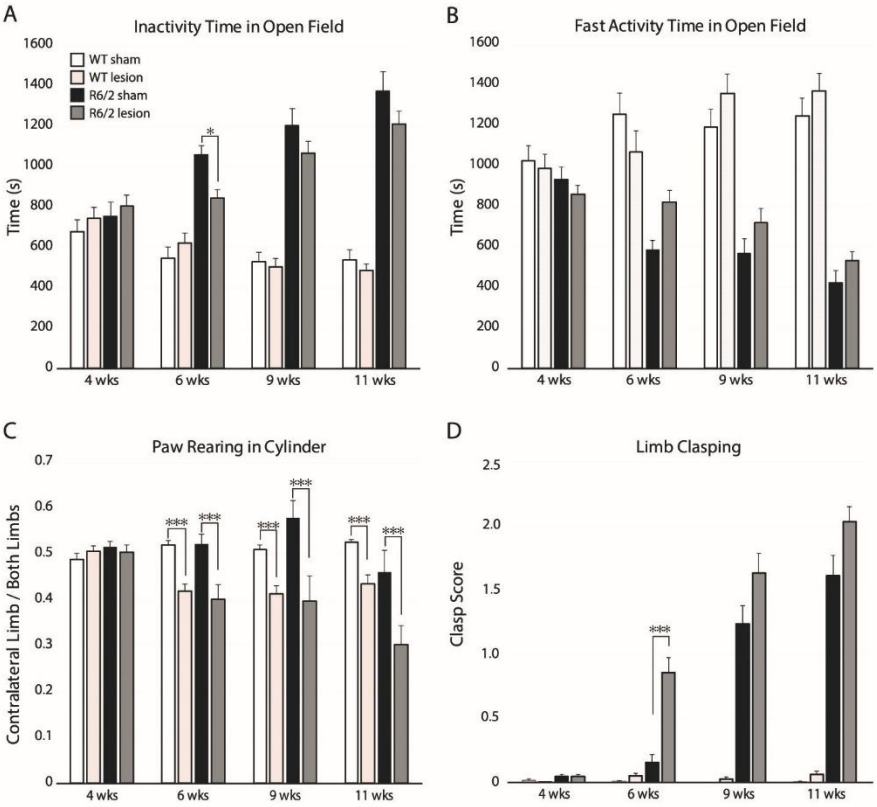


Fig. 2 Characterization of motor behavior following unilateral PF lesions in R6/2 and WT mice. Time spent at rest (**a**) or moving rapidly (**b**) during a one-hour open field session demonstrating a transient decrease in rest time at 6 wks in R6/2 mice following PF lesions compared to sham-lesions ($p = 0.01$), that is not sustained at later time-points. (**c**) The cylinder test assessing voluntary paw reaching motor behaviour shows a persistent decrease in contralateral limb use in both WT and R6/2 mice after PF lesions (6 wks $p = 0.00004$, 9 wks $p = 0.00004$, 11 wks $p = 0.00008$). (**d**) A significant increase in dystonic clasping is noted in PF lesioned R6/2 compared to sham treated R6/2 mice at 6 wks ($p = 0.00008$). A 3-way non-parametric ANOVA was applied to each data set, followed by a Bonferroni post hoc correction; *$p < 0.05$, ***$p < 0.001$. For Fig. 2 a-d: WT sham: $n = 18$, WT lesion: $n = 17$, R6/2 sham: $n = 15$, R6/2 lesion: $n = 22$

The open field test assesses spontaneous voluntary loco- motor activity [39]. To determine the effect of PF lesions on locomotor activity, R6/2 and WT mice were placed in an open field for one hour at 4, 6, 9 and 11 wks. In keeping with previous studies [34], we found a progressive increase in inactivity time over the R6/2 mouse lifespan starting at 6 wks in both sham-lesioned and lesioned groups compared to their respective WT groups (Fig. 2a, f (TimeXGenotypeXLesion)3201 = 2.82, p = 0.04, post hoc all p < 0.02 for sham R6/2 mice vs. WT sham as of the 6-week time- point). Lesioned R6/2 mice spent significantly less time resting at 6

wks compared to sham R6/2 mice ($p = 0.01$), but not at later time-points. Periods of fast movement time reflected inactivity time, with progressive decrease in loco- motion in R6/2 compared to WT mice. There was a non- significant ($p = 0.07$) trend to increased locomotion at 6 wks in lesioned compared to sham-lesioned R6/2 mice (Fig. 2b, f (TimeXGenotypeXLesion)$3201 = 4.64$, $p = 0.004$). Thus, PF lesioned R6/2 mice progress to the same hypokinetic state with poverty of spontaneous voluntary movement as sham-lesioned counterparts.

The cylinder test assesses exploratory vertical paw reaching limb asymmetry, a complex voluntary behav- iour requiring spatial sensorimotor coordination [45]. The number of paw touches on the walls of a cylinder were quantified during a five-minute session in PF lesioned and sham-lesioned WT and R6/2 mice at 4, 6, 9 and 11 wks. A significant reduction in the percent of contralateral limb touches occurs at all post-operative ages after PF lesions in both WT and R6/2 mice com- pared to sham counterparts (Fig. 2c, f (TimeXGenotypeXlesion)$3167 = 0.43$, $p = 0.73$, F(TimeXLesion)$3167 = 13.4$, $p < 0.00001$; F(TimeXGenotype)$3167 = 4.82$, $p = 0.003$, post hoc all $p < 0.001$). Thus, both R6/2 and WT mice preferentially explored vertical cylinder space with the ipsilateral limb following PF lesions.

The tail suspension test or clasping test is a widely used method for eliciting dystonic movements in HD and dystonia mouse models [34, 46, 47]. To determine if PF lesions affect the clasping phenotype, mice were tested prior to lesions and at three post-operative time- points. R6/2 mice had a worsening in dystonic clasping behaviour with aging in both sham and lesion groups with a significant increase in limb clasping in R6/2 mice at 6 wks following PF lesions compared to sham- lesioned R6/2 mice (Fig. 2d, f (TimeXGenotypeXLesion)$3210 = 26.63$, $p < 0.00001$, post hoc: 6-week R6/2 sham vs 6-week R6/2 lesion $p = 0.00008$). Virtually none of the WT mice

exhibited clasping and PF lesions did not induce dystonic behaviour in this group. Thus, PF lesions significantly worsen the clasping phenotype in R6/2 mice.

2.4.3 Striatal Morphology After Early PF Lesions

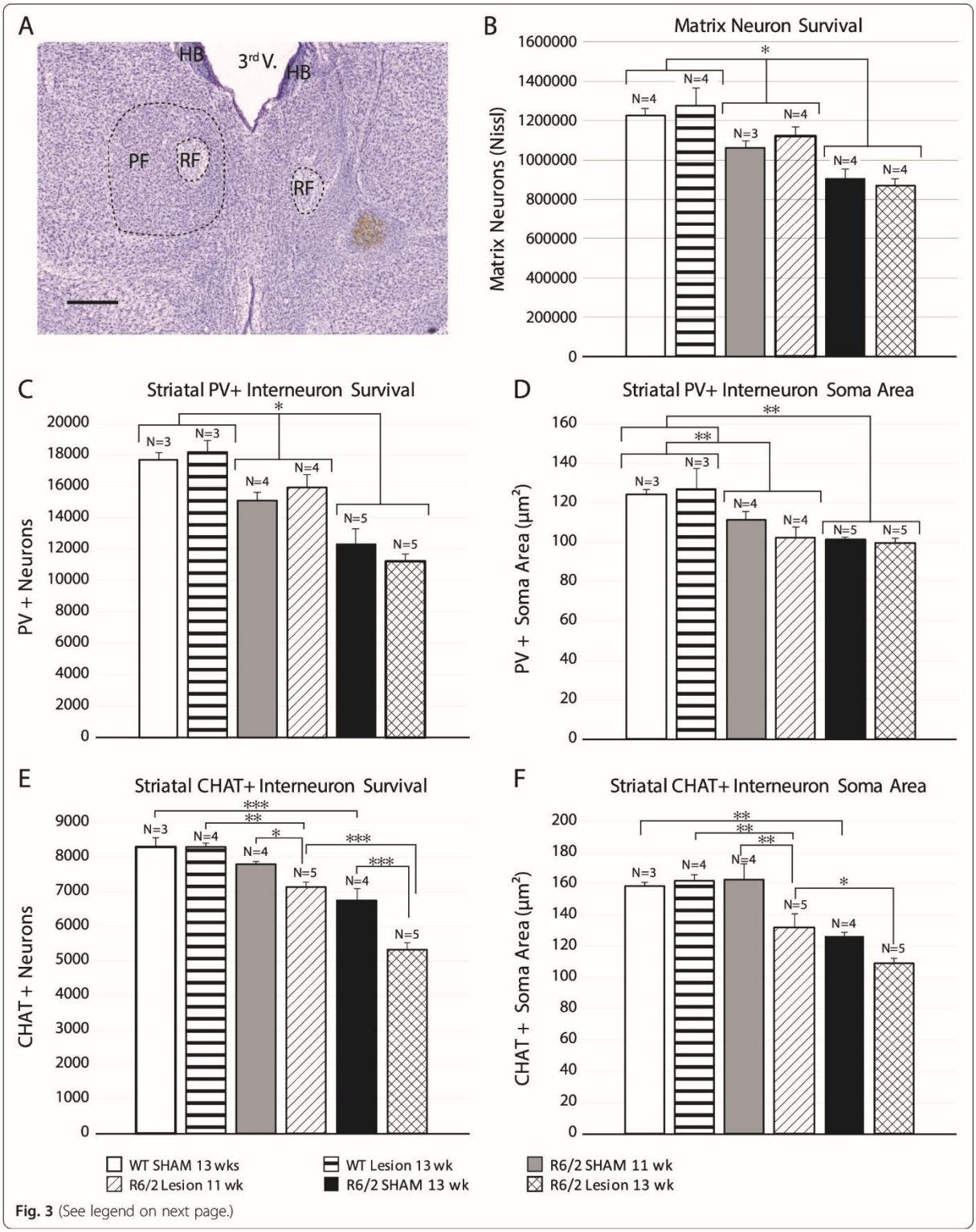


Fig. 3 (See legend on next page.)

(See figure on previous page.)
Fig. 3 Analysis of number and size of striatal neuron subtypes after PF lesions. These subtypes are known to receive PF input. (**a**) A typical PF lesion in an R6/2 mouse (scale bar: 0.5 mm). (**b**) Unbiased stereology using the optical fractionator reveals loss of matrix neurons in R6/2 mice at 11 wks with further loss at 13 wks. PF lesions do not alter neuron number in the striatal matrix compartmentin either WT or R6/2 mice. (**c, d**) Unbiased stereology analysis of striatal PV+ interneurons using the optical fractionator (**c**) or the nucleator (**d**) reveals progressive cell loss and atrophy in R6/2 vs. WT mice, with no effect of PF lesions. (**e, f**) Optical fractionator cell counts (**e**) and nucleator-derived soma area (**f**) of ChAT + interneurons show earlier, more severe and progressive cell loss and atrophy in PF lesioned R6/2 mice compared to sham-lesioned R6/2 mice at both 11 wks and 13 wks. Morphology of ChAT + interneurons is not altered by PF lesions in WT mice. Scale bar: 250 μm. A 2-way between subject ANOVA was applied to each data set followed by a Tukey HSD post hoc test; * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$. Abbreviations: PF = Parafascicular, FR = Fasciculus Retroflexus, 3rd V. = 3rd Ventricle, HB = Habenula

Previous work in R6/2 mice using unbiased stereology on Nissl stained sections demonstrates that significant striatal cell loss and atrophy occurs at 11 and 13 wks [34]. To determine if the PF has a trophic role for striatal neurons faced with degenerative stress in HD, we quantified the number and soma size of striatal neurons at 11 and 13 wks following PF lesions at 4 wks of age. Since the posterior intralaminar nuclei preferentially afferent the matrix compartment of the striatal mosaic [25–27], neurons of the striosome and matrix compartments were analyzed separately using μ-opiate receptor (MOR) as a marker of striosomes.

The number of matrix neurons in R6/2 mice undergoes a significant and progressive reduction over time com- pared to WT mice, and there is no effect of PF lesions (Fig. 3b, w(23) = 0.967, p = 0.72, F(GenotypeXLesion)2,17 = 0.49, p = 0.62, F(Lesion)1,17 = 0.27, p = 0.61, F(Genotype)2,17 = 23.45, p = 0.00001. post hoc: WT vs 11 wks R6/ 2, p = 0.03, WT vs 13 wks R6/2, p = 0.0002, 11 wks R6/2 vs 13 wks R6/2, p = 0.003). As with neuronal counts, there is a significant reduction in the soma area in R6/2 mice at 11 and 13 wks compared to WT, with no effect of PF lesions (Appendix: Figure S1, W(23) = 0.981, p = 0.90, F(GenotypeXLesion)2,17 = 2.82, p = 0.09, F(Genotype)2,17 =48.78, p < 0.00001, F(Lesion)1,17 = 1.13, p = 0.30).

The number of neurons in striosomes is significantly reduced in R6/2 mice compared to WT at 13 wks, but not at 11 wks. PF lesions did not alter striosome neuron number in any group (Appendix: Figure S2, W(23) = 0.986, p = 0.63 F(GenotypeXLesion)2,17 = 0.31, p = 0.74,

F(Lesion)1,17 = 1.00, p = 0.33, F(Genotype)2,17 = 10.65, p = 0.001). Striosomal soma area was significantly decreased in 11 and 13 week old R6/2 mice compared to WT but there was no significant effect of PF lesions (Appendix: Figure S3, W(23) = 0.963, p = 0.5362, F(GenotypeXLesion)2,17 = 0.69, p = 0.51, F(Genotype)2,17 = 43.06, p < 0.00001, F(Lesion)1,17 = 4.97, p = 0.04). In summary, striatal neuronal loss occurs in R6/2 mice with onset in the matrix compartment, followed by loss in both compartments at late time-points. However, striatal projection neuron loss is not altered in either compartment by early PF lesions.

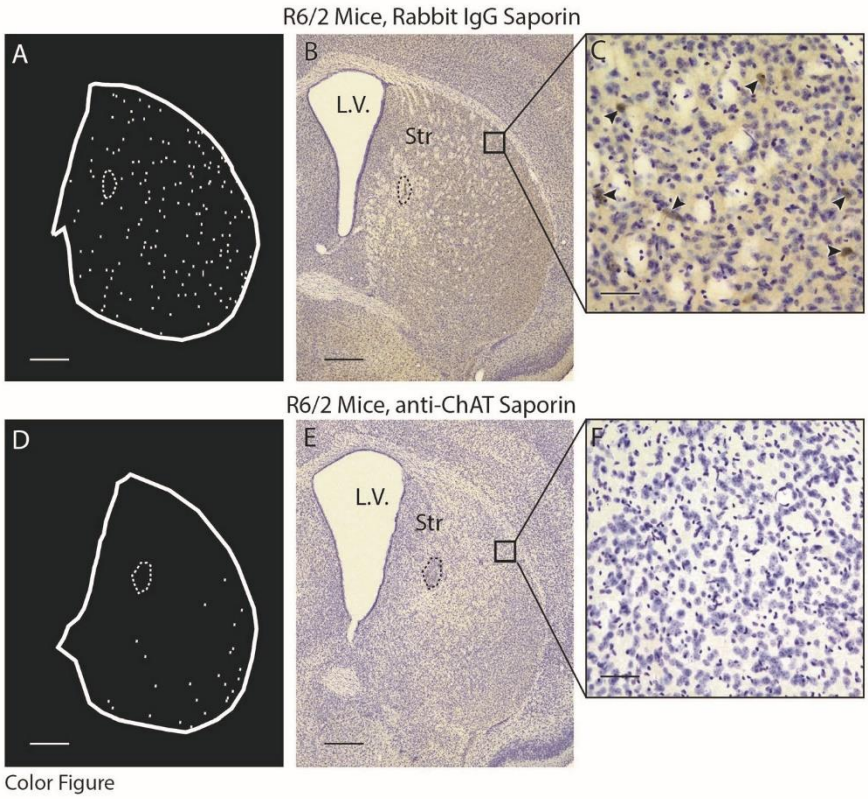
To determine if TS afferents sustain PV striatal interneurons in the face of degenerative stress [48], neurons were quantified by unbiased stereology in R6/2 mice at 11 and 13 wks following PF lesions at 4 wks. There is a significant and progressive reduction in the number of PV+ neurons in both PF lesioned and sham-lesioned R6/2 mice at 11 and 13 wks compared to WT. However, PF lesions did not alter PV+ interneuron number (Fig. 3c, w (24) = 0.965, p = 0.56, F(GenotypeXLesion)2,18 = 0.97, p = 0.40, F(Lesion)1,18 = 0.017, p = 0.90, F(Genotype)2,18 = 34.36, p < 0.00001, post hoc: WT vs 11 wks R6/2, p = 0.02; WT vs 13 wks R6/2, p = 0.0002; 11 wks R6/2 vs 13 wks R6/2, p = 0.0003). Furthermore, PV+ soma area was reduced with age in R6/2 mice, without an additional effect of PF lesions (Fig. 3d, w (24) = 0.978, p = 0.86, F(GenotypeXLesion)2,18 = 0.86, p = 0.44; F(Lesion)1,18 = 0.42, p = 0.53, F(Genotype)2,18 = 16.72, p = 0.00008, post hoc: WT vs 11 week p = 0.003; WT vs 13 week R6/2 p = 0.0002). Thus, PV+ cells undergo progressive atrophy and cell loss in R6/ 2 mice at late stages, but this degeneration is not affected by TS deafferentation.

The TS is the predominant source of glutamatergic input to striatal cholinergic interneurons [30, 49–52] and modulates their physiology [53]. To determine if loss of trophic support from the TS system alters striatal cholinergic neuron survival in R6/2 mice, we quantified choline acetyl-

transferase (ChAT) + cell number and soma size at 11 and 13 wks following PF lesions at 4 wks. Compared to WT mice, sham-lesioned R6/2 mice show a relative resistance to cholinergic neuron loss compared to MSNs or PV+ interneurons, with detectable reduction in numbers occurring at 13 wks, but not at 11 wks (Fig. 3e, w (25) = 0.982, p = 0.92, F(GenotypeXLesion)2,19 = 5.81, p = 0.01, post hoc: WT sham vs 13 wks R6/2 sham p = 0.0005; 11 wks R6/2 sham vs 13 wks R6/2 sham p = 0.005). PF lesioned R6/2 mice show accelerated loss of cholinergic neurons by 11 wks compared to both PF lesioned WT mice and sham-treated R6/2 mice, with further neuronal loss noted in PF lesioned R6/2 mice at 13 wks (Fig. 3e, post hoc: WT lesion vs 11 week R6/2 lesion, p = 0.002; WT lesion vs 13 wks R6/2 lesion, p = 0.0001; 11 wks R6/2 lesion vs 13 wks R6/2 lesion, p = 0.0002; 11 week R6/2 lesion vs 11 wks R6/2 sham p = 0.03, 13 wks R6/2 lesion vs 13 wks R6/2 sham, p = 0.0002). PF lesions did not induce cholinergic cell loss in WT mice. Thus, cholinergic degeneration occurs at a very late time- point in sham R6/2 mice (13 wks) while PF lesioned R6/2 mice show an accelerated cholinergic cell loss at 11 wks that progresses at 13 wks.

Cholinergic soma area is also reduced at 13 wks in R6/2 compared to WT mice. PF lesions in R6/2 mice are associated with a further decrease in soma area beginning at 11 wks, which progresses by 13 wks (Fig. 3f, w (25) = 0.98, p = 0.90, F(GenotypeXLesion)2,19 = 3.36, p = 0.05, post hoc: WT sham vs 13 wks R6/2 sham p = 0.003; 11 wks R6/ 2 sham vs 13 wks R6/2 sham p = 0.02; WT lesion vs 11 wks R6/2 lesion, p = 0.009; WT lesion vs 13 wks R6/2 lesion p = 0.0002; 11 wks R6/2 lesion vs 13 wks R6/2 lesion p = 0.04). In summary, there is more severe cholinergic neuron atrophy in PF lesioned R6/2 mice compared to sham-lesioned R6/2 mice at 11 wks (p = 0.003), with further atrophy noted at 13 wks.

2.4.4 Cholinergic Interneuron Loss Following Intrastriatal Injection of Immunotoxin



Color Figure

Fig. 4 Cholinergic interneuron distribution in representative striatal sections of R6/2 mce 7 wks after intrastriatal injection of either anti-ChAT-saporin immunotoxin or Rabbit IgG-saporin (control). (**a, d**) Contours demonstrating the location of ChAT+ profiles in coronal sections of neostriatum. (**b, c, e, f**) Photomicrographs of corresponding sections immunohistochemically stained for ChAT and Nissl after treatment with either Rabbit IgG-saporin (**b**, 4X; **c**, 20X) or anti-ChAT-saporin (**e**, 4X; **f**, 20X). Arrowheads demonstrate ChAT+ striatal neurons. Stippled contours indicate the hemosiderin artifact from the injection site. Squares in (**b**) and (**e**) represent the area magnified in (**c**) and (**f**) respectively. Scale bars: A, B, D, E = 500 µm, C, F = 100 µm. Abbreviations: L.V. = Lateral Ventricle, Str = Striatum

To determine if cholinergic neuron loss is associated with changes in motor phenotype, anti-ChAT conjugated saporin toxins were used to selectively ablate striatal cholinergic interneurons. Mice received intrastriatal injections of either anti-ChAT-saporin or Rabbit IgG-saporin at 4 wks of age and were euthanized at 11 wks (Fig. 4). There was a large reduction in the number of cholinergic neurons assessed using unbiased stereology in both R6/2 and WT mice injected with anti-ChAT-saporin (Fig. 5, W(14) = 0.895 p = 0.09, F(GenotypeXSaporin)1,10 = 8.08, p= 0.02; post hoc: anti-ChAT- saporin WT vs Rabbit-IgG -saporin WT: p = 0.0003, anti-ChAT-saporin R6/2 vs Rabbit IgG-saporin R6/2: p = 0.0002). The reduction in cholinergic

number in anti-ChAT -saporin injected R6/2 mice was greater than in anti-ChAT-saporin injected WT mice (p = 0.004).

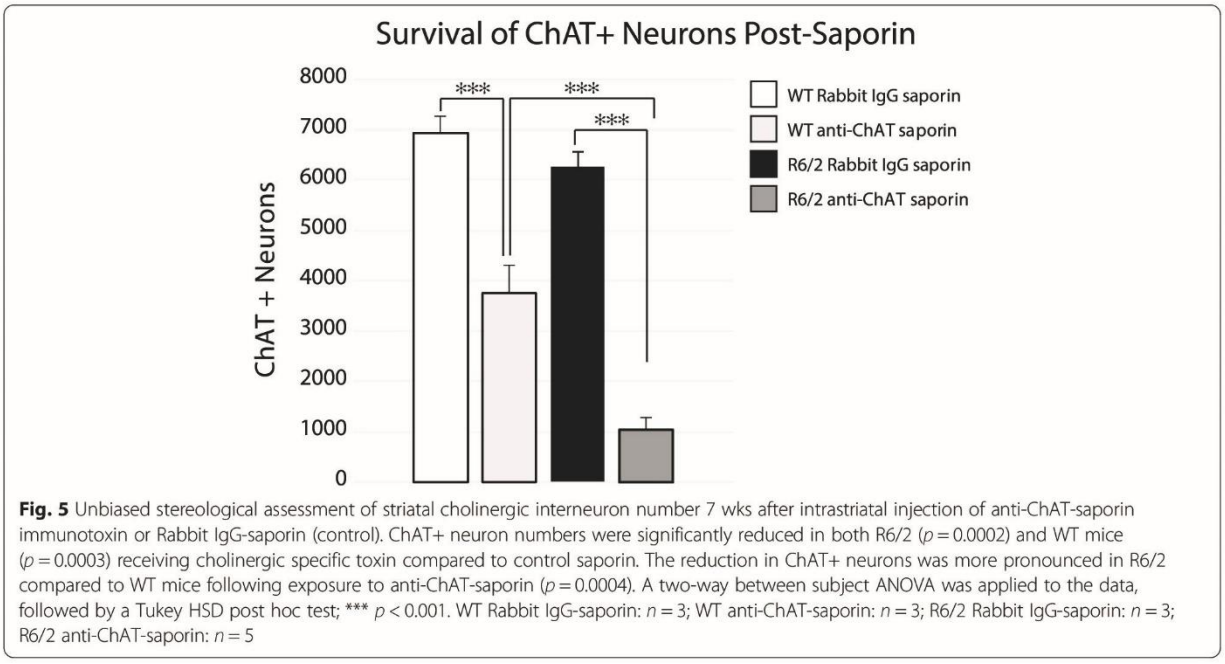


Fig. 5 Unbiased stereological assessment of striatal cholinergic interneuron number 7 wks after intrastriatal injection of anti-ChAT-saporin immunotoxin or Rabbit IgG-saporin (control). ChAT+ neuron numbers were significantly reduced in both R6/2 ($p = 0.0002$) and WT mice ($p = 0.0003$) receiving cholinergic specific toxin compared to control saporin. The reduction in ChAT+ neurons was more pronounced in R6/2 compared to WT mice following exposure to anti-ChAT-saporin ($p = 0.0004$). A two-way between subject ANOVA was applied to the data, followed by a Tukey HSD post hoc test; *** $p < 0.001$. WT Rabbit IgG-saporin: $n = 3$; WT anti-ChAT-saporin: $n = 3$; R6/2 Rabbit IgG-saporin: $n = 3$; R6/2 anti-ChAT-saporin: $n = 5$

Soma size of the surviving cells was not different among the four groups (Appendix: Figure S4, W(14) = 0.944 p = 0.4754, F(GenotypeXSaporin)1,10 = 0.46, p = 0.51, F(Genotype)1,10 = 3.28, p = 0.10, F(Saporin)1,10 = 0.23, p = 0.64). Thus, an intrastriatal anti-ChAT-saporin injection was effective in eliminating a substantial proportion of striatal cholinergic neurons in both WT and R6/2 mice. Moreover, striatal cholinergic neurons were significantly more vulnerable to the cholinergic immunotoxin in R6/2 compared to WT mice.

2.4.5 The Effect of Striatal Cholinergic Ablation on Motor Behavior in R6/2 and WT Mice

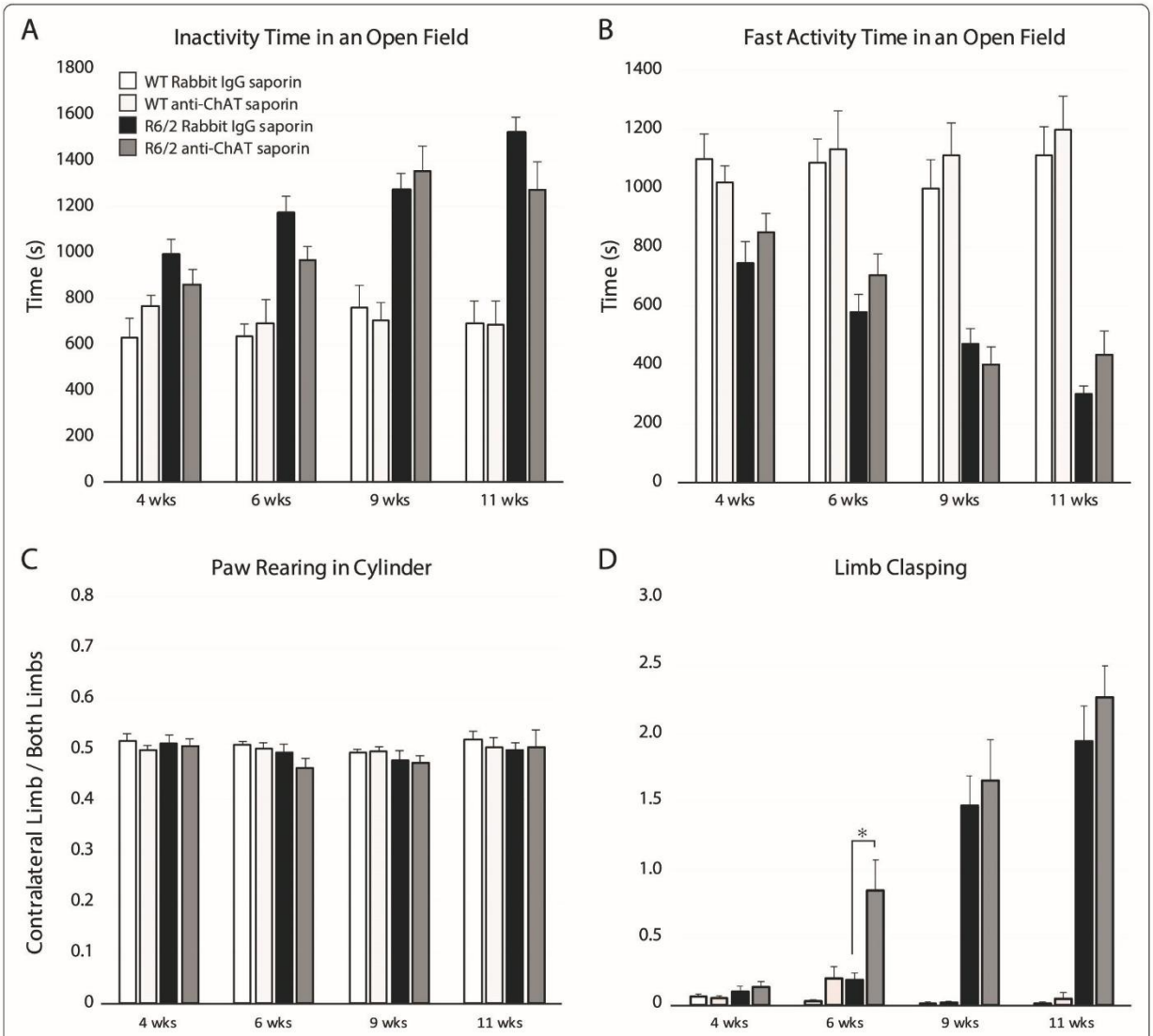


Fig. 6 Characterization of motor behavior following unilateral striatal cholinergic ablation in R6/2 and WT mice. (**a**) Time spent at rest and (**b**) on rapid movements during a one-hour open field session demonstrating a decrease in spontaneous voluntary locomotor activity over time in R6/2 mice with no significant effect of anti-ChAT- or Rabbit-IgG-saporin injections. (**c**) Cylinder test assessing limb use asymmetry shows no effect of saporin injection on paw preference. (**d**) Evaluation of dystonia shows an increase in limb clasping at 6 wks in anti-ChAT saporin treated R6/2 mice compared to control-saporin treated R6/2 mice ($p = 0.04$). A 3-way non-parametric ANOVA was applied to each data set, followed by a post hoc Bonferroni correction; *$p < 0.05$. For all panels of Fig. 6, WT Rabbit-IgG-saporin: $n = 9$, WT anti-ChAT-saporin: $n = 10$, R6/2 Rabbit-IgG-saporin: $n = 12$, R6/2 anti-ChAT-saporin: $n = 11$

To determine the effect of striatal cholinergic ablation on spontaneous locomotor activity, anti-ChAT-saporin or Rabbit-IgG-saporin injected R6/2 and WT mice were placed in an open field for one hour at 4, 6, 9 and 11 wks. Compared to WT mice, R6/2 mice showed a decrease in

spontaneous locomotion as revealed by increased time spent resting in an open field at 6,9 and 11 wks, with no effect of anti-ChAT-saporin injection (Fig. 6a, f (GenotypeXSaporinXTime) 3120 = 2.50, $p = 0.06$, F(GenotypeXTime) 3120 = 13.8, $p < 0.0001$, post hoc: R6/2 vs WT after 6 wks, all $p < 0.005$). Decreased time spent on fast activity reflected the rest time results (Fig. 6b F(TimeXGenotypeXSaporin) 3120 = 2.77, $p = 0.04$; post hoc WT vs R6/2 after 6 weeks all $p < 0.005$, post hoc: all comparisons within genotype for Rabbit-IgG-saporin vs anti-ChAT- saporin were not significant). Thus, striatal cholinergic ablation does not affect spontaneous voluntary locomotor behaviour of R6/2 and WT mice.

The cylinder test assessing limb use asymmetry while performing vertical exploration revealed no differences in paw reaching for either of the saporin treated groups (Fig. 6c, w (60) = 0.974 $p = 0.2354$, F(GenotypeXSaporinXTime)3142 = 0.33, $p = 0.80$, F(GenotypeXSaporin)1142 = 0.001, $p = 0.80$, F(TimeXSaporin)3142 = 0.14, $p = 0.94$, F(TimeXGenotype)1142 = 0.51, $p = 0.67$). Thus, unilateral striatal cholinergic ablation does not induce a paw preference during voluntary movement in either R6/2 or WT mice.

To determine if cholinergic ablation affected the development of the dystonic phenotype in R6/2 mice, limb clasping was assessed at 4, 6, 9 and 11 weeks. Dystonic clasping behaviour with aging worsened in R6/2 mice in both anti-ChAT-saporin and Rabbit IgG-saporin treated groups. There was a significant increase in clasping at 6 wks in anti-ChAT-saporin treated R6/2 mice compared to control anti-Rabbit IgG-saporin treated R6/2 mice (Fig. 6d, f (TimeXGenotypeXSaporin)3114 = 4.31, $p = 0.006$; post hoc: 6-week anti-chat saporin R6/2 vs 6-week control anti-Rabbit IgG-saporin R6/2 $p = 0.04$). Both anti-ChAT-saporin and Rabbit-IgG-saporin injected WT mice exhibited virtually no clasping behaviour. Thus, striatal cholinergic ablation significantly accelerates the development of a dystonic phenotype in R6/2 mice.

2.5 Discussion

Dysfunctional striatal afferents may play an important role in mechanisms leading to motor symptoms of HD (Deng and Reiner, 2016; Deng et al., 2013; Heinsen et al., 1996; Tanimura et al., 2016). Here we demonstrate that the major source of thalamostriatal (TS) projections, the CM-PF complex in primates or the PF in rodents, degenerates in the R6/2 mouse model of HD. Furthermore, depriving the R6/2 striatum of TS inputs prior to onset of motor signs results in an acceleration of dystonic involuntary movements. Complex voluntary motor behaviours such as spontaneous paw reaching are also impaired following TS deafferentation in R6/2 compared to WT mice. The time course of worsening of spontaneous locomotion in an open field is not altered after unilateral TS lesions. Morphological analysis of degenerating striatal neurons indicates that the cholinergic interneuron subtype is especially vulnerable to TS denervation in the R6/2 mouse. In contrast, the time course of loss of MSNs and parvalbumin-positive interneurons is unaltered following PF lesions in R6/2. Finally, induction of striatal cholinergic loss in the R6/2 striatum using immunotoxins reproduces the acceleration of dystonia seen after TS denervation in R6/2 mice, suggesting that abnormal TS-cholinergic interactions are an important contributor to the dystonia phenotype in HD.

2.5.1 The Role of Afferents in Loss of Striatal Neurons in HD

In HD, *mhtt* protein is expressed throughout the organism, but the striatum is especially vulnerable to degeneration (Vonsattel and DiFiglia, 1998). Striatal neurons are likely lost due to multiple cell autonomous mechanisms (Crotti et al., 2014; Fernandes et al., 2007; Gladding et al., 2012; Li et al., 2003; Nguyen et al., 2016; Zhang et al., 2008b). Striatal afferents may contribute to cell non-autonomous mechanisms of neuron dysfunction or death by loss of anterograde neurotrophic support (Gharami et al., 2008; Xie et al., 2010; Zuccato et al., 2005), excitotoxicity

related to abnormal ionotropic receptor signaling (Li et al., 2003; Milnerwood et al., 2012; Shehadeh et al., 2006; Zeron et al., 2002) or abnormal synaptic transmission (Deng and Reiner, 2016; Ikonomidou et al., 2000).

Glutamatergic afferents from the cerebral cortex to the striatum may participate in neuronal loss in HD by inducing excitotoxicity (Li et al., 2003; Milnerwood et al., 2012; Shehadeh et al., 2006; Zeron et al., 2002). Depriving the HD striatum of cortical afferents in the R6/2 model using lesions restricted mainly to the motor cortex appears to protect neurons sampled from the dorsolateral striatum from atrophy, although neuronal counts were not available (Stack et al., 2007). These lesioned mice also showed reduced clasping (Stack et al., 2007), a finding that may be confounded by pyramidal effects from lesioning the motor cortex. An excitotoxic role for corticostriatal (CS) glutamatergic afferents on striatal MSNs in HD was suggested. This is in keeping with previous work by several groups indicating that aberrant calcium signaling through extra-synaptic NMDA receptor (NMDAR) stimulation and increased sensitivity of NMDARs is linked to MSN excitotoxicity in HD (Dau et al., 2014; Fernandes et al., 2007; Gladding et al., 2012; Li et al., 2003; Milnerwood et al., 2012; Shehadeh et al., 2006; Zeron et al., 2002; Zhang et al., 2008b). On the other hand, CS afferents are potentially protective for MSNs, an effect that may be mediated by synaptic glutamatergic mechanisms (Hardingham and Bading, 2010; Kaufman et al., 2012) or by anterograde neurotrophin-related effects (Gharami et al., 2008; Sadikot et al., 2005; Samadi et al., 2013; Xie et al., 2010; Zuccato and Cattaneo, 2009).

In comparison to the cerebral cortex, relatively little is known of the role of the other major source of glutamatergic striatal afferents, the posterior intralaminar nuclei, in mechanisms of striatal dysfunction in HD. *In vivo* imaging of patients demonstrates that thalamic atrophy occurs early in the course of HD (Kassubek et al., 2005), and autopsy studies provide evidence for

significant loss of CM-PF neurons (Heinsen et al., 1996). Ultrastructural evidence from the Q140 heterozygous mouse model of HD provides morphological evidence of early TS dysfunction, since TS synapses on MSNs are lost by one month, whereas loss of corticostriatal synapses is observed relatively late, at one year (Deng et al., 2013). Recent work in 9-12 week old R6/2 mice also suggests abnormal morphology of TS inputs (Parievsky et al., 2017). The present work demonstrates that PF neurons, the main source of TS inputs, are lost in R6/2 mice coincident with the onset of detectable neurodegenerative changes in the neostriatum on Nissl stains (Samadi et al., 2013). Indeed. PF neuron atrophy is already detected 9 weeks, prior to significant striatal neuronal loss. There is progressive loss of PF neurons at 11 and 13 wks correlating with worsening dystonia and other locomotor deficits. Interestingly, the early significant reduction of average neuronal soma size at 9 wks is followed by apparent normalization of average soma size at 11 wks. Neuronal loss and average cell size do not necessarily correlate. Indeed, as degeneration progresses, it is expected that neurons with decreased cell size will be lost preferentially. As a result, there would be a relative abundance of larger neurons with apparent normalization of cell size. With further progression of degeneration, the remaining neurons that were initially spared also degenerate resulting in the observed reduction in soma area at 13wks. Alternatively, the degenerating PF neurons may represent a specific subpopulation. For example, different cellular subpopulations within the mouse PF may provide preferential inputs to MSNs or striatal cholinergic interneurons (Tanimura et al., 2019). It would be of interest to determine whether specific subpopulations degenerate in post-mortem HD brains and in HD models.

Another important differentiating factor between glutamatergic striatal afferents is that physiological studies in slice preparations indicate that the PF preferentially elicits NMDA currents in MSNs while CS afferents evoke a higher proportion of AMPA- mediated post-synaptic

currents (Ellender et al., 2013; Raju et al., 2006). The apparently larger contribution of NMDA mediated post-synaptic currents from PF inputs compared to CS afferents (Ellender et al., 2013; Raju et al., 2006), may suggest a differential role for the TS or CS in excitotoxicity (Kolodziejczyk and Raymond, 2016; Parievsky et al., 2017). Differential inputs to patch and matrix compartments that comprise the striatal mosaic may provide a clue to differences in thalamic or cortical-derived afferent effects on MSN survival in HD. Unlike the cerebral cortex which innervates all MSNs, the PF provides dense afferents almost exclusively to the matrix compartment of the striatum (Fujiyama et al., 2006; Raju et al., 2006; Sadikot et al., 1992b). Therefore, potential excitotoxicity from the PF would be expected to have differential effects on MSNs in either compartment. Alternatively, TS afferents may also provide a sustaining role for vulnerable striatal neurons in HD (Samadi et al., 2013), analogous to their trophic role in normal striatal development(Sadikot et al., 2005). Indeed, BDNF is enriched in PF neurons (Conner et al., 1997; Sadikot et al., 2005), and there is an early reduction in BDNF mRNA in striatal afferents including in the PF of R6/2 mice (Samadi et al., 2013). Furthermore, the ability to activate striatal TrkB receptors in the R6/2 striatum is impaired (Nguyen et al., 2016)**.** Importantly, the present findings indicate that early TS lesions in R6/2 mice have no significant effect on MSN size or number using unbiased stereology performed separately on either patch or matrix compartments of sham and lesioned R6/2 mice. These findings suggest that loss of projection neurons in HD likely involves a complex interplay between neurotrophic, excitotoxic and cell autonomous mechanisms, and loss of glutamatergic TS afferents is not a major factor determining survival of MSNs in the HD striatum.

2.5.2 Vulnerability of Specific Interneuron Subtypes

Although striatal interneuron subtypes comprise only 5-10% of the striatal population, they are important modulators of striatal function in health and disease states (Aoki et al., 2015;

Bradfield and Balleine, 2017; Bradfield et al., 2013; Ding et al., 2010; Gernert et al., 2000; Gonzales and Smith, 2015; Kimura et al., 2004; Nakano et al.; Nanda et al., 2009; Reiner et al., 2013; Tepper and Bolam, 2004). Striatal interneurons include cholinergic neurons, and GABAergic subtypes that express somatostatin, parvalbumin, or calretinin (Tepper and Bolam, 2004). Striatal interneurons modulate MSNs via local synapses, and also at a distance across patch/matrix boundaries (Kawaguchi, 1992; Penny et al., 1988). In rodents, the PF contributes only a small proportion of excitatory synapses to striatal PV neurons (Nakano et al.; Rudkin and Sadikot, 1999). In contrast, striatal PV interneurons receive dense asymmetric inputs from the cerebral cortex (Nakano et al.; Ramanathan et al., 2002b) suggesting they may be more sensitive to pathological changes affecting the cortex rather then the PF in HD. Early work suggested that striatal PV interneurons may be spared in HD, (Harrington and Kowall, 1991) but more recent findings in autopsied HD brains indicate an important reduction in PV interneurons (Reiner et al., 2013). The present results provide stereological evidence for a decrease in soma size and number of striatal PV interneurons in the R6/2 model of HD. Early PF lesions in the R6/2 model do not accelerate the time course of degeneration of PV interneurons. As with MSNs, degeneration of PV neurons is likely due to a combination of cell autonomous and non-autonomous mechanisms (Giampa et al., 2009; Simmons et al., 2013), but the TS projection does not play a major survival role for PV interneurons in the face of neurodegenerative stress in HD.

Cholinergic interneurons make up 1% of all striatal neurons, synapse on most MSNs and other interneurons, and modulate dopaminergic, and glutamatergic terminals in the striatum (Gonzales and Smith, 2015). Ultrastructural studies indicate that the predominant glutamatergic input to cholinergic interneurons is from the posterior intralaminar nuclei in rodents and monkeys (Bennett and Bolam, 1994; Bolam et al., 1984; Doig et al., 2014; Lapper et al., 1992; Sidibe and

Smith, 1999). Although physiological and viral-based tracing studies (Guo et al., 2015; Wilson et al., 1990) suggest that cholinergic interneurons may receive cortical input, there is little ultrastructural evidence for inputs from the cerebral cortex in rodents (Lapper and Bolam, 1992). Classically, cholinergic interneurons were thought to be spared in HD (Ferrante et al., 1987a). However, recent evidence points to significant striatal cholinergic dysfunction in HD patients, including reduced synthetic and vesicular proteins (Smith et al., 2006; Suzuki et al., 2001), and decreased ChAT + cell numbers (Massouh et al., 2008). Several electrophysiological studies have shown abnormal cholinergic responses to afferent stimulation and decreased acetylcholine release in slice preparations in R6/2 or Q175 mouse models (Farrar et al., 2011; Picconi et al., 2006; Tanimura et al., 2016; Vetter et al., 2003). In the R6/1 mouse model of HD, striatal vesicular choline acetylcholine transporter and ChAT mRNA and protein concentrations are reduced in tissue lysates, and *mhtt* aggregates accumulate in cholinergic neurons (Smith et al., 2006). Ultrastructural evidence in the Q140 mouse model of HD indicates that striatal cholinergic interneurons have a decreased number of TS synapses, reduced cell diameter and fewer dendritic branches (Deng and Reiner, 2016). In keeping with this work, *ex vivo* brain slices derived from the Q175 mouse model of HD show decreased synaptic facilitation at cholinergic targets in response to PF stimulation (Tanimura et al., 2016). The present results from R6/2 mice suggest that neuronal degeneration in the PF occurs early in the course of HD, and therefore contributes to loss of TS synaptic integrity and function (Parievsky et al., 2017). The observed loss of PF neurons precedes cholinergic neuron atrophy and cell loss which normally only occurs at late timepoints suggesting a relative resistance of cholinergic neurons to degeneration in HD. Early PF lesions accelerate the atrophy and loss of cholinergic neurons in R6/2 mice, suggesting that these neurons

are especially dependent on sustaining thalamic input in the face of *mhtt* related neurodegenerative stress.

Multiple mechanisms may underlie the differential vulnerability of striatal cholinergic interneurons to TS deafferentation compared to other striatal populations. The fact that the glutamatergic TS system provides more prominent input to cholinergic interneurons (Bennett and Bolam, 1994; Bolam et al., 1984; Lapper et al., 1992; Sidibe and Smith, 1999) compared to PV interneurons (Nakano et al.; Rudkin and Sadikot, 1999) may explain their sensitivity to TS deafferentation in R6/2 mice through both glutamatergic and trophic factor receptor mechanisms. For example, cholinergic interneurons express lower levels of ionotropic NMDA-2A and metabotropic GluR1/5 glutamate receptors (Tallaksen-Greene et al., 1998) then other striatal cell types, but maintain high NMDA-2B expression (Kuppenbender et al., 2000; Landwehrmeyer et al., 1995; Standaert et al., 1999). Signaling from mGLUR5 and synaptic NMDA receptors enriched in NMDA-2A subunits can stabilize mitochondrial membranes and promote cell survival, (Hardingham and Bading, 2010; Kaufman et al., 2012) while neurotoxic extrasynaptic NMDA receptors rich in NMDA-2B subunits contribute to mitochondrial failure and cell death in MSNs in various HD models (Dau et al., 2014; Gladding et al., 2012; Hardingham and Bading, 2010; Ikonomidou et al., 2000; Milnerwood et al., 2012; Zeron et al., 2002; Zhang et al., 2008b). In keeping with this evidence, the present *in vivo* results demonstrate striatal cholinergic interneurons in R6/2 mice are more susceptible to a mitochondrial toxin than WT neurons suggesting that they are more vulnerable to cellular energy failure.

In addition to glutamatergic modulation of cell death, neurotrophins may also play an important role in cholinergic neuron vulnerability to TS loss in HD. The neurotrophin brain-derived neurotrophic factor (BDNF) promotes forebrain cholinergic neuron maintenance, growth

(Martinez-Serrano et al., 1996), and survival (Larsson et al., 1999; Lucidi-Phillipi et al., 1995). In particular, ChAT+ striatal neurons express both TrkA and TrkB receptors (Richardson et al., 2000; Steininger et al., 1993) and contain BDNF protein (Fusco et al., 2003). Given that the striatum lacks BDNF mRNA (Altar et al., 1997; Conner et al., 1997; Lein et al., 2007; Malkovska et al., 2006; Schmidt-Kastner et al., 1996; Zuccato et al., 2001), the BDNF protein in ChAT+ cells may derive from post-synaptic internalization and endosomal trafficking of BDNF released from afferents (Ayloo et al., 2017; Braunstein et al., 2010; Cohen et al., 2011; Kononenko et al., 2017; Liot et al., 2013; Moya-Alvarado et al., 2018; Sasi et al., 2017). Endosomal trafficking of TrkB/BDNF complexes towards the soma provides trophic support to neurons, is regulated by htt and is reduced in the presence of mhtt (del Toro et al., 2006; Gauthier et al., 2004; Liot et al., 2013; Numakawa et al., 2010; Zheng et al., 2008). Since a higher proportion of cholinergic neurons express htt compared to other striatal subpopulations (Fusco et al., 1999; Fusco et al., 2003), they may be especially vulnerable to loss of BDNF. Importantly, the TS system is the main source of glutamatergic afferents to cholinergic neurons (Bennett and Bolam, 1994; Bolam et al., 1984; Doig et al., 2014; Lapper et al., 1992; Sidibe and Smith, 1999) is enriched in BDNF mRNA (Conner et al., 1997; Samadi et al., 2013), and PF lesions reduce striatal BDNF-TrkB signaling in neonatal rodents (Sadikot et al., 2005). The PF may therefore contribute to the relative resistance to degeneration of cholinergic neurons. Loss of BDNF following PF lesions or degeneration in HD may make cholinergic neurons more vulnerable to degeneration in HD.

2.5.3 Implications of Thalamostriatal and Cholinergic Dysfunction for Dystonia and HD

Dystonia can be a motor feature of both hypokinetic and hyperkinetic disorders including primary and secondary dystonic syndromes, and other neurodegenerative diseases (Marsden, 1976; Neychev et al., 2011; Phukan et al., 2011). Secondary dystonia can occur following lesions

in different parts of the thalamic, cerebellar or basal ganglia network (LeDoux and Brady, 2003; Lee and Marsden, 1994; Marsden et al., 1985; Meissner et al., 1987; Rumbach et al., 1995; Zadro et al., 2008). Dystonia is also a common symptom in HD, and worsens with disease progression, but appears not to correlate with chorea or bradykinesia (Louis et al., 1999). Clasping behaviour is considered a surrogate for dystonia in rodent models as it mimics the sustained muscle contractions and abnormal postures seen in humans (Marsden, 1976). Clasping behaviour occurs in many animal models of HD and primary dystonia (Gernert et al., 2000; Pappas et al., 2015; Samadi et al., 2013; Wilson and Hess, 2013). Furthermore, clasping behavior is well studied in the R6/2 mouse and worsens significantly as the model progresses (Samadi et al., 2013), similar to the age related increase in dystonia seen in HD patients (Louis et al., 1999).

Basal ganglia, cerebellar, brainstem and cortical dysfunction are proposed in both human dystonia and in the many animal models exhibiting clasping behaviours reminiscent of dystonia (Eskow Jaunarajs et al., 2015; Wilson and Hess, 2013). Interrogation of different components of the striatal micro-circuitry in animal models allows better understanding of dystonia. DYT1 mouse models of primary dystonia show decreased intrastriatal dopamine release possibly due to reduced nicotinic cholinergic tone (Bao et al., 2010), and paradoxical dopamine D2 receptor mediated excitation of cholinergic neurons (Pisani et al., 2006; Sciamanna et al., 2012a). These altered cholinergic-dopaminergic interactions in DYT1 mice impair long-term depression in MSNs and increase corticostriatal synaptic long-term potentiation, leading to abnormal striatal output (Bao et al., 2010; Eskow Jaunarajs et al., 2015; Sciamanna et al., 2012a; Sciamanna et al., 2012b). These synaptic deficits may even occur early in brain development, as mice with selective forebrain DYT1 knockout show an early clasping phenotype associated with post-natal loss of cholinergic striatal interneurons and decreased striatal acetylcholine release (Pappas et al., 2015).

Similar changes in striatal cholinergic micro-circuitry are described in HD models. These include: the inability of striatal cholinergic cells to undergo long-term potentiation with an associated inability of MSNs to undergo long-term synaptic depression (Picconi et al., 2006), decreased acetylcholine (Farrar et al., 2011; Vetter et al., 2003), and increased MSN and cholinergic responses to cortical excitation (Parievsky et al., 2017; Tanimura et al., 2016). These abnormalities are compounded in HD by atrophy and loss of striatal cells (Cicchetti and Parent, 1996; Massouh et al., 2008; Reiner et al., 2013; Vonsattel and DiFiglia, 1998).

The posterior intralaminar nuclei are important drivers of cholinergic activity in the normal striatum (Aoki et al., 2015; Bradfield and Balleine, 2017; Bradfield et al., 2013; Brown et al., 2010; Ding et al., 2010; Kimura et al., 2004; Nanda et al., 2009; Threlfell et al., 2012). Cholinergic neurons modulate long-term plasticity of MSNs by regulating dopamine and glutamate co-release onto MSNs through pre-synaptic acetylcholine receptors on glutamatergic and dopaminergic terminals (Threlfell et al., 2012). More specifically, the pause-response of cholinergic neurons to TS stimulation, which is mediated by D2-receptors on cholinergic cells as well as presynaptic nicotinic receptors on dopaminergic terminals, helps to transiently inhibit both direct and indirect pathway MSNs responses to cortical stimulation and then later facilitate post-synaptic cortical glutamatergic excitation of indirect pathway neurons, thereby preferentially driving the network towards action cessation (Ding et al., 2010). Loss of the PF-cholinergic mediated tuning of striatal projection neurons leads to an imbalance between competing basal ganglia pathways and is thought to impair saliency estimation and motor program selection (Bradfield and Balleine, 2017; Bradfield et al., 2013; Brown et al., 2010; Kimura et al., 2004; Matsumoto et al., 2001), and contribute to the generation of dystonia (Eskow Jaunarajs et al., 2015). Indeed, TS-cholinergic deficits have been shown in HD and dystonia models. For example, in a DYT1 mutant mouse, the

normal pause-response is replaced by erratic firing of cholinergic cells to TS stimulation (Sciamanna et al., 2012b) and in the Q175 HD mouse, there is reduced TS synaptic facilitation of cholinergic interneurons and loss of the normal pause-spike response to TS stimulation (Tanimura et al., 2016). The present work demonstrates that lesioning either the TS system or striatal cholinergic interneurons exacerbates dystonia in the R6/2 HD mouse. Altogether these findings suggest that dysfunction of both the TS system and loss of cholinergic interneurons plays an important role in the generation of dystonia in HD and in primary dystonia models.

In addition to the striatum and the TS system, the cerebellum is proposed as an important part of the dystonia network. Evidence from imaging studies suggests reduced cerebellar activity, degeneration of the cerebello-thalamocortical pathway and abnormal cerebellar sensorimotor integration in dystonia patients (Dresel et al., 2014; Hutchinson et al., 2000; Yang et al., 2014). HD patients show cerebellar degeneration that correlates with a worse motor score (Rees et al., 2014; Rub et al., 2013; Vonsattel and DiFiglia, 1998). Furthermore, mouse models which have a severe clasping phenotype such as the R6/2 and Hdh100 HD mouse models also demonstrate a loss of Purkinje cells at late timepoints (Dougherty et al., 2013; Dougherty et al., 2012).

The intralaminar nuclei receive afferents from deep cerebellar nuclei and form a disynaptic link between the basal ganglia and the cerebellar nuclei (Hoshi et al., 2005; Melik-Musyan and Fanardjyan, 1998; Pelzer et al., 2017). The output of the cerebello-thalamic circuit plays an important role in saliency estimation and action selection (Bradfield and Balleine, 2017; Bradfield et al., 2013; Brown et al., 2010; Kimura et al., 2004). Similar to the effect of unilateral cerebellar lesions in rats (Edalatmanesh et al., 2014), PF lesions in the present work led to decreased spontaneous unilateral paw use in both WT and R6/2 when exploring a cylinder. This provides evidence for a role for the TS system in evaluation of salient sensory information and appropriate

motor program selection. Furthermore, both thalamic and cerebellar strokes lead to secondary dystonia in susceptible individuals (LeDoux and Brady, 2003; Lee and Marsden, 1994; Marsden et al., 1985; Meissner et al., 1987; Rumbach et al., 1995; Zadro et al., 2008). Atrophy and cell loss of the TS system and the cerebellum in HD (Heinsen et al., 1996; Rees et al., 2014; Vonsattel and DiFiglia, 1998) may therefore contribute to the dystonia network in HD. TS afferents degenerate in R6/2 mice expressing a dystonic clasping phenotype, and early PF lesions in R6/2 lead to a worsening of dystonia. We therefore propose that TS degeneration, with downstream pathology at cholinergic targets plays an important part in the network leading to expression of dystonia in HD and possibly in other dystonic syndromes.

2.6 Conclusion

Thalamostriatal afferents provide important trophic support to striatal cholinergic neurons in Huntington's disease. Furthermore, pathological dysfunction of the TS system and cholinergic interneurons is closely linked to the generation of a dystonic phenotype in HD models. This work provides a new understanding of mechanisms of striatal degeneration and motor symptoms in HD, and may pave the way for development of effective therapies for those affected by this currently incurable neurodegenerative disorder.

Chapter 3: How Thalamostriatal Afferents Participate in Dystonia in HD and Other Movement Disorders

3.1 The Thalamostriatal System Degenerates in HD

There exists a growing body of evidence that striatal afferents undergo significant atrophy in early stages of HD. For example, *In vivo* imaging demonstrates that prodromal HD patients have thalamic and cortical atrophy (Aylward et al., 2011; Kassubek et al., 2005; Kassubek et al., 2004; Matsui et al., 2014; Rosas et al., 2008). Degeneration of intralaminar nuclei in the context of HD is further supported by autopsy studies which show a significant loss of CM-PF neurons (Heinsen et al., 1996). Recent work with premanifest models of HD such as the heterozygous Q140 mouse (Hickey et al., 2008) demonstrated an early deafferentation of PF afferents at one and four months of age before late cortical deafferentation at one year (Deng et al., 2013; Deng et al., 2014). Interestingly, PF deafferentation at four months of age in the Q140 model coincides with deficits in cholinergic interneurons including: reduced cholinergic cell diameter and dendritic branching (Deng and Reiner, 2016). TS deafferentation has also been shown in the YAC 128 mouse (Kim et al., 2015). Changes in trophic factor receptors, and cell scaffolding proteins in the posterior intralaminar nuclei of post-mortem studies of HD patients are also present in R6/2 mice (Freeman and Morton, 2004; Kusakabe et al., 2001; Samadi et al., 2013; Tang et al., 2011)

This work led us to hypothesize that the thalamostriatal system also degenerates in R6/2 mice, a highly symptomatic mouse model of HD. Unbiased cell counting experiments in R6/2 and WT mice confirmed this hypothesis by demonstrating that the PF, the main source of TS afferents, undergoes neuronal atrophy at nine weeks and neuron loss by 11 weeks compared to WT mice**.** Interestingly, the changes in cell soma area occurred relatively early in the time course of both motor and striatal dysfunction in that model. This led us to formulate the hypothesis that TS degeneration represents an important loss of trophic support to downstream striatal neurons and

affects the motor phenotype in HD. These hypotheses are discussed further in the following sections.

3.2 Changes to the Striatal Microcircuit and its Glutamatergic Afferents in HD

Glutamatergic afferents such as the cortex and thalamus are known to provide trophic support in the form of NMDA receptor stimulation and anterograde trophic factor delivery to striatal neurons (Baquet et al., 2004; Hetman and Kharebava, 2006; Ikonomidou et al., 2000; Loopuijt et al., 1997a; Loopuijt et al., 1997b). On the other hand, depriving the HD striatum of cortical afferents in the R6/2 model using lesions restricted mainly to the motor cortex appears to protect from striatal neuronal atrophy, as measured by sampling in the dorsolateral striatum, although neuronal counts were not available (Stack et al., 2007). These lesions also resulted in reduced clasping in the contralateral limb, although pyramidal effects may be a confound (Stack et al., 2007). Those results suggested an excitotoxic role for the corticostriatal (CS) glutamatergic afferents on striatal MSNs in HD.

In keeping with those findings, several groups have shown that aberrant calcium signaling through extra-synaptic NMDAR stimulation and increased sensitivity of NMDAR is linked to MSN excitotoxicity in HD (Dau et al., 2014; Fernandes et al., 2007; Gladding et al., 2012; Graham et al., 2006; Li et al., 2003; Milnerwood et al., 2012; Shehadeh et al., 2006; Zeron et al., 2002; Zhang et al., 2008b). Physiological studies in slice preparations indicate that the PF preferentially elicits NMDA-mediated post-synaptic currents while CS and non-PF thalamic afferents preferentially induce AMPA-mediated post-synaptic currents (Ellender et al., 2013; Raju et al., 2006). The apparent larger contribution of NMDA mediated post-synaptic currents from TS compared to CS afferents, may suggest a role for the TS system in excitotoxicity (Kolodziejczyk and Raymond, 2016; Parievsky et al., 2017). On the other hand, synaptic NMDAR stimulation

appears to be protective in various HD models (Hardingham, 2009; Hardingham and Bading, 2010; Hetman and Kharebava, 2006) and striatal NMDA receptors are downregulated early in HD (Albin et al., 1990a; Albin et al., 1990b; Young et al., 1988). These seemingly contradictory effects of NMDAR stimulation muddle the role of glutamatergic afferents on striatal neurodegeneration in HD.

Alternatively, both the TS and CS afferents are proposed to be potentially protective for MSNs through anterograde transport of neurotrophins including BDNF (Baquet et al., 2004; Bartkowska et al., 2010; Baydyuk et al., 2013; Baydyuk and Xu, 2014; Causing et al., 1997; Dragatsis et al., 2000; Duyao et al., 1995b; Fawcett et al., 1998; Reilly, 2001; Xie et al., 2010; Xu et al., 2000; Zhang et al., 2008a; Zuccato et al., 2007; Zuccato and Cattaneo, 2009; Zuccato et al., 2001; Zuccato et al., 2005; Zuccato et al., 2003). Furthermore, BDNF is enriched in PF neurons (Conner et al., 1997). The neurotrophic effects of afferent derived BDNF are abnormal in HD as *mhtt* is known to interfere with the synthesis of BDNF (Zuccato et al., 2001). In keeping with this, there is an early reduction in BDNF mRNA in striatal afferents including in the PF (Samadi et al., 2013). Furthermore, the dysfunction in BDNF mRNA synthesis coincides with impaired activation of striatal trkB receptors in the R6/2 striatum (Brito et al., 2013; Nguyen et al., 2016). As discussed below, our results suggest that the sum of neuroprotective and potentially neurodegenerative effects of glutamatergic TS afferents is to support vulnerable cholinergic interneurons from degeneration in HD. On the other hand, lesions of the TS do not increase MSNs or PV interneuron cell loss.

The striatum is a mosaic comprised of two intermingled compartments called the patch and matrix (Gerfen, 1984; Herkenham and Pert, 1981). The matrix compartment comprises roughly 80% of all striatal neurons and the patch compartment making up the rest (Fujiyama et al., 2015).

The literature suggests that there is a differential patch-matrix degeneration in HD patients with different repeat lengths. Patients with longer repeat length have equal degeneration in both patch-matrix compartments along with more severe motor symptoms (Ferrante et al., 1987b; Seto-Ohshima et al., 1988; Tippett et al., 2007; Waldvogel et al., 2012). Lower repeat lengths are associated with a later age at onset, fewer motor symptoms and a predominant patch degeneration (Hedreen and Folstein, 1995; Morton et al., 1993; Tippett et al., 2007). This preferential patch atrophy was also noted in the YAC128 mouse which also has a mild and late-onset phenotype (Lawhorn et al., 2008). Conversely, the current work suggests that both the patch and matrix compartments degenerate in the R6/2 mouse thereby mimicking the pattern of degeneration which occurs in patients with higher repeat numbers and a prominent motor phenotype. This finding is broadly consistent with the R6/2 being a model of juvenile HD with a severe motor phenotype which has a relatively rapid onset and early death.

Unlike the cortex which innervates all MSNs, PF afferents are known to project exclusively to the matrix striatal compartment MSNs (Bennett and Bolam, 1994; Bolam et al., 1984; Bracci et al., 2002; Centonze et al., 2003; Dube et al., 1988; Fujiyama et al., 2006; Guo et al., 2015; Herkenham and Pert, 1981; Koos and Tepper, 2002; Lacey et al., 2007; Lapper and Bolam, 1992; Lapper et al., 1992; Nakano et al.; Raju et al., 2006; Ramanathan et al., 2002b; Rudkin and Sadikot, 1999; Sadikot et al., 1992a; Sadikot et al., 1992b; Sidibe and Smith, 1999) therefore potential excitotoxicity from the PF would be expected to have differential effects on MSNs in the patch and matrix compartments. Since TS lesions promoted neither neuronal survival nor enhanced neuronal death of MSPs in either compartment, it is apparent that MSN cell loss occurs by complex mechanisms that may depend on an interplay between neurotrophic, excitotoxic and cell-autonomous mechanisms.

Parvalbumin interneurons are fast-spiking and receive a rich cortical innervation as well as some innervation by intralaminar and other nuclei and project preferentially to direct pathway MSNs (Bracci et al., 2002; Centonze et al., 2003; Gittis et al., 2010; Koos and Tepper, 2002; Nakano et al.; Ramanathan et al., 2002b; Rudkin and Sadikot, 1999). Early work suggested the PV interneuron subtype may be spared in HD (Harrington and Kowall, 1991) but more recent findings in autopsied HD brains have shown an important reduction in PV+ interneurons (Reiner et al., 2013). The present results are in agreement with more recent studies and provide stereological evidence for a decrease in soma size and number of striatal PV+ interneurons in the R6/2 model.

The mechanisms for PV+ loss in HD is unknown but likely includes a combination of cell autonomous and non-autonomous mechanisms (Giampa et al., 2006; Giampa et al., 2009; Leuti et al., 2013; Simmons et al., 2013)(Anzilotti et al., 2012; Fusco et al., 2012; Giampa et al., 2006; Giampa et al., 2009; Meade et al., 2002). However, cell-specific knock-in HD models in which the mutated huntingtin gene was added to these cells produces a mild phenotype and no PV+ cell loss even at very late timepoints (Dougherty et al., 2014). These findings suggest that cell non-autonomous mechanism may be more important contributors to the degenerative stress of PV interneurons in HD. Indeed, ultrastructural evidence in R6/2 suggests that striatal PV+ interneurons had reduced dendrite and synapse loss after chronic administration of a trkB ligand (Simmons et al., 2013). This is of interest because BDNF mRNA is reduced in all main striatal afferents including the cerebral cortex and PF at early timepoints (Samadi et al., 2013).

As with MSNs, early PF lesions do not accelerate the time course of degeneration of PV+ interneurons, suggesting that the TS projection does not play major survival role for PV+ interneurons in the face of neurodegenerative stress in HD. This is likely due to the fact that only a minority of glutamatergic synapses onto PV interneurons come from thalamic sources (Rudkin

and Sadikot, 1999, Nakano et al. 2018) instead they are robustly innervated by the cerebral cortex (Kawaguchi et al., 1997; Nakano et al.; Ramanathan et al., 2002b). Therefore, these neurons may be more sensitive to pathological changes affecting the cortex rather than the PF in HD.

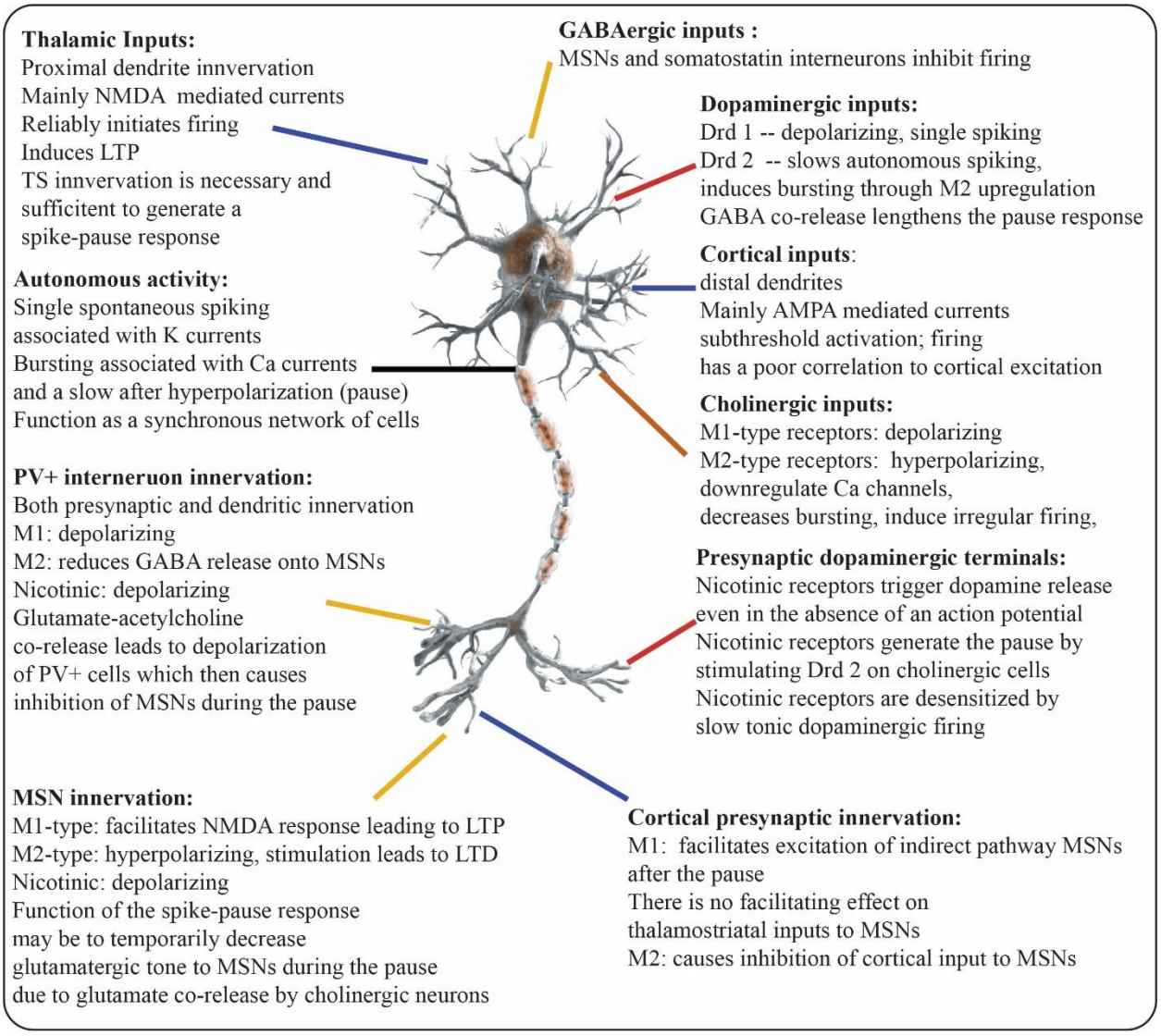
In the brain there are two main types of cholinergic neurons: projection cholinergic neurons arising from the mesopontine and basal forebrain which send ascending projections to the thalamus, cortex and hippocampus, and local cholinergic interneurons (Bolam et al., 1984b; Woolf and Butcher, 1986). In the striatum, cholinergic interneurons make up 1-2% of all striatal neurons but they synapse onto most MSNs, striatal afferents and other interneurons (Gonzales and Smith, 2015). They exert their effects on other striatal neurons through muscarinic (M 1-5) and nicotinic receptors (α4, β2). M-1 type muscarinic receptors can directly depolarize MSN while M-2 type receptors inhibit MSNs and modulate their responsiveness to NMDA receptor stimulation (Picconi et al., 2006). Cholinergic output onto MSN and presynaptic nicotinic receptors on dopaminergic terminals allows for a sophisticated and real-time modulation of MSN plasticity through timing of dopamine co-release in coordination with glutamatergic afferent stimulation (Bradfield and Balleine, 2017; Bradfield et al., 2013; Brown et al., 2010; Cachope et al., 2012; Kimura et al., 2004; Threlfell et al., 2012).

Cholinergic interneurons are also known as tonically active neurons because of their propensity to autonomously produce spikes due to selective potassium and calcium channels which cyclically depolarize the cell membrane (Goldberg and Wilson, 2017). They also display bursts of activity which are often preceded by an inhibitory pause which affects local MSNs and local cholinergic neurons in response to both aversive and reward stimuli. As a result, this pause-response motif is thought to be important in detecting saliency (Nanda et al., 2009). Nearby cholinergic interneurons display synchronized activity and therefore this “saliency” signal has an

effect on vast portions of the striatal circuit at once (Goldberg and Wilson, 2017). The posterior intralaminar nuclei are important drivers of striatal cholinergic activity as PF inputs to cholinergic cells are necessary and sufficient for the latter to undergo the pause-response (Ding et al., 2010; Matsumoto et al., 2001; Nanda et al., 2009).

In summary, the physiological input and output properties of cholinergic interneurons are a very sophisticated and integral part of the striatal microcircuit. The extensive literature exploring cholinergic interneuron physiology has been reviewed in depth by (Goldberg and Wilson, 2017) and (Madeo and Pisani, 2016) and is summarized in Figure 3.2.0 below.

Figure 3.2.0 Normal Striatal Cholinergic Interneuron Physiology



For details see reviews by (Goldberg and Wilson, 2017) and (Madeo and Pisani, 2016)

Cholinergic interneurons are targets of degeneration in HD. For example, reductions in cholinergic acetylcholine vesicular transporters have been demonstrated in the striatum of HD patients (Smith et al., 2006; Spokes, 1980; Suzuki et al., 2001), there is decreased CHAT synthetic activity from HD tissue lysates (Ferrante et al., 1987a; Spokes, 1980; Suzuki et al., 2001), and HD patients have a modest improvement in motor and cognitive symptoms with pro-cholinergic medication (de Tommaso et al., 2007; de Tommaso et al., 2004). While initial studies did not find

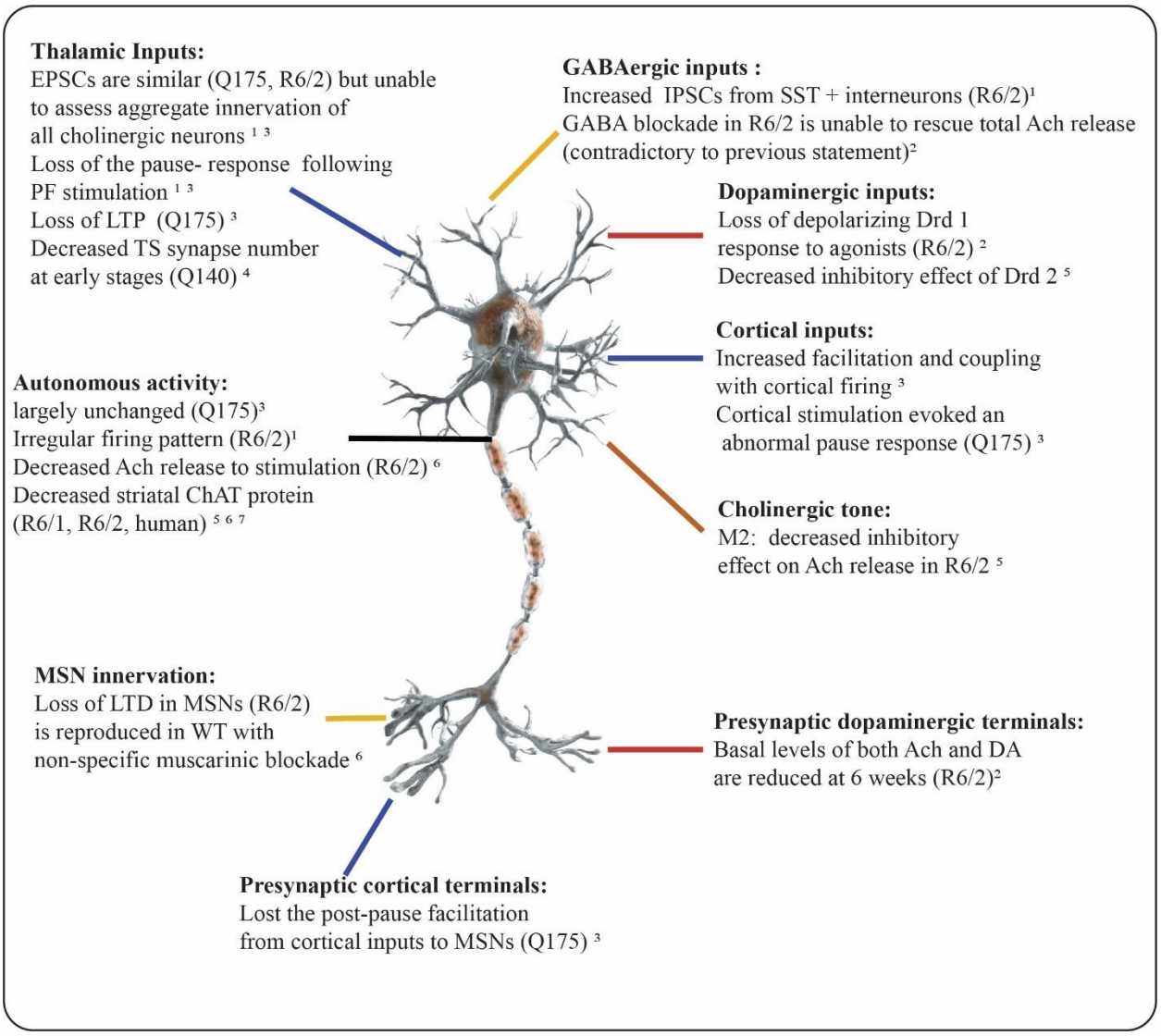
cholinergic interneuron cell loss (Ferrante et al., 1987a), more recent studies using stereological methods have found a large reduction in the number of cells expressing choline acetyltransferase, the main synthetic enzyme of cholinergic cells (Massouh et al., 2008).

Mouse models of HD reflect the deficits observed in post-mortem human HD samples and have added to our understanding of the possible mechanisms of cholinergic degeneration in HD. These include: a unique glutamatergic receptor expression profile, a reduction in trophic factor stimulation, a propensity to mitochondrial dysfunction as was demonstrated and extensively reviewed in Chapter 2 as well as an accumulation of intranuclear mhtt inclusions which are believed to be cytotoxic (Cicchetti et al., 2014; Cicchetti et al., 2009). Indeed, R6/1 mice had intranuclear mhtt inclusions in 72% of CHAT+ cells (Smith et al., 2006). This finding is not surprising as striatal cholinergic cells preferentially express *htt* protein in normal rodents (Fusco et al., 1999; Fusco et al., 2003).

Cholinergic degeneration in HD has important effects on the striatal microcircuit. For example, in the R6/2 model, striatal cholinergic interneurons are more strongly inhibited and display an irregular firing pattern (Holley et al., 2015), have decreased synthetic protein mRNA, as well as decreased production and release of acetylcholine (Farrar et al., 2011; Smith et al., 2006; Vetter et al., 2003). As a result, R6/2 mouse cholinergic interneurons are unable to undergo LTP. This is associated with an inability of MSNs to undergo LTD (Picconi et al., 2006). Deficits in MSN LTD are corrected with the addition of a cholinergic receptor agonist onto slice preparations suggesting that LTD deficits are caused by reduced cholinergic tone (Picconi et al., 2006). The loss of normal cholinergic tone in HD models may indicate a dysfunction in their glutamatergic afferents. For example, PF deafferentation occurs in the Q175 model and leads to an inability to undergo long-term potentiation of cholinergic cells, a loss the pause-spike motif which is thought

to mediate saliency estimation, and is associated with abnormal MSN and cholinergic responses to cortical excitation (Tanimura et al., 2016). Overall, these findings suggest that abnormal cholinergic interneurons and thalamostriatal afferents may lead to important striatal dysfunction in HD and are summarized in Figure 3.2.1 below.

Figure 3.2.1 Changes in Striatal Cholinergic Interneurons in Huntington's Disease



List of numerical references: 1- (Holley et al., 2015), 2- (Farrar et al., 2011), 3- (Tanimura et al., 2016), 4- (Deng and Reiner, 2016), 5- (Vetter et al., 2003), 6- (Picconi et al., 2006), 7- (Smith et al., 2006)

Despite these important deficits, no prior studies have shown a reduction in the number of striatal cholinergic neurons in HD models (Holley et al., 2015; Smith et al., 2006) although none have quantified the numbers using unbiased stereology at highly symptomatic (11 weeks) or terminal ages (13 weeks). At the 13-week timepoint, R6/2 mice undergo cholinergic degeneration suggesting that those cells may be more resistant to cell loss compared to PV interneurons and MSN which have reduced cell numbers earlier at 11 weeks.

Interestingly, the time course of PF atrophy at 9 weeks prior to cholinergic cell loss at 13 weeks in R6/2 mice led us to hypothesize that PF afferents play a trophic role for those striatal neurons. Early TS deafferentation accelerates the atrophy and loss of cholinergic neurons in R6/2 mice and confirmed our second hypothesis that TS afferents provide trophic support to striatal neurons in HD. Overall, this suggests that rescuing the thalamostriatal circuit from degeneration may be of potential therapeutic benefit and help slow degeneration of cholinergic interneurons and preserve striatal function in HD.

3.3 Dystonia Arises from Multiple Hits to the Motor Circuit

Dystonia can be a motor symptom of many hypo and hyperkinetic diseases including a dozen primary dystonic syndromes, Parkinson's patients both on and off L-DOPA, after chronic neuroleptic use, and other neurodegenerative diseases (D'Abreu et al., 2011; Goto et al., 2005; Kanovsky et al., 2015; Marálek, 2000; Marsden, 1976; Phukan et al., 2011; Steele et al., 1964; Tinazzi et al., 2003; Tolosa and Compta, 2006; van Harten et al., 1997; Wilson and Hess, 2013). This is also the case in HD as dystonia is a common symptom but does not correlate with worse chorea or bradykinesia (Louis et al., 1999). Dystonia also occurs after thalamic, cerebellar and basal ganglia lesions such as strokes; lesional cases are termed secondary dystonia (Bhatia and Marsden, 1994; Demierre and Rondot, 1983; LeDoux and Brady, 2003; Lee and Marsden, 1994;

Marsden et al., 1985; Meissner et al., 1987; Rantamäki et al., 2001; Rumbach et al., 1995; Zadro et al., 2008).

Several studies have described paw clasping behaviours in models of HD, other diseases with a dystonic phenotype as well as lesional models mimicking secondary dystonia (Fernagut et al., 2002; Gernert et al., 2000; Jardim et al., 2001; Lalonde and Strazielle, 2011; Menalled et al., 2009; Neychev et al., 2011; Pappas et al., 2015; Pizoli et al., 2002; Samadi et al., 2013; Wilson and Hess, 2013). Furthermore, clasping behaviour has face validity as a surrogate for dystonia in rodent models as it mimics the sustained muscle contractions and abnormal postures which define dystonia in humans (Klein and Fahn, 2013; Marsden, 1976).

Basal ganglia, cerebellar, nigral and cortical dysfunction co-occurs in dystonia and many models exhibiting clasping behaviours reminiscent of dystonia (Eskow Jaunarajs et al., 2015; Hess and Jinnah, 2015; Lalonde and Strazielle, 2011; Lehericy et al., 2013; Neychev et al., 2011; Wilson and Hess, 2013). Some imaging and transcranial magnetic stimulation studies suggest that abnormal inhibition or plasticity within the sensorimotor cortex may be associated with dystonia (Hallett, 2011; Quartarone, 2013; Quartarone et al., 2005; Quartarone et al., 2008; Quartarone and Pisani, 2011). However, others have suggested that these changes are also present in psychogenic dystonia (Espay et al., 2006; Quartarone et al., 2009) or are compensatory (O'Dwyer et al., 2005; Scontrini et al., 2009). Overall, this contradictory evidence suggests that changes in the cerebral cortex may not provide a major contribution to the development of this movement disorder.

On the other hand, *in vivo* imaging has shown that the cerebellum is hypoactive on PET studies while fMRI studies suggest that there are changes in cerebellar sensorimotor integration which can normalize after treatment (Dresel et al., 2014; Hutchinson et al., 2000; Koch et al., 2014; Lehericy et al., 2013; Niethammer et al., 2011). Others demonstrated abnormal cerebello-

thalamocortical anisotropy implying decreased cerebellar afferentation of the thalamus (Ramdhani et al., 2014; Yang et al., 2014). Cerebellar degeneration in HD, both morphologically and on *in vivo* imaging, correlates with a worse motor score (GOEBEL et al., 1978; Rees et al., 2014; Rub et al., 2013). Purkinje cell atrophy occurs in HD models with a dystonic clasping phenotype such as the Hdh150 and R6/2 models (Dougherty et al., 2013; Dougherty et al., 2012).

Despite this evidence, dystonia cannot be completely explained by cerebellar pathology as patients often lack other cerebellar signs such as ataxia and dysmetria (Filip et al., 2013; Hendrix and Vitek, 2012; Lehericy et al., 2013). *In vivo* imaging and neuronal recordings in dystonia patients have shown that the basal ganglia is hypermetabolic and has decreased D2 receptor binding (Blood et al., 2012; Galardi et al., 1996; Lehericy et al., 2013; Naumann et al., 1998). Neurophysiological studies have shown that the striatum is abnormally locked to cortical beta oscillations (Neumann et al., 2015) while others have shown increased pallidal activation in response to repeated tasks (Lenz et al., 1998). Finally, deep brain stimulation of the globus pallidus improves symptoms further suggesting that abnormal basal ganglia plasticity is important to dystonia pathogenesis (Mink, 2018; Ostrem and Starr, 2008).

The intralaminar thalamic nuclei are a disynaptic link between basal ganglia and cerebellar networks since the intralaminar nuclei receive afferents from deep cerebellar nuclei in rats, cats, non-human primates and in humans (Bostan et al., 2010; Bostan and Strick, 2010; Cornwall and Phillipson, 1988; Hendry et al., 1979; Hoshi et al., 2005; Melik-Musyan and Fanardjyan, 1998; Pelzer et al., 2017) for review see (Hintzen et al., 2018). The PF therefore integrates cerebellar inputs and sends its output to the striatum. The TS-cholinergic interaction in the striatum is important for action selection (Bradfield and Balleine, 2017; Bradfield et al., 2013; Brown et al.,

2010; Nanda et al., 2009). More specifically, it was shown to modulate MSN output pathways towards action cessation (Ding et al., 2010).

Moreover, disruption of cerebello-thalamo-striatal pathways such as in thalamic or cerebellar strokes can cause secondary dystonia (LeDoux and Brady, 2003; Lee and Marsden, 1994; Marsden et al., 1985; Meissner et al., 1987; Rantamäki et al., 2001; Rumbach et al., 1995; Zadro et al., 2008). Atrophy and cell loss of the TS system and the cerebellum in HD (GOEBEL et al., 1978; Heinsen et al., 1996; Rees et al., 2014; Rub et al., 2013) may have a similar effect to lesions of those structures in secondary dystonia. Indeed, either PF lesions or induction of striatal cholinergic loss in the R6/2 striatum using immunotoxins leads to an acceleration of the dystonia phenotype in that model. This confirmed our third hypothesis that dysfunction of TS-cholinergic system affects the motor phenotype in HD. Interestingly, neither PF lesions nor striatal cholinergic immunotoxin lesions produced a dystonic phenotype in WT mice. Therefore, we conclude that the system can compensate for loss of single elements of the TS-cholinergic circuit. However, a combination of cerebellar, TS and striatal pathology in R6/2 mice overwhelms the systems ability to suppress abnormal motor programs and leads to the generation of a dystonic phenotype. This adds to the growing body of evidence which supports a motor network theory of dystonia which requires pathological changes in multiple brain structures (Hess and Jinnah, 2015; Jinnah et al., 2017; Neychev et al., 2011).

3.4 Strengths and Limitations

The strengths of this study included the large animal cohorts used in behavioural experiments, the rigorous planning of behavioural experiments to reproduce the same environment including precise timing of experiments at each behavioural timepoint, attention to the time of day at which these were performed, and this over many cohorts of animals. Also, optimal

concentrations of recently purchased reagents were used for all histological processing allowing for high quality material which can be analyzed for years to come. Finally, blinded behavioural scoring and blinded cell counting with state-of-the-art unbiased stereological techniques allowed for impartial and unbiased assessments of histological materials and phenotypic progression.

Some limitations of these experiments include lack of earlier timepoints in the histological analysis. It is possible that there was an early effect of thalamic lesion at these timepoints on matrix and parvalbuminergic neurons which was lost at the later timepoints. Another limitation was that these experiments did not address the glial response to thalamic deafferentation or cholinergic ablation. There is evidence that excessive microglial activation (Crotti et al., 2014; Franciosi et al., 2012; Pavese et al., 2006; Politis et al., 2011; Sapp et al., 2001; Tai et al., 2007) and astrogliosis (Bayram-Weston et al., 2012; Cirillo et al., 2010; Hickey et al., 2008; Meunier et al., 2016) contribute to the disease process in HD and may very well have affected the time course of degeneration in these experiments. The current work did not explain mechanistically how thalamic afferents provide trophic support to cholinergic neurons. NMDA receptor stimulation (Ellender et al., 2013; Hardingham, 2009; Hardingham and Bading, 2010; Hetman and Kharebava, 2006) and anterograde BDNF transport (Zuccato et al., 2001; Zuccato et al., 2005; Zuccato et al., 2003) were proposed. However, how each mechanism contributes to neuronal cell loss and at which point these processes overlap or diverge was not elucidated. An experiment rescuing TS afferents or striatal interneuron cell groups such as the cholinergic interneurons through modulation of their afferents, overexpression of trophic factors or knock-down of extra-synaptic NMDA receptors would be an important next step in teasing out these effects. This study does not address what happens physiologically to MSNs, parvalbumin or cholinergic interneurons as R6/2 mice undergo their dystonic episodes. This would be of interest as it would give a functional understanding of

how abnormal TS-cholinergic interactions contribute to dystonia *in vivo*. Finally, we did not assess which combination of motor circuit element pathology is required to generate a dystonic phenotype in normal mice. This finding would have broader implications for all types of dystonic syndromes and could lead to more effective treatments for this common movement disorder.

3.5 Future Applications

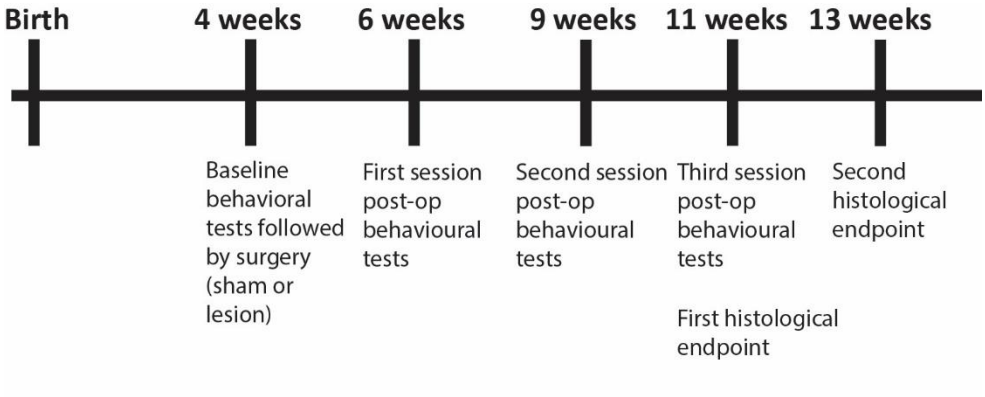
Accelerating the degenerative process in R6/2 through thalamostriatal lesion, as was hypothesized and later shown by these experiments, was done in the hopes of being able to characterize the circuit anatomy and pathophysiology of HD to guide the development of future cures. It is now within our grasp to modulate most cell specific processes in a neural circuit (Mohan, 2017), knock down unwanted proteins (Cheng et al., 2015), induce gene expression (Kells et al., 2008) and even edit the genome *in vivo* (Hwang et al., 2013). Determining which techniques are most likely to succeed by accurately targeting the earliest pathological changes to the circuit has never been more important given this plethora of possibilities. It is easier than ever to get lost in a technique and to fall into the trap of treating every problem like it is a nail because the only tool we know how to use is a hammer. The challenge of future experiments will be to link the proposed pathophysiological mechanisms (hyperexcitability, deafferentation, loss of trophic factors and their receptors, impaired vesicular transport, protein transcription, calcium signaling, prion effects and inappropriate immune activation), target common elements and get at the root of the problem in HD and various other neurodegenerative diseases.

The PF is a relatively small structure compared to the cortex or striatum but has far reaching projections to the striatum, cortex, subthalamic nucleus and substantia nigra (Parr-Brownlie et al., 2009; Sadikot et al., 1992a). These connections may be harnessed to deliver pro-survival factors throughout vulnerable circuits and potentially prevent neurodegeneration in diseases like HD,

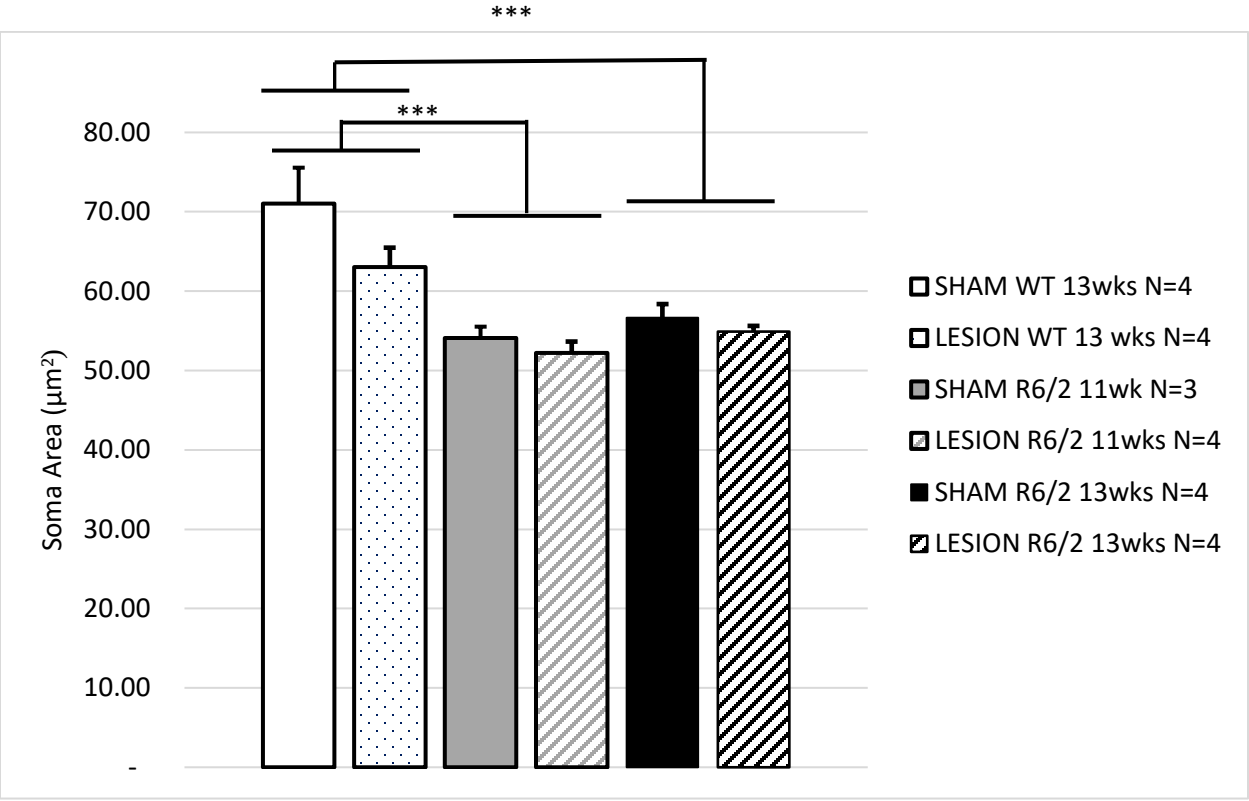
Parkinson's, PSP, and Alzheimer's disease. We propose that the target molecules may include BDNF or NGF expressing viruses, trkB agonists, NMDA receptors, and *htt* knock-down molecules. In summary, science has just scratched the surface and more work is needed to find effective treatments for neurodegenerative diseases.

Appendix

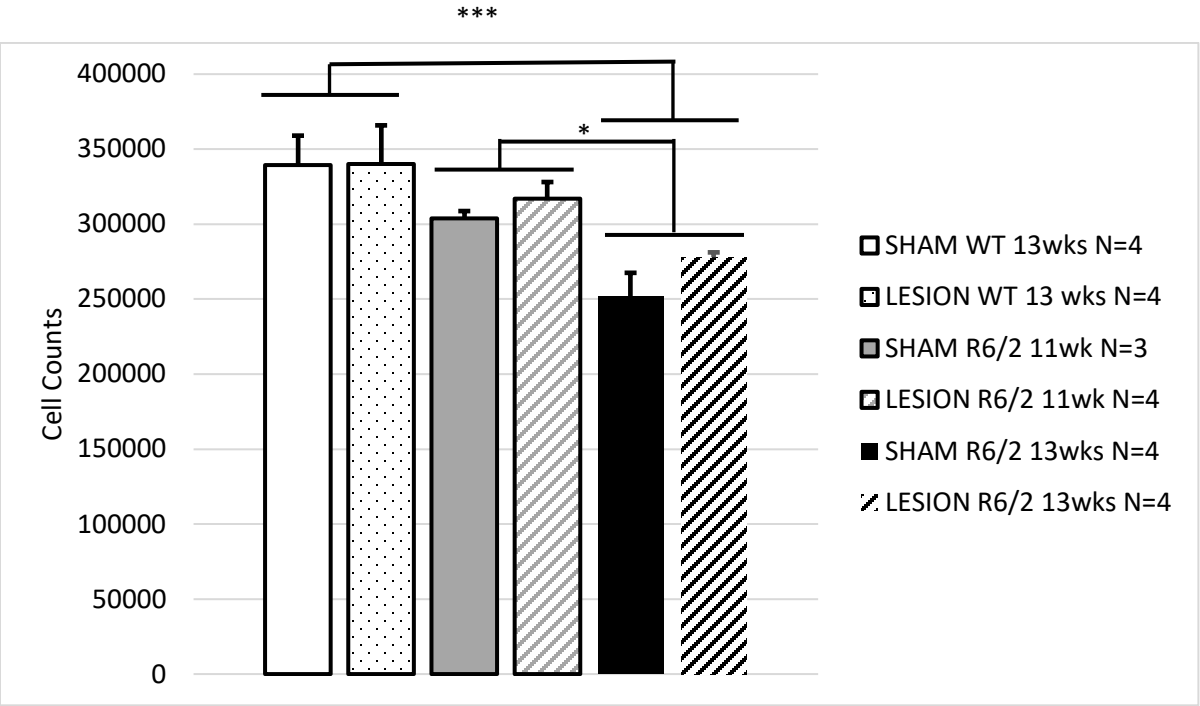
A1.1 Experimental Timeline



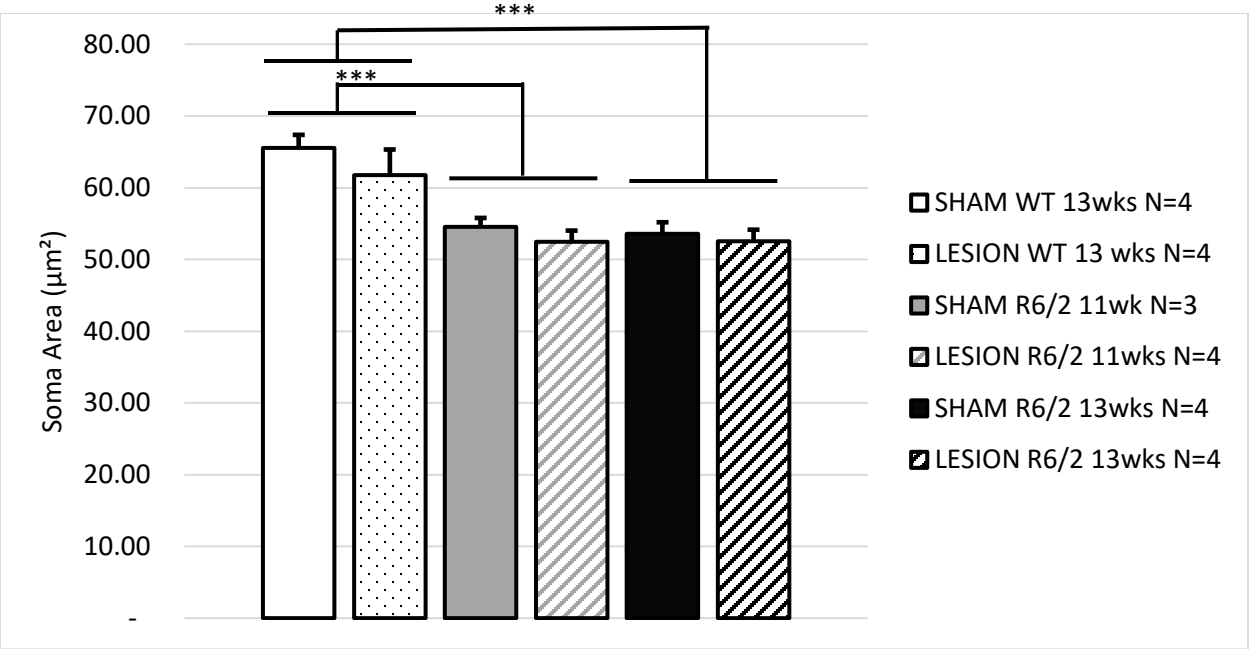
A1.2 Supplementary Figures



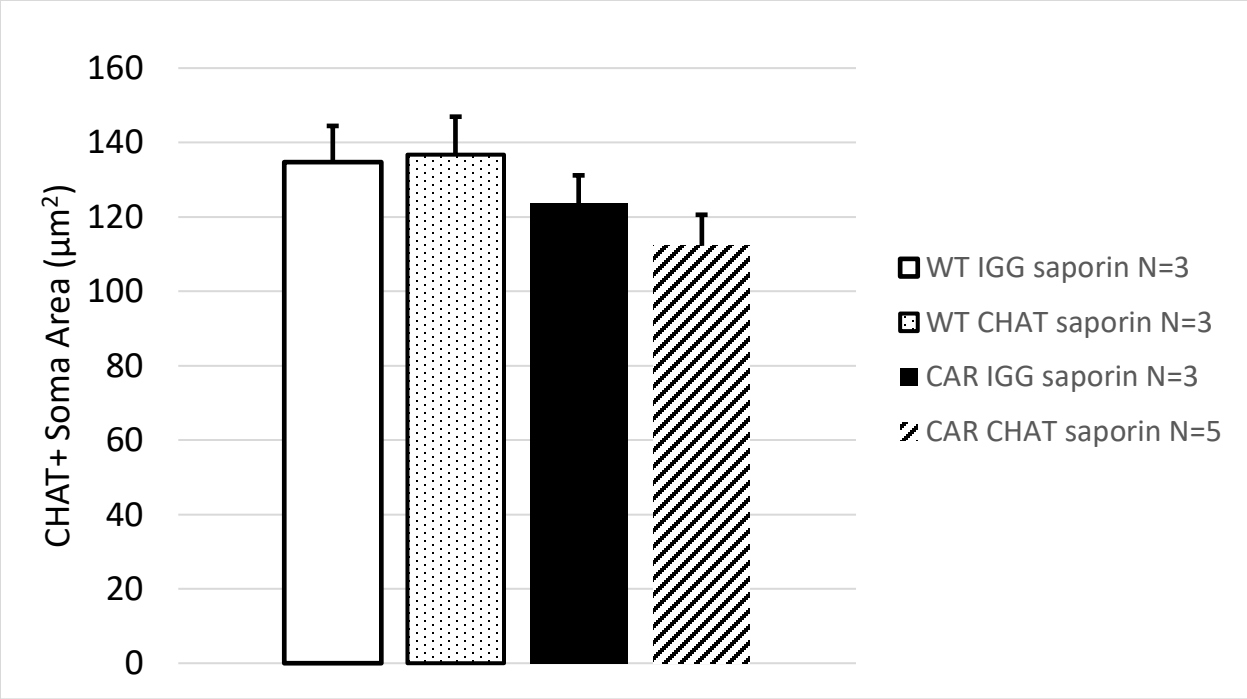
Supplementary Fig. 1: Matrix Neuron Soma Area in WT and R6/2 mice. Demonstrates a reduction in the R6/2 soma area of matrix neurons compared to WT at both 11 and 13wks with no significant effect of PF lesion. A 2-way between subject ANOVA was applied to the data and was followed by a Tukey HSD *post hoc* test where appropriate. ***$p<0.001$.



Supplementary Fig. 2: Striosome cell count in WT and R6/2 mice. Demonstrates a trend to reduction in number of striosome neurons is seen in R6/2 compared WT mice that was not significant (~8% decrease, $p=0.23$) at 11wks but was significant ($p<0.05$) at 13wks. There was no significant effect of thalamic lesion on striosome neuron number. A 2-way between subject ANOVA was applied, followed by a Tukey HSD *post hoc* test.*$p<0.05$; ***$p<0.001$.



Supplementary Fig. 3: Striosome Neuron Soma Area in WT and R6/2. Demonstrates a reduction in soma area of striosome neurons in R6/2 animals at both 11 and 13 wks compared to WT. There is no significant effect of lesion on striosome neuron soma area. A 2-way between subject ANOVA was applied followed by a Tukey HSD *post hoc* test. ***$p<0.001$.



Supplementary Fig. 4: CHAT+ Cell Soma Area in Saporin Treated Animals. No significant difference between remaining CHAT+ cells in either WT or R6/2 animals treated with saporin toxins. A 2-way between subject ANOVA was applied to the data and was followed by a Tukey HSD *post hoc* test.

References

(1993). A novel gene containing a trinucleotide repeat that is expanded and unstable on Huntington's disease chromosomes. The Huntington's Disease Collaborative Research Group. Cell *72*, 971-983.

Abiru, Y., Nishio, C., and Hatanaka, H. (1996a). The survival of striatal cholinergic neurons cultured from postnatal 2-week-old rats is promoted by neurotrophins. Brain Res Dev Brain Res *91*, 260-267.

Abiru, Y., Nishio, C., and Hatanaka, H. (1996b). The survival of striatal cholinergic neurons cultured from postnatal 2-week-old rats is promoted by neurotrophins. Developmental Brain Research *91*, 260-267.

Acklin, S.E., and van der Kooy, D. (1993). Clonal heterogeneity in the germinal zone of the developing rat telencephalon. Development *118*, 175-192.

Airaksinen, M.S., and Panula, P. (1988). The histaminergic system in the guinea pig central nervous system: an immunocytochemical mapping study using an antiserum against histamine. J Comp Neurol *273*, 163-186.

Albin, R.L., Reiner, A., Anderson, K.D., Penney, J.B., and Young, A.B. (1990a). Striatal and nigral neuron subpopulations in rigid Huntington's disease: implications for the functional anatomy of chorea and rigidity-akinesia. Ann Neurol *27*, 357-365.

Albin, R.L., Young, A.B., and Penney, J.B. (1989). The functional anatomy of basal ganglia disorders. Trends Neurosci *12*, 366-375.

Albin, R.L., Young, A.B., Penney, J.B., Handelin, B., Balfour, R., Anderson, K.D., Markel, D.S., Tourtellotte, W.W., and Reiner, A. (1990b). Abnormalities of striatal projection neurons and N-methyl-D-aspartate receptors in presymptomatic Huntington's disease. N Engl J Med *322*, 1293-1298.

Ali, N.J., and Levine, M.S. (2006). Changes in expression of N-methyl-D-aspartate receptor subunits occur early in the R6/2 mouse model of Huntington's disease. Dev Neurosci *28*, 230-238.

Alloway, K.D., Smith, J.B., and Watson, G.D. (2014). Thalamostriatal projections from the medial posterior and parafascicular nuclei have distinct topographic and physiologic properties. Journal of neurophysiology *111*, 36-50.

Altar, C.A., Cai, N., Bliven, T., Juhasz, M., Conner, J.M., Acheson, A.L., Lindsay, R.M., and Wiegand, S.J. (1997). Anterograde transport of brain-derived neurotrophic factor and its role in the brain. Nature *389*, 856-860.

Ambrose, C.M., Duyao, M.P., Barnes, G., Bates, G.P., Lin, C.S., Srinidhi, J., Baxendale, S., Hummerich, H., Lehrach, H., Altherr, M.*, et al.* (1994). Structure and expression of the Huntington's disease gene: Evidence against simple inactivation due to an expanded CAG repeat. Somat Cell Mol Genet *20*, 27-38.

Anborgh, P.H., Godin, C., Pampillo, M., Dhami, G.K., Dale, L.B., Cregan, S.P., Truant, R., and Ferguson, S.S. (2005). Inhibition of metabotropic glutamate receptor signaling by the huntingtin-binding protein optineurin. J Biol Chem *280*, 34840-34848.

Anderson, K.E., Louis, E.D., Stern, Y., and Marder, K.S. (2001). Cognitive correlates of obsessive and compulsive symptoms in Huntington's disease. The American journal of psychiatry *158*, 799-801.

Anzilotti, S., Giampa, C., Laurenti, D., Perrone, L., Bernardi, G., Melone, M.A., and Fusco, F.R. (2012). Immunohistochemical localization of receptor for advanced glycation end (RAGE) products in the R6/2 mouse model of Huntington's disease. Brain Res Bull *87*, 350-358.

Aoki, S., Liu, A.W., Zucca, A., Zucca, S., and Wickens, J.R. (2015). Role of Striatal Cholinergic Interneurons in Set-Shifting in the Rat. J Neurosci *35*, 9424-9431.

Aosaki, T., Kimura, M., and Graybiel, A.M. (1995). Temporal and spatial characteristics of tonically active neurons of the primate's striatum. Journal of neurophysiology *73*, 1234-1252.

Avanzino, L., and Fiorio, M. (2014). Proprioceptive dysfunction in focal dystonia: from experimental evidence to rehabilitation strategies. Front Hum Neurosci *8*, 1000.

Ayloo, S., Guedes-Dias, P., Ghiretti, A.E., and Holzbaur, E.L.F. (2017). Dynein efficiently navigates the dendritic cytoskeleton to drive the retrograde trafficking of BDNF/TrkB signaling endosomes. Molecular biology of the cell *28*, 2543-2554.

Aylward, E.H., Nopoulos, P.C., Ross, C.A., Langbehn, D.R., Pierson, R.K., Mills, J.A., Johnson, H.J., Magnotta, V.A., Juhl, A.R., Paulsen, J.S*., et al.* (2011). Longitudinal change in regional brain volumes in prodromal Huntington disease. J Neurol Neurosurg Psychiatry *82*, 405-410.

Bacci, J.J., Goff, L.K.L., and Salin, P. (2002). Effects of intralaminar thalamic nuclei lesion on glutamic acid decarboxylase (GAD65 and GAD67) and cytochrome oxidase subunit I mRNA expression in the basal ganglia of the rat. European Journal of Neuroscience *15*, 1918-1928.

Bailey, C.D., and Johnson, G.V. (2005). Tissue transglutaminase contributes to disease progression in the R6/2 Huntington's disease mouse model via aggregate-independent mechanisms. J Neurochem *92*, 83-92.

Bailey, C.D., and Johnson, G.V. (2006). The protective effects of cystamine in the R6/2 Huntington's disease mouse involve mechanisms other than the inhibition of tissue transglutaminase. Neurobiol Aging *27*, 871-879.

Bailoo, J.D., Bohlen, M.O., and Wahlsten, D. (2010). The precision of video and photocell tracking systems and the elimination of tracking errors with infrared backlighting. J Neurosci Methods *188*, 45-52.

Bao, L., Patel, J.C., Walker, R.H., Shashidharan, P., and Rice, M.E. (2010). Dysregulation of striatal dopamine release in a mouse model of dystonia. J Neurochem *114*, 1781-1791.

Baquet, Z.C., Gorski, J.A., and Jones, K.R. (2004). Early Striatal Dendrite Deficits followed by Neuron Loss with Advanced Age in the Absence of Anterograde Cortical Brain-Derived Neurotrophic Factor. The Journal of Neuroscience *24*, 4250-4258.

Bartkowska, K., Turlejski, K., and Djavadian, R.L. (2010). Neurotrophins and their receptors in early development of the mammalian nervous system. Acta neurobiologiae experimentalis *70*, 454-467.

Baydyuk, M., Xie, Y., Tessarollo, L., and Xu, B. (2013). Midbrain-Derived Neurotrophins Support Survival of Immature Striatal Projection Neurons. The Journal of Neuroscience *33*, 3363-3369.

Baydyuk, M., and Xu, B. (2014). BDNF signaling and survival of striatal neurons. Frontiers in cellular neuroscience *8*, 254.

Bayram-Weston, Z., Jones, L., Dunnett, S.B., and Brooks, S.P. (2012). Light and electron microscopic characterization of the evolution of cellular pathology in YAC128 Huntington's disease transgenic mice. Brain Res Bull *88*, 137-147.

Beal, M.F., Ferrante, R., Swartz, K., and Kowall, N. (1991). Chronic quinolinic acid lesions in rats closely resemble Huntington's disease. Journal of Neuroscience *11*, 1649-1659.

Beal, M.F., Kowall, N.W., Ellison, D.W., Mazurek, M.F., Swartz, K.J., and Martin, J.B. (1986). Replication of the neurochemical characteristics of Huntington's disease by quinolinic acid. Nature *321*, 168.

Beckstead, R.M. (1984). A projection to the striatum from the medial subdivision of the posterior group of the thalamus in the cat. Brain Res *300*, 351-356.

Behar, T.N., Scott, C.A., Greene, C.L., Wen, X., Smith, S.V., Maric, D., Liu, Q.Y., Colton, C.A., and Barker, J.L. (1999). Glutamate acting at NMDA receptors stimulates embryonic cortical neuronal migration. J Neurosci *19*, 4449-4461.

Bemelmans, A.-P., Horellou, P., Pradier, L., Brunet, I., Colin, P., and Mallet, J. (1999). Brain-Derived Neurotrophic Factor-Mediated Protection of Striatal Neurons in an Excitotoxic Rat Model of Huntington's Disease, as Demonstrated by Adenoviral Gene Transfer. Human gene therapy *10*, 2987-2997.

Bennett, B.D., and Bolam, J.P. (1993). Characterization of calretinin-immunoreactive structures in the striatum of the rat. Brain Res *609*, 137-148.

Bennett, B.D., and Bolam, J.P. (1994). Synaptic input and output of parvalbumin-immunoreactive neurons in the neostriatum of the rat. Neuroscience *62*, 707-719.

Berendse, H.W., and Groenewegen, H.J. (1990). Organization of the thalamostriatal projections in the rat, with special emphasis on the ventral striatum. J Comp Neurol *299*, 187-228.

Berendse, H.W., and Groenewegen, H.J. (1991). Restricted cortical termination fields of the midline and intralaminar thalamic nuclei in the rat. Neuroscience *42*, 73-102.

Bernacer, J., Prensa, L., and Gimenez-Amaya, J.M. (2012). Distribution of GABAergic interneurons and dopaminergic cells in the functional territories of the human striatum. PLoS One *7*, e30504.

Bezard, E., Boraud, T., Bioulac, B., and Gross, C.E. (1997). Compensatory effects of glutamatergic inputs to the substantia nigra pars compacta in experimental Parkinsonism. Neuroscience *81*, 399-404.

Bhatia, K.P., and Marsden, C.D. (1994). The behavioural and motor consequences of focal lesions of the basal ganglia in man. Brain *117*, 859-876.

Blood, A.J., Kuster, J.K., Woodman, S.C., Kirlic, N., Makhlouf, M.L., Multhaupt-Buell, T.J., Makris, N., Parent, M., Sudarsky, L.R., Sjalander, G.*, et al.* (2012). Evidence for altered basal ganglia-brainstem connections in cervical dystonia. PLoS One *7*, e31654.

Bolam, J.P., and Ellender, T.J. (2016). Histamine and the striatum. Neuropharmacology *106*, 74-84.

Bolam, J.P., Hanley, J.J., Booth, P.A., and Bevan, M.D. (2000). Synaptic organisation of the basal ganglia. Journal of anatomy *196 (Pt 4)*, 527-542.

Bolam, J.P., Ingham, C.A., Izzo, P.N., Levey, A.I., Rye, D.B., Smith, A.D., and Wainer, B.H. (1986). Substance P-containing terminals in synaptic contact with cholinergic neurons in the neostriatum and basal forebrain: a double immunocytochemical study in the rat. Brain Res *397*, 279-289.

Bolam, J.P., Izzo, P.N., and Graybiel, A.M. (1988). Cellular substrate of the histochemically defined striosome/matrix system of the caudate nucleus: a combined Golgi and immunocytochemical study in cat and ferret. Neuroscience *24*, 853-875.

Bolam, J.P., Wainer, B.H., and Smith, A.D. (1984). Characterization of cholinergic neurons in the rat neostriatum. A combination of choline acetyltransferase immunocytochemistry, Golgi-impregnation and electron microscopy. Neuroscience *12*, 711-718.

Bostan, A.C., Dum, R.P., and Strick, P.L. (2010). The basal ganglia communicate with the cerebellum. Proceedings of the National Academy of Sciences *107*, 8452-8456.

Bostan, A.C., and Strick, P.L. (2010). The cerebellum and basal ganglia are interconnected. Neuropsychology review *20*, 261-270.

Braak, H., Del Tredici, K., Rub, U., de Vos, R.A., Jansen Steur, E.N., and Braak, E. (2003). Staging of brain pathology related to sporadic Parkinson's disease. Neurobiol Aging *24*, 197-211.

Bracci, E., Centonze, D., Bernardi, G., and Calabresi, P. (2002). Dopamine excites fast-spiking interneurons in the striatum. Journal of neurophysiology *87*, 2190-2194.

Bradfield, L.A., and Balleine, B.W. (2017). Thalamic Control of Dorsomedial Striatum Regulates Internal State to Guide Goal-Directed Action Selection. J Neurosci *37*, 3721-3733.

Bradfield, L.A., Bertran-Gonzalez, J., Chieng, B., and Balleine, B.W. (2013). The thalamostriatal pathway and cholinergic control of goal-directed action: interlacing new with existing learning in the striatum. Neuron *79*, 153-166.

Braunstein, K.E., Eschbach, J., Ròna-Vörös, K., Soylu, R., Mikrouli, E., Larmet, Y., René, F., De Aguilar, J.-L.G., Loeffler, J.-P., Müller, H.-P.*, et al.* (2010). A point mutation in the dynein heavy chain gene leads to striatal atrophy and compromises neurite outgrowth of striatal neurons. Hum Mol Genet *19*, 4385-4398.

Brazel, C.Y., Nunez, J.L., Yang, Z., and Levison, S.W. (2005). Glutamate enhances survival and proliferation of neural progenitors derived from the subventricular zone. Neuroscience *131*, 55-65.

Breakefield, X.O., Blood, A.J., Li, Y., Hallett, M., Hanson, P.I., and Standaert, D.G. (2008). The pathophysiological basis of dystonias. Nature Reviews Neuroscience *9*, 222.

Brezun, J.M., and Daszuta, A. (1999). Depletion in serotonin decreases neurogenesis in the dentate gyrus and the subventricular zone of adult rats. Neuroscience *89*, 999-1002.

Brooks, D., and Halliday, G.M. (2009). Intralaminar nuclei of the thalamus in Lewy body diseases. Brain Res Bull *78*, 97-104.

Brown, H.D., Baker, P.M., and Ragozzino, M.E. (2010). The parafascicular thalamic nucleus concomitantly influences behavioral flexibility and dorsomedial striatal acetylcholine output in rats. J Neurosci *30*, 14390-14398.

Cachope, R., Mateo, Y., Mathur, B.N., Irving, J., Wang, H.L., Morales, M., Lovinger, D.M., and Cheer, J.F. (2012). Selective activation of cholinergic interneurons enhances accumbal phasic dopamine release: setting the tone for reward processing. Cell Rep *2*, 33-41.

Calabresi, P., Centonze, D., Gubellini, P., Pisani, A., and Bernardi, G. (2000). Acetylcholine-mediated modulation of striatal function. Trends in Neurosciences *23*, 120-126.

Calabresi, P., Centonze, D., Pisani, A., Sancesario, G., North, R.A., and Bernardi, G. (1998). Muscarinic IPSPs in rat striatal cholinergic interneurones. The Journal of physiology *510*, 421-427.

Calabresi, P., Picconi, B., Tozzi, A., Ghiglieri, V., and Di Filippo, M. (2014). Direct and indirect pathways of basal ganglia: a critical reappraisal. Nature neuroscience *17*, 1022.

Cameron, H., McEwen, B., and Gould, E. (1995). Regulation of adult neurogenesis by excitatory input and NMDA receptor activation in the dentate gyrus. The Journal of Neuroscience *15*, 4687-4692.

Canudas, A.M., Pezzi, S., Canals, J.M., Pallas, M., and Alberch, J. (2005). Endogenous brain-derived neurotrophic factor protects dopaminergic nigral neurons against transneuronal degeneration induced by striatal excitotoxic injury. Brain research Molecular brain research *134*, 147-154.

Carter, R.J., Lione, L.A., Humby, T., Mangiarini, L., Mahal, A., Bates, G.P., Dunnett, S.B., and Morton, A.J. (1999). Characterization of progressive motor deficits in mice transgenic for the human Huntington's disease mutation. J Neurosci *19*, 3248-3257.

Cattaneo, E., Zuccato, C., and Tartari, M. (2005). Normal huntingtin function: an alternative approach to Huntington's disease. Nature Reviews Neuroscience *6*, 919.

Causing, C.G., Gloster, A., Aloyz, R., Bamji, S.X., Chang, E., Fawcett, J., Kuchel, G., and Miller, F.D. (1997). Synaptic Innervation Density Is Regulated by Neuron-Derived BDNF. Neuron *18*, 257-267.

Centonze, D., Grande, C., Usiello, A., Gubellini, P., Erbs, E., Martín, A.B., Pisani, A., Tognazzi, N., Bernardi, G., Moratalla, R*., et al.* (2003). Receptor Subtypes Involved in the Presynaptic and Postsynaptic Actions of Dopamine on Striatal Interneurons. The Journal of Neuroscience *23*, 6245-6254.

Chakraborty, A., Murphy, S., and Coleman, N. (2017). The Role of NMDA Receptors in Neural Stem Cell Proliferation and Differentiation. Stem cells and development *26*, 798-807.

Chang, H.T., Wilson, C.J., and Kitai, S.T. (1981). Single neostriatal efferent axons in the globus pallidus: a light and electron microscopic study. Science *213*, 915.

Chao, M.V. (2003). Neurotrophins and their receptors: a convergence point for many signalling pathways. Nat Rev Neurosci *4*, 299-309.

Chen, L., Chatterjee, M., and Li, J.Y.H. (2010). The Mouse Homeobox Gene Gbx2 Is Required for the Development of Cholinergic Interneurons in the Striatum. The Journal of Neuroscience *30*, 14824-14834.

Cheng, H.-M., Chern, Y., Chen, I.H., Liu, C.-R., Li, S.-H., Chun, S.J., Rigo, F., Bennett, C.F., Deng, N., Feng, Y.*, et al.* (2015). Effects on Murine Behavior and Lifespan of Selectively Decreasing Expression of Mutant Huntingtin Allele by Supt4h Knockdown. PLoS Genet *11*, e1005043.

Chesselet, M.F., and Graybiel, A.M. (1986). Striatal neurons expressing somatostatin-like immunoreactivity: evidence for a peptidergic interneuronal system in the cat. Neuroscience *17*, 547-571.

Cicchetti, F., Lacroix, S., Cisbani, G., Vallières, N., Saint-Pierre, M., St-Amour, I., Tolouei, R., Skepper, J.N., Hauser, R.A., Mantovani, D.*, et al.* (2014). Mutant huntingtin is present in neuronal grafts in huntington disease patients. Ann Neurol *76*, 31-42.

Cicchetti, F., and Parent, A. (1996). Striatal interneurons in Huntington's disease: selective increase in the density of calretinin-immunoreactive medium-sized neurons. Mov Disord *11*, 619-626.

Cicchetti, F., Saporta, S., Hauser, R.A., Parent, M., Saint-Pierre, M., Sanberg, P.R., Li, X.J., Parker, J.R., Chu, Y., Mufson, E.J.*, et al.* (2009). Neural transplants in patients with Huntington's disease undergo disease-like neuronal degeneration. Proceedings of the National Academy of Sciences *106*, 12483-12488.

Cirillo, G., Maggio, N., Bianco, M.R., Vollono, C., Sellitti, S., and Papa, M. (2010). Discriminative behavioral assessment unveils remarkable reactive astrocytosis and early molecular correlates in basal ganglia of 3-nitropropionic acid subchronic treated rats. Neurochem Int *56*, 152-160.

Cohen, M.S., Bas Orth, C., Kim, H.J., Jeon, N.L., and Jaffrey, S.R. (2011). Neurotrophin-mediated dendrite-to-nucleus signaling revealed by microfluidic compartmentalization of dendrites. Proc Natl Acad Sci U S A *108*, 11246-11251.

Conner, J.M., Lauterborn, J.C., Yan, Q., Gall, C.M., and Varon, S. (1997). Distribution of brain-derived neurotrophic factor (BDNF) protein and mRNA in the normal adult rat CNS: evidence for anterograde axonal transport. J Neurosci *17*, 2295-2313.

Cornwall, J., and Phillipson, O.T. (1988). Afferent projections to the parafascicular thalamic nucleus of the rat, as shown by the retrograde transport of wheat germ agglutinin. Brain Res Bull *20*, 139-150.

Cowan, R.L., and Wilson, C.J. (1994). Spontaneous firing patterns and axonal projections of single corticostriatal neurons in the rat medial agranular cortex. Journal of neurophysiology *71*, 17-32.

Cowan, R.L., Wilson, C.J., Emson, P.C., and Heizmann, C.W. (1990). Parvalbumin-containing GABAergic interneurons in the rat neostriatum. J Comp Neurol *302*, 197-205.

Coyle, J., Price, D., and DeLong, M. (1983). Alzheimer's disease: a disorder of cortical cholinergic innervation. Science *219*, 1184-1190.

Coyle, J.T., and Schwarcz, R. (1976). Lesion of striatal neurons with kainic acid provides a model for Huntington's chorea. Nature *263*, 244.

Cramer, S.C., Sampat, A., Haske-Palomino, M., Nguyen, S., Procaccio, V., and Hermanowicz, N. (2010). Increased prevalence of val66met BDNF genotype among subjects with cervical dystonia. Neurosci Lett *468*, 42-45.

Crotti, A., Benner, C., Kerman, B.E., Gosselin, D., Lagier-Tourenne, C., Zuccato, C., Cattaneo, E., Gage, F.H., Cleveland, D.W., and Glass, C.K. (2014). Mutant Huntingtin promotes autonomous microglia activation via myeloid lineage-determining factors. Nature neuroscience *17*, 513-521.

D'Abreu, A., Jr, M.C.F., Yasuda, C.L., Souza, M.S.A., Lopes-Cendes, Í., and Cendes, F. (2011). Thalamic Volume and Dystonia in Machado–Joseph Disease. Journal of Neuroimaging *21*, e91-e93.

Dau, A., Gladding, C.M., Sepers, M.D., and Raymond, L.A. (2014). Chronic blockade of extrasynaptic NMDA receptors ameliorates synaptic dysfunction and pro-death signaling in Huntington disease transgenic mice. Neurobiol Dis *62*, 533-542.

de Carvalho Aguiar, P., Sweadner, K.J., Penniston, J.T., Zaremba, J., Liu, L., Caton, M., Linazasoro, G., Borg, M., Tijssen, M.A., Bressman, S.B*., et al.* (2004). Mutations in the Na+/K+ -ATPase alpha3 gene ATP1A3 are associated with rapid-onset dystonia parkinsonism. Neuron *43*, 169-175.

de Carvalho Aguiar, P.M., and Ozelius, L.J. (2002). Classification and genetics of dystonia. Lancet Neurol *1*, 316-325.

de Tommaso, M., Difruscolo, O., Sciruicchio, V., Specchio, N., and Livrea, P. (2007). Two Years' Follow-up of Rivastigmine Treatment in Huntington Disease. Clinical neuropharmacology *30*, 43-46.

de Tommaso, M., Specchio, N., Sciruicchio, V., Difruscolo, O., and Specchio, L.M. (2004). Effects of rivastigmine on motor and cognitive impairment in Huntington's disease. Movement Disorders *19*, 1516-1518.

Dedeoglu, A., Kubilus, J.K., Jeitner, T.M., Matson, S.A., Bogdanov, M., Kowall, N.W., Matson, W.R., Cooper, A.J., Ratan, R.R., Beal, M.F*., et al.* (2002). Therapeutic effects of cystamine in a murine model of Huntington's disease. J Neurosci *22*, 8942-8950.

Dehay, C., Savatier, P., Cortay, V., and Kennedy, H. (2001). Cell-Cycle Kinetics of Neocortical Precursors Are Influenced by Embryonic Thalamic Axons. The Journal of Neuroscience *21*, 201-214.

del Toro, D., Canals, J.M., Gines, S., Kojima, M., Egea, G., and Alberch, J. (2006). Mutant huntingtin impairs the post-Golgi trafficking of brain-derived neurotrophic factor but not its Val66Met polymorphism. J Neurosci *26*, 12748-12757.

Del Tredici, K., and Braak, H. (2013). Dysfunction of the locus coeruleus–norepinephrine system and related circuitry in Parkinson's disease-related dementia. Journal of Neurology, Neurosurgery & Psychiatry *84*, 774.

Demierre, B., and Rondot, P. (1983). Dystonia caused by putamino-capsulo-caudate vascular lesions. Journal of Neurology, Neurosurgery & Psychiatry *46*, 404-409.

Deng, Y.P., and Reiner, A. (2016). Cholinergic interneurons in the Q140 knockin mouse model of Huntington's disease: Reductions in dendritic branching and thalamostriatal input. J Comp Neurol *524*, 3518-3529.

Deng, Y.P., Wong, T., Bricker-Anthony, C., Deng, B., and Reiner, A. (2013). Loss of corticostriatal and thalamostriatal synaptic terminals precedes striatal projection neuron pathology in heterozygous Q140 Huntington's disease mice. Neurobiol Dis *60*, 89-107.

Deng, Y.P., Wong, T., Wan, J.Y., and Reiner, A. (2014). Differential loss of thalamostriatal and corticostriatal input to striatal projection neuron types prior to overt motor symptoms in the Q140 knock-in mouse model of Huntington's disease. Front Syst Neurosci *8*, 198.

DeVito, J.L., and Anderson, M.E. (1982). An autoradiographic study of efferent connections of the globus pallidus in Macaca mulatta. Exp Brain Res *46*, 107-117.

DeVito, J.L., Anderson, M.E., and Walsh, K.E. (1980). A horseradish peroxidase study of afferent connections of the globus pallidus in Macaca mulatta. Exp Brain Res *38*, 65-73.

DiFiglia, M., Sapp, E., Chase, K., Schwarz, C., Meloni, A., Young, C., Martin, E., Vonsattel, J.-P., Carraway, R., and Reeves, S.A. (1995). Huntingtin is a cytoplasmic protein associated with vesicles in human and rat brain neurons. Neuron *14*, 1075-1081.

Ding, J., Guzman, J.N., Tkatch, T., Chen, S., Goldberg, J.A., Ebert, P.J., Levitt, P., Wilson, C.J., Hamm, H.E., and Surmeier, D.J. (2006). RGS4-dependent attenuation of M4 autoreceptor function in striatal cholinergic interneurons following dopamine depletion. Nature neuroscience *9*, 832.

Ding, J.B., Guzman, J.N., Peterson, J.D., Goldberg, J.A., and Surmeier, D.J. (2010). Thalamic gating of corticostriatal signaling by cholinergic interneurons. Neuron *67*, 294-307.

Doig, N.M., Magill, P.J., Apicella, P., Bolam, J.P., and Sharott, A. (2014). Cortical and thalamic excitation mediate the multiphasic responses of striatal cholinergic interneurons to motivationally salient stimuli. The Journal of neuroscience : the official journal of the Society for Neuroscience *34*, 3101-3117.

Doig, N.M., Moss, J., and Bolam, J.P. (2010). Cortical and Thalamic Innervation of Direct and Indirect Pathway Medium-Sized Spiny Neurons in Mouse Striatum. The Journal of Neuroscience *30*, 14610.

Donoghue, J.P., and Herkenham, M. (1986). Neostriatal projections from individual cortical fields conform to histochemically distinct striatal compartments in the rat. Brain Res *365*, 397-403.

Dooley, A.E., Pappas, I.S., and Parnavelas, J.G. (1997). Serotonin Promotes the Survival of Cortical Glutamatergic Neuronsin Vitro. Exp Neurol *148*, 205-214.

Dougherty, S.E., Hollimon, J.J., McMeekin, L.J., Bohannon, A.S., West, A.B., Lesort, M., Hablitz, J.J., and Cowell, R.M. (2014). Hyperactivity and cortical disinhibition in mice with restricted expression of mutant huntingtin to parvalbumin-positive cells. Neurobiol Dis *62*, 160-171.

Dougherty, S.E., Reeves, J.L., Lesort, M., Detloff, P.J., and Cowell, R.M. (2013). Purkinje cell dysfunction and loss in a knock-in mouse model of Huntington disease. Exp Neurol *240*, 96-102.

Dougherty, S.E., Reeves, J.L., Lucas, E.K., Gamble, K.L., Lesort, M., and Cowell, R.M. (2012). Disruption of Purkinje cell function prior to huntingtin accumulation and cell loss in an animal model of Huntington disease. Exp Neurol *236*, 171-178.

Dragatsis, I., Levine, M.S., and Zeitlin, S. (2000). Inactivation of Hdh in the brain and testis results in progressive neurodegeneration and sterility in mice. Nat Genet *26*, 300-306.

Dresel, C., Li, Y., Wilzeck, V., Castrop, F., Zimmer, C., and Haslinger, B. (2014). Multiple changes of functional connectivity between sensorimotor areas in focal hand dystonia. J Neurol Neurosurg Psychiatry *85*, 1245-1252.

Dube, L., Smith, A.D., and Bolam, J.P. (1988). Identification of synaptic terminals of thalamic or cortical origin in contact with distinct medium-size spiny neurons in the rat neostriatum. J Comp Neurol *267*, 455-471.

Duchen, L.W. (1976). Dystonia musculorum--an inherited disease of the nervous system in the mouse. Adv Neurol *14*, 353-365.

Duchen, L.W., Strich, S.J., and Falconer, D.S. (1964). Clinical and Pathological Studies of an Hereditary Neuropathy in Mice (Dystonia Musculorum). Brain *87*, 367-378.

Duyao, M., Auerbach, A., Ryan, A., Persichetti, F., Barnes, G., McNeil, S., Ge, P., Vonsattel, J., Gusella, J., Joyner, A.*, et al.* (1995a). Inactivation of the mouse Huntington's disease gene homolog Hdh. Science *269*, 407-410.

Duyao, M.P., Auerbach, A.B., Ryan, A., Persichetti, F., Barnes, G.T., McNeil, S.M., Ge, P., Vonsattel, J.P., Gusella, J.F., Joyner, A.L.*, et al.* (1995b). Inactivation of the mouse Huntington's disease gene homolog Hdh. Science *269*, 407-410.

Eblen, F., and Graybiel, A.M. (1995). Highly restricted origin of prefrontal cortical inputs to striosomes in the macaque monkey. J Neurosci *15*, 5999-6013.

Edalatmanesh, M.A., Nikfarjam, H., Moghadas, M., Haddad-Mashadrizeh, A., Robati, R., and Hashemzadeh, M.R. (2014). Histopathological and behavioral assessment of toxin-produced cerebellar lesion: a potent model for cell transplantation studies in the cerebellum. Cell J *16*, 325-334.

Ellender, T.J., Harwood, J., Kosillo, P., Capogna, M., and Bolam, J.P. (2013). Heterogeneous properties of central lateral and parafascicular thalamic synapses in the striatum. The Journal of physiology *591*, 257-272.

Epping, E.A., and Paulsen, J.S. (2011). Depression in the early stages of Huntington disease. Neurodegener Dis Manag *1*, 407-414.

Esapa, C.T., Waite, A., Locke, M., Benson, M.A., Kraus, M., McIlhinney, R.A.J., Sillitoe, R.V., Beesley, P.W., and Blake, D.J. (2007). SGCE missense mutations that cause myoclonus-dystonia syndrome impair ε-sarcoglycan trafficking to the plasma membrane: modulation by ubiquitination and torsinA. Hum Mol Genet *16*, 327-342.

Eskow Jaunarajs, K.L., Bonsi, P., Chesselet, M.F., Standaert, D.G., and Pisani, A. (2015). Striatal cholinergic dysfunction as a unifying theme in the pathophysiology of dystonia. Progress in Neurobiology *127-128*, 91-107.

Espay, A.J., Morgante, F., Purzner, J., Gunraj, C.A., Lang, A.E., and Chen, R. (2006). Cortical and spinal abnormalities in psychogenic dystonia. Ann Neurol *59*, 825-834.

Farrar, A.M., Callahan, J.W., and Abercrombie, E.D. (2011). Reduced striatal acetylcholine efflux in the R6/2 mouse model of Huntington's disease: an examination of the role of altered inhibitory and excitatory mechanisms. Exp Neurol *232*, 119-125.

Fawcett, J.P., Bamji, S.X., Causing, C.G., Aloyz, R., Ase, A.R., Reader, T.A., McLean, J.H., and Miller, F.D. (1998). Functional Evidence that BDNF Is an Anterograde Neuronal Trophic Factor in the CNS. The Journal of Neuroscience *18*, 2808-2821.

Fenelon, G., Francois, C., Percheron, G., and Yelnik, J. (1991). Topographic distribution of the neurons of the central complex (centre median-parafascicular complex) and of other thalamic neurons projecting to the striatum in macaques. Neuroscience *45*, 495-510.

Fernagut, P.O., Diguet, E., Stefanova, N., Biran, M., Wenning, G.K., Canioni, P., Bioulac, B., and Tison, F. (2002). Subacute systemic 3-nitropropionic acid intoxication induces a distinct motor disorder in adult C57Bl/6 mice: behavioural and histopathological characterisation. Neuroscience *114*, 1005-1017.

Fernandes, H.B., Baimbridge, K.G., Church, J., Hayden, M.R., and Raymond, L.A. (2007). Mitochondrial sensitivity and altered calcium handling underlie enhanced NMDA-induced apoptosis in YAC128 model of Huntington's disease. J Neurosci *27*, 13614-13623.

Fernandez-Nogales, M., Santos-Galindo, M., Hernandez, I.H., Cabrera, J.R., and Lucas, J.J. (2016). Faulty splicing and cytoskeleton abnormalities in Huntington's disease. Brain Pathol *26*, 772-778.

Ferrante, R.J., Beal, M.F., Kowall, N.W., Richardson, E.P., Jr., and Martin, J.B. (1987a). Sparing of acetylcholinesterase-containing striatal neurons in Huntington's disease. Brain Res *411*, 162-166.

Ferrante, R.J., Gutekunst, C.-A., Persichetti, F., McNeil, S.M., Kowall, N.W., Gusella, J.F., MacDonald, M.E., Beal, M.F., and Hersch, S.M. (1997). Heterogeneous topographic and cellular distribution of huntingtin expression in the normal human neostriatum. Journal of Neuroscience *17*, 3052-3063.

Ferrante, R.J., Kowall, N.W., Beal, M.F., Martin, J.B., Bird, E.D., and Richardson, E.P., Jr. (1987b). Morphologic and histochemical characteristics of a spared subset of striatal neurons in Huntington's disease. J Neuropathol Exp Neurol *46*, 12-27.

Ferrante, R.J., Kowall, N.W., Cipolloni, P.B., Storey, E., and Beal, M.F. (1993). Excitotoxin Lesions in Primates as a Model for Huntington's Disease: Histopathologic and Neurochemical Characterization. Exp Neurol *119*, 46-71.

Fiedorowicz, J.G., Mills, J.A., Ruggle, A., Langbehn, D., Paulsen, J.S., and Group, P.-H.I.o.t.H.S. (2011). Suicidal behavior in prodromal Huntington disease. Neurodegener Dis *8*, 483-490.

Filip, P., Lungu, O.V., and Bares, M. (2013). Dystonia and the cerebellum: a new field of interest in movement disorders? Clin Neurophysiol *124*, 1269-1276.

Fishell, G., Mason, C.A., and Hatten, M.E. (1993). Dispersion of neural progenitors within the germinal zones of the forebrain. Nature *362*, 636-638.

Flaherty, A.W., and Graybiel, A.M. (1994). Input-output organization of the sensorimotor striatum in the squirrel monkey. J Neurosci *14*, 599-610.

Franciosi, S., Ryu, J.K., Shim, Y., Hill, A., Connolly, C., Hayden, M.R., McLarnon, J.G., and Leavitt, B.R. (2012). Age-dependent neurovascular abnormalities and altered microglial morphology in the YAC128 mouse model of Huntington disease. Neurobiol Dis *45*, 438-449.

Francois, C., Percheron, G., Parent, A., Sadikot, A.F., Fenelon, G., and Yelnik, J. (1991). Topography of the projection from the central complex of the thalamus to the sensorimotor striatal territory in monkeys. J Comp Neurol *305*, 17-34.

Franklin, K.B.J., and Paxinos, G. (2008). The mouse brain in stereotaxic coordinates, 3rd edn (Amsterdam ; Boston: Elsevier/Academic Press).

Freeman, W., and Morton, A.J. (2004). Regional and progressive changes in brain expression of complexin II in a mouse transgenic for the Huntington's disease mutation. Brain Res Bull *63*, 45-55.

Fryer, R.H., Kaplan, D.R., Feinstein, S.C., Radeke, M.J., Grayson, D.R., and Kromer, L.F. (1996). Developmental and mature expression of full-length and truncated TrkB, receptors in the rat forebrain. Journal of Comparative Neurology *374*, 21-40.

Fujiyama, F., Sohn, J., Nakano, T., Furuta, T., Nakamura, K.C., Matsuda, W., and Kaneko, T. (2011). Exclusive and common targets of neostriatofugal projections of rat striosome neurons: a single neuron-tracing study using a viral vector. Eur J Neurosci *33*, 668-677.

Fujiyama, F., Takahashi, S., and Karube, F. (2015). Morphological elucidation of basal ganglia circuits contributing reward prediction. Front Neurosci *9*, 6.

Fujiyama, F., Unzai, T., Nakamura, K., Nomura, S., and Kaneko, T. (2006). Difference in organization of corticostriatal and thalamostriatal synapses between patch and matrix compartments of rat neostriatum. Eur J Neurosci *24*, 2813-2824.

Furukawa, Y., Hornykiewicz, O., Fahn, S., and Kish, S.J. (2000). Striatal dopamine in early-onset primary torsion dystonia with the DYT1 mutation. Neurology *54*, 1193-1195.

Fusco, F.R., Anzilotti, S., Giampa, C., Dato, C., Laurenti, D., Leuti, A., Colucci D'Amato, L., Perrone, L., Bernardi, G., and Melone, M.A. (2012). Changes in the expression of extracellular regulated kinase (ERK 1/2) in the R6/2 mouse model of Huntington's disease after phosphodiesterase IV inhibition. Neurobiol Dis *46*, 225-233.

Fusco, F.R., Chen, Q., Lamoreaux, W.J., Figueredo-Cardenas, G., Jiao, Y., Coffman, J.A., Surmeier, D.J., Honig, M.G., Carlock, L.R., and Reiner, A. (1999). Cellular Localization of Huntingtin in Striatal and

Cortical Neurons in Rats: Lack of Correlation with Neuronal Vulnerability in Huntington's Disease. The Journal of Neuroscience *19*, 1189-1202.

Fusco, F.R., Zuccato, C., Tartari, M., Martorana, A., De March, Z., Giampa, C., Cattaneo, E., and Bernardi, G. (2003). Co-localization of brain-derived neurotrophic factor (BDNF) and wild-type huntingtin in normal and quinolinic acid-lesioned rat brain. Eur J Neurosci *18*, 1093-1102.

Galardi, G., Perani, D., Grassi, F., Bressi, S., Amadio, S., Antoni, M., Comi, G.C., Canal, N., and Fazio, F. (1996). Basal ganglia and thalamo-cortical hypermetabolism in patients with spasmodic torticollis. Acta Neurol Scand *94*, 172-176.

Gandhi, R., Luk, K.C., Rymar, V.V., and Sadikot, A.F. (2008). Group I mGluR5 metabotropic glutamate receptors regulate proliferation of neuronal progenitors in specific forebrain developmental domains. J Neurochem *104*, 155-172.

Gauthier, L.R., Charrin, B.C., Borrell-Pages, M., Dompierre, J.P., Rangone, H., Cordelieres, F.P., De Mey, J., MacDonald, M.E., Lessmann, V., Humbert, S*., et al.* (2004). Huntingtin controls neurotrophic support and survival of neurons by enhancing BDNF vesicular transport along microtubules. Cell *118*, 127-138.

Gerfen, C., Engber, T., Mahan, L., Susel, Z., Chase, T., Monsma, F., and Sibley, D. (1990). D1 and D2 dopamine receptor-regulated gene expression of striatonigral and striatopallidal neurons. Science *250*, 1429-1432.

Gerfen, C.R. (1984). The neostriatal mosaic: compartmentalization of corticostriatal input and striatonigral output systems. Nature *311*, 461-464.

Gerfen, C.R. (1985). The neostriatal mosaic. I. compartmental organization of projections from the striatum to the substantia nigra in the rat. Journal of Comparative Neurology *236*, 454-476.

Gerfen, C.R. (1992). The neostriatal mosaic: multiple levels of compartmental organization. Trends Neurosci *15*, 133-139.

Gernert, M., Hamann, M., Bennay, M., Loscher, W., and Richter, A. (2000). Deficit of striatal parvalbumin-reactive GABAergic interneurons and decreased basal ganglia output in a genetic rodent model of idiopathic paroxysmal dystonia. J Neurosci *20*, 7052-7058.

Gharami, K., Xie, Y., An, J.J., Tonegawa, S., and Xu, B. (2008). Brain-derived neurotrophic factor over-expression in the forebrain ameliorates Huntington's disease phenotypes in mice. Journal of Neurochemistry *105*, 369-379.

Ghosh, A., Carnahan, J., and Greenberg, M. (1994). Requirement for BDNF in activity-dependent survival of cortical neurons. Science *263*, 1618-1623.

Giampa, C., DeMarch, Z., D'Angelo, V., Morello, M., Martorana, A., Sancesario, G., Bernardi, G., and Fusco, F.R. (2006). Striatal modulation of cAMP-response-element-binding protein (CREB) after excitotoxic lesions: implications with neuronal vulnerability in Huntington's disease. Eur J Neurosci *23*, 11-20.

Giampa, C., Middei, S., Patassini, S., Borreca, A., Marullo, F., Laurenti, D., Bernardi, G., Ammassari-Teule, M., and Fusco, F.R. (2009). Phosphodiesterase type IV inhibition prevents sequestration of CREB binding protein, protects striatal parvalbumin interneurons and rescues motor deficits in the R6/2 mouse model of Huntington's disease. Eur J Neurosci *29*, 902-910.

Giampà, C., Montagna, E., Dato, C., Melone, M.A.B., Bernardi, G., and Fusco, F.R. (2013). Systemic Delivery of Recombinant Brain Derived Neurotrophic Factor (BDNF) in the R6/2 Mouse Model of Huntington's Disease. PLoS One *8*, e64037.

Gittis, A.H., Nelson, A.B., Thwin, M.T., Palop, J.J., and Kreitzer, A.C. (2010). Distinct roles of GABAergic interneurons in the regulation of striatal output pathways. J Neurosci *30*, 2223-2234.

Gladding, C.M., Sepers, M.D., Xu, J., Zhang, L.Y., Milnerwood, A.J., Lombroso, P.J., and Raymond, L.A. (2012). Calpain and STriatal-Enriched protein tyrosine phosphatase (STEP) activation contribute to extrasynaptic NMDA receptor localization in a Huntington's disease mouse model. Hum Mol Genet *21*, 3739-3752.

GOEBEL, H.H., HEIPERTZ, R., SCHOLZ, W., IQBAL, K., and TELLEZ-NAGEL, I. (1978). Juvenile Huntington chorea. Clinical, ultrastructural, and biochemical studies *28*, 23-23.

Goldberg, J., and Wilson, C.J. (2017). The Cholinergic Interneuron of the Striatum. In, pp. 137-155.

Goldstein, L.S.B. (2012). Axonal transport and neurodegenerative disease: Can we see the elephant? Progress in neurobiology *99*, 10.1016/j.pneurobio.2012.1003.1006.

Gomez-Tortosa, E., MacDonald, M.E., Friend, J.C., Taylor, S.A., Weiler, L.J., Cupples, L.A., Srinidhi, J., Gusella, J.F., Bird, E.D., Vonsattel, J.P.*, et al.* (2001). Quantitative neuropathological changes in presymptomatic Huntington's disease. Ann Neurol *49*, 29-34.

Gonzales, K.K., and Smith, Y. (2015). Cholinergic interneurons in the dorsal and ventral striatum: anatomical and functional considerations in normal and diseased conditions. Ann N Y Acad Sci *1349*, 1-45.

Goodchild, R.E., Kim, C.E., and Dauer, W.T. Loss of the Dystonia-Associated Protein TorsinA Selectively Disrupts the Neuronal Nuclear Envelope. Neuron *48*, 923-932.

Goto, S., Lee, L.V., Munoz, E.L., Tooyama, I., Tamiya, G., Makino, S., Ando, S., Dantes, M.B., Yamada, K., Matsumoto, S.*, et al.* (2005). Functional anatomy of the basal ganglia in X-linked recessive dystonia-parkinsonism. Ann Neurol *58*, 7-17.

Graham, R.K., Slow, E.J., Deng, Y., Bissada, N., Lu, G., Pearson, J., Shehadeh, J., Leavitt, B.R., Raymond, L.A., and Hayden, M.R. (2006). Levels of mutant huntingtin influence the phenotypic severity of Huntington disease in YAC128 mouse models. Neurobiol Dis *21*, 444-455.

Granata, A., and Warner, T.T. (2010). The role of torsinA in dystonia. Eur J Neurol *17 Suppl 1*, 81-87.

Gray, M., Shirasaki, D.I., Cepeda, C., Andre, V.M., Wilburn, B., Lu, X.H., Tao, J., Yamazaki, I., Li, S.H., Sun, Y.E.*, et al.* (2008). Full-length human mutant huntingtin with a stable polyglutamine repeat can elicit progressive and selective neuropathogenesis in BACHD mice. J Neurosci *28*, 6182-6195.

Graybiel, A.M., Baughman, R.W., and Eckenstein, F. (1986). Cholinergic neuropil of the striatum observes striosomal boundaries. Nature *323*, 625-627.

Groenewegen, H.J., and Berendse, H.W. (1994). The specificity of the 'nonspecific' midline and intralaminar thalamic nuclei. Trends Neurosci *17*, 52-57.

Gundersen, H.J., Jensen, E.B., Kieu, K., and Nielsen, J. (1999). The efficiency of systematic sampling in stereology--reconsidered. J Microsc *193*, 199-211.

Guo, Q., Wang, D., He, X., Feng, Q., Lin, R., Xu, F., Fu, L., and Luo, M. (2015). Whole-Brain Mapping of Inputs to Projection Neurons and Cholinergic Interneurons in the Dorsal Striatum. PLoS One *10*, e0123381.

Hallett, M. (2011). Neurophysiology of dystonia: The role of inhibition. Neurobiol Dis *42*, 177-184.

Halliday, A.L., and Cepko, C.L. (1992). Generation and migration of cells in the developing striatum. Neuron *9*, 15-26.

Halliday, G.M. (2009). Thalamic changes in Parkinson's disease. Parkinsonism Relat Disord *15 Suppl 3*, S152-155.

Halliday, G.M., Li, Y.W., Blumbergs, P.C., Joh, T.H., Cotton, R.G.H., Howe, P.R.C., Blessing, W.W., and Geffen, L.B. (1990). Neuropathology of immunohistochemically identified brainstem neurons in Parkinson's disease. Annals of Neurology *27*, 373-385.

Hammond, E.N.L., Tetzlaff, W., Mestres, P., and Giehl, K.M. (1999). BDNF, but not NT-3, promotes long-term survival of axotomized adult rat corticospinal neurons in vivo. Neuroreport *10*, 2671-2675.

Hardingham, G.E. (2009). Coupling of the NMDA receptor to neuroprotective and neurodestructive events. Biochemical Society transactions *37*, 1147-1160.

Hardingham, G.E., and Bading, H. (2010). Synaptic versus extrasynaptic NMDA receptor signalling: implications for neurodegenerative disorders. Nat Rev Neurosci *11*, 682-696.

Harrington, K., and Kowall, N. (1991). Parvalbumin immunoreactive neurons resist degeneration in Huntingtons-disease striatum. Paper presented at: Journal of Neuropathology and Experimental Neurology (AMER ASSN NEUROPATHOLOGISTS INC 1041 NEW HAMPSHIRE ST, LAWRENCE, KS 66044).

Hassani, O.K., Rymar, V.V., Nguyen, K.Q., Huo, L., Cloutier, J.-F., Miller, F.D., and Sadikot, A.F. (2020). The noradrenergic system is necessary for survival of vulnerable midbrain dopaminergic neurons: implications for development and Parkinson's disease. Neurobiol Aging *85*, 22-37.

Hedreen, J.C., and Folstein, S.E. (1995). Early loss of neostriatal striosome neurons in Huntington's disease. J Neuropathol Exp Neurol *54*, 105-120.

Hedreen, J.C., Zweig, R.M., DeLong, M.R., Whitehouse, P.J., and Price, D. (1988). Primary dystonias: a review of the pathology and suggestions for new directions of study. Advances in neurology *50*, 123.

Heinsen, H., Rub, U., Gangnus, D., Jungkunz, G., Bauer, M., Ulmar, G., Bethke, B., Schuler, M., Bocker, F., Eisenmenger, W.*, et al.* (1996). Nerve cell loss in the thalamic centromedian-parafascicular complex in patients with Huntington's disease. Acta Neuropathol *91*, 161-168.

Hendrix, C.M., and Vitek, J.L. (2012). Toward a network model of dystonia. Ann N Y Acad Sci *1265*, 46-55.

Hendry, S.H.C., Jones, E.G., and Graham, J. (1979). Thalamic relay nuclei for cerebellar and certain related fiber systems in the cat. Journal of Comparative Neurology *185*, 679-713.

Herkenham, M., and Pert, C.B. (1981). Mosaic distribution of opiate receptors, parafascicular projections and acetylcholinesterase in rat striatum. Nature *291*, 415-418.

Herzfeld, T., Nolte, D., Grznarova, M., Hofmann, A., Schultze, J.L., and Müller, U. (2013). X-linked dystonia parkinsonism syndrome (XDP, lubag): disease-specific sequence change DSC3 in TAF1/DYT3 affects genes in vesicular transport and dopamine metabolism. Hum Mol Genet *22*, 941-951.

Hess, E.J., and Jinnah, H.A. (2015). Chapter 27 - Mouse Models of Dystonia A2 - LeDoux, Mark S. In Movement Disorders (Second Edition) (Boston: Academic Press), pp. 465-481.

Hetman, M., and Kharebava, G. (2006). Survival signaling pathways activated by NMDA receptors. Curr Top Med Chem *6*, 787-799.

Hewett, J., Johansen, P., Sharma, N., Standaert, D., and Balcioglu, A. (2010). Function of dopamine transporter is compromised in DYT1 transgenic animal model in vivo. J Neurochem *113*, 228-235.

Hewett, J.W., Tannous, B., Niland, B.P., Nery, F.C., Zeng, J., Li, Y., and Breakefield, X.O. (2007). Mutant torsinA interferes with protein processing through the secretory pathway in DYT1 dystonia cells. Proceedings of the National Academy of Sciences *104*, 7271-7276.

Hickey, M.A., Gallant, K., Gross, G.G., Levine, M.S., and Chesselet, M.F. (2005). Early behavioral deficits in R6/2 mice suitable for use in preclinical drug testing. Neurobiol Dis *20*, 1-11.

Hickey, M.A., Kosmalska, A., Enayati, J., Cohen, R., Zeitlin, S., Levine, M.S., and Chesselet, M.F. (2008). Extensive early motor and non-motor behavioral deficits are followed by striatal neuronal loss in knock-in Huntington's disease mice. Neuroscience *157*, 280-295.

Hintzen, A., Pelzer, E.A., and Tittgemeyer, M. (2018). Thalamic interactions of cerebellum and basal ganglia. Brain Structure and Function *223*, 569-587.

Hirsch, E.C., Graybiel, A.M., Duyckaerts, C., and Javoy-Agid, F. (1987). Neuronal loss in the pedunculopontine tegmental nucleus in Parkinson disease and in progressive supranuclear palsy. Proceedings of the National Academy of Sciences *84*, 5976-5980.

Hockly, E., Cordery, P.M., Woodman, B., Mahal, A., van Dellen, A., Blakemore, C., Lewis, C.M., Hannan, A.J., and Bates, G.P. (2002). Environmental enrichment slows disease progression in R6/2 Huntington's disease mice. Ann Neurol *51*, 235-242.

Hodgson, J.G., Agopyan, N., Gutekunst, C.-A., Leavitt, B.R., LePiane, F., Singaraja, R., Smith, D.J., Bissada, N., McCutcheon, K., and Nasir, J. (1999). A YAC mouse model for Huntington's disease with full-length mutant huntingtin, cytoplasmic toxicity, and selective striatal neurodegeneration. Neuron *23*, 181-192.

Holley, S.M., Joshi, P.R., Parievsky, A., Galvan, L., Chen, J.Y., Fisher, Y.E., Huynh, M.N., Cepeda, C., and Levine, M.S. (2015). Enhanced GABAergic Inputs Contribute to Functional Alterations of Cholinergic Interneurons in the R6/2 Mouse Model of Huntington's Disease. eNeuro *2*.

Holton, J.L., Schneider, S.A., Ganesharajah, T., Gandhi, S., Strand, C., Shashidharan, P., Barreto, J., Wood, N.W., Lees, A.J., Bhatia, K.P.*, et al.* (2008). Neuropathology of primary adult-onset dystonia. Neurology *70*, 695-699.

Hoshi, E., Tremblay, L., Féger, J., Carras, P.L., and Strick, P.L. (2005). The cerebellum communicates with the basal ganglia. Nature neuroscience *8*, 1491.

Huerta-Ocampo, I., Mena-Segovia, J., and Bolam, J.P. (2014). Convergence of cortical and thalamic input to direct and indirect pathway medium spiny neurons in the striatum. Brain Structure and Function *219*, 1787-1800.

Hutchinson, M., Nakamura, T., Moeller, J.R., Antonini, A., Belakhlef, A., Dhawan, V., and Eidelberg, D. (2000). The metabolic topography of essential blepharospasm: a focal dystonia with general implications. Neurology *55*, 673-677.

Hwang, W.Y., Fu, Y., Reyon, D., Maeder, M.L., Tsai, S.Q., Sander, J.D., Peterson, R.T., Yeh, J.J., and Joung, J.K. (2013). Efficient genome editing in zebrafish using a CRISPR-Cas system. Nature biotechnology *31*, 227.

Ichinose, H., Ohye, T., Takahashi, E.-i., Seki, N., Hori, T.-a., Segawa, M., Nomura, Y., Endo, K., Tanaka, H., Tsuji, S.*, et al.* (1994). Hereditary progressive dystonia with marked diurnal fluctuation caused by mutations in the GTP cyclohydrolase I gene. Nat Genet *8*, 236.

Ikonomidou, C., Stefovska, V., and Turski, L. (2000). Neuronal death enhanced by N-methyl-D-aspartate antagonists. Proc Natl Acad Sci U S A *97*, 12885-12890.

Jackson, G.R., Salecker, I., Dong, X., Yao, X., Arnheim, N., Faber, P.W., MacDonald, M.E., and Zipursky, S.L. (1998). Polyglutamine-Expanded Human Huntingtin Transgenes Induce Degeneration of Drosophila Photoreceptor Neurons. Neuron *21*, 633-642.

Jansson, L.C., and Akerman, K.E. (2014). The role of glutamate and its receptors in the proliferation, migration, differentiation and survival of neural progenitor cells. J Neural Transm (Vienna) *121*, 819-836.

Jardim, L.B., Pereira, M.L., Silveira, I., Ferro, A., Sequeiros, J., and Giugliani, R. (2001). Neurologic findings in Machado-Joseph disease: relation with disease duration, subtypes, and (CAG)n. Arch Neurol *58*, 899-904.

Jimenez-Castellanos, J., and Graybiel, A.M. (1987). Subdivisions of the dopamine-containing A8-A9-A10 complex identified by their differential mesostriatal innervation of striosomes and extrastriosomal matrix. Neuroscience *23*, 223-242.

Jimenez-Castellanos, J., and Graybiel, A.M. (1989). Compartmental origins of striatal efferent projections in the cat. Neuroscience *32*, 297-321.

Jinnah, H.A., Neychev, V., and Hess, E.J. (2017). The Anatomical Basis for Dystonia: The Motor Network Model. Tremor Other Hyperkinet Mov (N Y) *7*, 506.

Johnston, J.G., Gerfen, C.R., Haber, S.N., and van der Kooy, D. (1990). Mechanisms of striatal pattern formation: conservation of mammalian compartmentalization. Brain Res Dev Brain Res *57*, 93-102.

Jung, A.B., and Bennett, J.P. (1996). Development of striatal dopaminergic function. III: Pre- and postnatal development of striatal and cortical mRNAs for the neurotrophin receptors trkBTK+ and trkC and their regulation by synaptic dopamine. Developmental Brain Research *94*, 133-143.

Kachidian, P., Vuillet, J., Nieoullon, A., Lafaille, G., and Kerkerian-Le Goff, L. (1996). Striatal neuropeptide Y neurones are not a target for thalamic afferent fibres. Neuroreport *7*, 1665-1669.

Kanovsky, P., Bhatia, K.P., and Rosales, R.L. (2015). Dystonia and Dystonic Syndromes (Springer).

Kassubek, J., Juengling, F.D., Ecker, D., and Landwehrmeyer, G.B. (2005). Thalamic atrophy in Huntington's disease co-varies with cognitive performance: a morphometric MRI analysis. Cerebral cortex *15*, 846-853.

Kassubek, J., Juengling, F.D., Kioschies, T., Henkel, K., Karitzky, J., Kramer, B., Ecker, D., Andrich, J., Saft, C., Kraus, P.*, et al.* (2004). Topography of cerebral atrophy in early Huntington's disease: a voxel based morphometric MRI study. J Neurol Neurosurg Psychiatry *75*, 213-220.

Kaufman, A.M., Milnerwood, A.J., Sepers, M.D., Coquinco, A., She, K., Wang, L., Lee, H., Craig, A.M., Cynader, M., and Raymond, L.A. (2012). Opposing Roles of Synaptic and Extrasynaptic NMDA Receptor Signaling in Cocultured Striatal and Cortical Neurons. The Journal of Neuroscience *32*, 3992.

Kawaguchi, Y. (1992). Large aspiny cells in the matrix of the rat neostriatum in vitro: physiological identification, relation to the compartments and excitatory postsynaptic currents. Journal of neurophysiology *67*, 1669-1682.

Kawaguchi, Y. (1993). Physiological, morphological, and histochemical characterization of three classes of interneurons in rat neostriatum. J Neurosci *13*, 4908-4923.

Kawaguchi, Y., Wilson, C.J., and Emson, P.C. (1989). Intracellular recording of identified neostriatal patch and matrix spiny cells in a slice preparation preserving cortical inputs. Journal of neurophysiology *62*, 1052-1068.

Kawaguchi, Y., Wilson, C.J., and Emson, P.C. (1990). Projection subtypes of rat neostriatal matrix cells revealed by intracellular injection of biocytin. J Neurosci *10*, 3421-3438.

Kells, A., Henry, R., and Connor, B. (2008). AAV–BDNF mediated attenuation of quinolinic acid-induced neuropathology and motor function impairment. Gene therapy *15*, 966.

Kemp, J.M., and Powell, T.P. (1971a). The synaptic organization of the caudate nucleus. Philos Trans R Soc Lond B Biol Sci *262*, 403-412.

Kemp, J.M., and Powell, T.P. (1971b). The termination of fibres from the cerebral cortex and thalamus upon dendritic spines in the caudate nucleus: a study with the Golgi method. Philos Trans R Soc Lond B Biol Sci *262*, 429-439.

Kemp, J.M., and Powell, T.P.S. (1971c). The Connexions of the Striatum and Globus Pallidus: Synthesis and Speculation. Philosophical Transactions of the Royal Society of London Series B, Biological Sciences *262*, 441-457.

Kerkerian-Le Goff, L., Bacci, J.-J., Jouve, L., Melon, C., and Salin, P. (2009). Impact of surgery targeting the caudal intralaminar thalamic nuclei on the pathophysiological functioning of basal ganglia in a rat model of Parkinson's disease. Brain Res Bull *78*, 80-84.

Kim, D., Jeon, J., Cheong, E., Kim, D.J., Ryu, H., Seo, H., and Kim, Y.K. (2015). Neuroanatomical Visualization of the Impaired Striatal Connectivity in Huntington's Disease Mouse Model. Mol Neurobiol.

Kimura, M., Minamimoto, T., Matsumoto, N., and Hori, Y. (2004). Monitoring and switching of cortico-basal ganglia loop functions by the thalamo-striatal system. Neuroscience research *48*, 355-360.

Kincaid, A.E., and Wilson, C.J. (1996). Corticostriatal innervation of the patch and matrix in the rat neostriatum. J Comp Neurol *374*, 578-592.

Kirschenbaum, B., and Goldman, S.A. (1995). Brain-derived neurotrophic factor promotes the survival of neurons arising from the adult rat forebrain subependymal zone. Proceedings of the National Academy of Sciences *92*, 210-214.

Klein, C., and Fahn, S. (2013). Translation of Oppenheim's 1911 paper on dystonia. Movement Disorders *28*, 851-862.

Klein, C., Liu, L., Doheny, D., Kock, N., Müller, B., Aguiar, P.D.C., Leung, J., Leon, D.D., Bressman, S.B., Silverman, J.*, et al.* (2002). ε-sarcoglycan mutations found in combination with other dystonia gene mutations. Ann Neurol *52*, 675-679.

Koch, G., Porcacchia, P., Ponzo, V., Carrillo, F., Caceres-Redondo, M.T., Brusa, L., Desiato, M.T., Arciprete, F., Di Lorenzo, F., Pisani, A.*, et al.* (2014). Effects of two weeks of cerebellar theta burst stimulation in cervical dystonia patients. Brain stimulation *7*, 564-572.

Kolb, B., Gibb, R., and van der Kooy, D. (1992). Cortical and striatal structure and connectivity are altered by neonatal hemidecortication in rats. J Comp Neurol *322*, 311-324.

Kolodziejczyk, K., and Raymond, L.A. (2016). Differential changes in thalamic and cortical excitatory synapses onto striatal spiny projection neurons in a Huntington disease mouse model. Neurobiol Dis *86*, 62-74.

Konakova, M., Huynh, D.P., Yong, W., and Pulst, S.M. (2001). Cellular distribution of torsin a and torsin b in normal human brain. Arch Neurol *58*, 921-927.

Kononenko, N.L., Claßen, G.A., Kuijpers, M., Puchkov, D., Maritzen, T., Tempes, A., Malik, A.R., Skalecka, A., Bera, S., Jaworski, J.*, et al.* (2017). Retrograde transport of TrkB-containing autophagosomes via the

adaptor AP-2 mediates neuronal complexity and prevents neurodegeneration. Nat Commun *8*, 14819-14819.

Koos, T., and Tepper, J.M. (2002). Dual cholinergic control of fast-spiking interneurons in the neostriatum. J Neurosci *22*, 529-535.

Krammer, E.B. (1980). Anterograde and transsynaptic degeneration 'en cascade' in basal ganglia induced by intrastriatal injection of kainic acid: an animal analogue of Huntington's disease. Brain Res *196*, 209-221.

Krushel, L.A., Johnston, J.G., Fishell, G., Tibshirani, R., and van der Kooy, D. (1993). Spatially localized neuronal cell lineages in the developing mammalian forebrain. Neuroscience *53*, 1035-1047.

Kuhar, M.J., Roth, R.H., and Aghajanian, G.K. (1971). Selective reduction of tryptophan hydroxylase activity in rat forebrain after midbrain raphe lesions. Brain Res *35*, 167-176.

Kuppenbender, K.D., Standaert, D.G., Feuerstein, T.J., Penney, J.B., Jr., Young, A.B., and Landwehrmeyer, G.B. (2000). Expression of NMDA receptor subunit mRNAs in neurochemically identified projection and interneurons in the human striatum. J Comp Neurol *419*, 407-421.

Kusakabe, M., Mangiarini, L., Laywell, E.D., Bates, G.P., Yoshiki, A., Hiraiwa, N., Inoue, J., and Steindler, D.A. (2001). Loss of cortical and thalamic neuronal tenascin-C expression in a transgenic mouse expressing exon 1 of the human Huntington disease gene. J Comp Neurol *430*, 485-500.

Lacey, C.J., Bolam, J.P., and Magill, P.J. (2007). Novel and distinct operational principles of intralaminar thalamic neurons and their striatal projections. J Neurosci *27*, 4374-4384.

Lalonde, R., and Strazielle, C. (2007). Spontaneous and induced mouse mutations with cerebellar dysfunctions: behavior and neurochemistry. Brain Res *1140*, 51-74.

Lalonde, R., and Strazielle, C. (2011). Brain regions and genes affecting limb-clasping responses. Brain Res Rev *67*, 252-259.

Lanciego, J.L., Rodríguez-Oroz, M.C., Blesa, F.J., Alvarez-Erviti, L., Guridi, J., Barroso-Chinea, P., Smith, Y., and Obeso, J.A. (2008). Lesion of the centromedian thalamic nucleus in MPTP-treated monkeys. Movement Disorders *23*, 708-715.

Landwehrmeyer, G., Standaert, D., Testa, C., Penney, J., and Young, A. (1995). NMDA receptor subunit mRNA expression by projection neurons and interneurons in rat striatum. The Journal of Neuroscience *15*, 5297-5307.

Lange, N., Hamann, M., Shashidharan, P., and Richter, A. (2011). Behavioural and pharmacological examinations in a transgenic mouse model of early-onset torsion dystonia. Pharmacology Biochemistry and Behavior *97*, 647-655.

Lapper, S.R., and Bolam, J.P. (1992). Input from the frontal cortex and the parafascicular nucleus to cholinergic interneurons in the dorsal striatum of the rat. Neuroscience *51*, 533-545.

Lapper, S.R., Smith, Y., Sadikot, A.F., Parent, A., and Bolam, J.P. (1992). Cortical input to parvalbumin-immunoreactive neurones in the putamen of the squirrel monkey. Brain Res *580*, 215-224.

Larsson, E., Nanobashvili, A., Kokaia, Z., and Lindvall, O. (1999). Evidence for neuroprotective effects of endogenous brain-derived neurotrophic factor after global forebrain ischemia in rats. J Cereb Blood Flow Metab *19*, 1220-1228.

Lawhorn, C., Smith, D.M., and Brown, L.L. (2008). Striosome-matrix pathology and motor deficits in the YAC128 mouse model of Huntington's disease. Neurobiol Dis *32*, 471-478.

LeDoux, M.S., and Brady, K.A. (2003). Secondary cervical dystonia associated with structural lesions of the central nervous system. Mov Disord *18*, 60-69.

Lee, H.Y., Xu, Y., Huang, Y., Ahn, A.H., Auburger, G.W., Pandolfo, M., Kwiecinski, H., Grimes, D.A., Lang, A.E., Nielsen, J.E.*, et al.* (2004). The gene for paroxysmal non-kinesigenic dyskinesia encodes an enzyme in a stress response pathway. Hum Mol Genet *13*, 3161-3170.

Lee, M.S., and Marsden, C.D. (1994). Movement disorders following lesions of the thalamus or subthalamic region. Mov Disord *9*, 493-507.

Lehericy, S., Tijssen, M.A., Vidailhet, M., Kaji, R., and Meunier, S. (2013). The anatomical basis of dystonia: current view using neuroimaging. Mov Disord *28*, 944-957.

Lei, W., Deng, Y., Liu, B., Mu, S., Guley, N.M., Wong, T., and Reiner, A. (2013). Confocal laser scanning microscopy and ultrastructural study of VGLUT2 thalamic input to striatal projection neurons in rats. Journal of Comparative Neurology *521*, 1354-1377.

Lein, E.S., Hawrylycz, M.J., Ao, N., Ayres, M., Bensinger, A., Bernard, A., Boe, A.F., Boguski, M.S., Brockway, K.S., and Byrnes, E.J. (2007). Genome-wide atlas of gene expression in the adult mouse brain. Nature *445*, 168.

Lenz, F.A., Suarez, J.I., Metman, L.V., Reich, S.G., Karp, B.I., Hallett, M., Rowland, L.H., and Dougherty, P.M. (1998). Pallidal activity during dystonia: somatosensory reorganisation and changes with severity. Journal of Neurology, Neurosurgery & Psychiatry *65*, 767-770.

Leveille, F., Papadia, S., Fricker, M., Bell, K.F., Soriano, F.X., Martel, M.A., Puddifoot, C., Habel, M., Wyllie, D.J., Ikonomidou, C.*, et al.* (2010). Suppression of the intrinsic apoptosis pathway by synaptic activity. J Neurosci *30*, 2623-2635.

Levesque, M., and Parent, A. (2005). The striatofugal fiber system in primates: a reevaluation of its organization based on single-axon tracing studies. Proc Natl Acad Sci U S A *102*, 11888-11893.

Li, L., Fan, M., Icton, C.D., Chen, N., Leavitt, B.R., Hayden, M.R., Murphy, T.H., and Raymond, L.A. (2003). Role of NR2B-type NMDA receptors in selective neurodegeneration in Huntington disease. Neurobiol Aging *24*, 1113-1121.

Linden, R. (1994). The survival of developing neurons: a review of afferent control. Neuroscience *58*, 671-682.

Liot, G., Zala, D., Pla, P., Mottet, G., Piel, M., and Saudou, F. (2013). Mutant Huntingtin Alters Retrograde Transport of TrkB Receptors in Striatal Dendrites. The Journal of Neuroscience *33*, 6298.

Loopuijt, L., Villablanca, J., Hovda, D., Huang, E., and Mancuso, S. (1997a). The effect of neocortical lesions on the number of cells in neonatal or adult feline caudate nucleus: comparison to fetal lesions. Neuroscience *77*, 403-418.

Loopuijt, L.D., Hovda, D.A., Ebrahim, A., Villablanca, J.R., and Chugani, H.T. (1998). Differences in D2 dopamine receptor binding in the neostriatum between cats hemidecorticated neonatally or in adulthood. Developmental brain research *107*, 113-122.

Loopuijt, L.D., Villablanca, J.R., Hovda, D.A., Huang, E., and Mancuso, S. (1997b). The effect of neocortical lesions on the number of cells in neonatal or adult feline caudate nucleus: comparison to fetal lesions. Neuroscience *77*, 403-418.

Lopez-Huerta, V.G., Nakano, Y., Bausenwein, J., Jaidar, O., Lazarus, M., Cherassse, Y., Garcia-Munoz, M., and Arbuthnott, G. (2015). The neostriatum: two entities, one structure? Brain structure & function.

LoTurco, J.J., Owens, D.F., Heath, M.J., Davis, M.B., and Kriegstein, A.R. (1995). GABA and glutamate depolarize cortical progenitor cells and inhibit DNA synthesis. Neuron *15*, 1287-1298.

Louis, E.D., Anderson, K.E., Moskowitz, C., Thorne, D.Z., and Marder, K. (2000). Dystonia-predominant adult-onset Huntington disease: association between motor phenotype and age of onset in adults. Arch Neurol *57*, 1326-1330.

Louis, E.D., Lee, P., Quinn, L., and Marder, K. (1999). Dystonia in Huntington's disease: prevalence and clinical characteristics. Mov Disord *14*, 95-101.

Lu, P., Blesch, A., and Tuszynski, M.H. (2001). Neurotrophism without neurotropism: BDNF promotes survival but not growth of lesioned corticospinal neurons. Journal of Comparative Neurology *436*, 456-470.

Lucidi-Phillipi, C.A., Gage, F.H., Shults, C.W., Jones, K.R., Reichardt, L.F., and Kang, U.J. (1995). Brain-derived neurotrophic factor-transduced fibroblasts: production of BDNF and effects of grafting to the adult rat brain. J Comp Neurol *354*, 361-376.

Luk, K.C., Kennedy, T.E., and Sadikot, A.F. (2003). Glutamate promotes proliferation of striatal neuronal progenitors by an NMDA receptor-mediated mechanism. J Neurosci *23*, 2239-2250.

Luk, K.C., and Sadikot, A.F. (2001). GABA promotes survival but not proliferation of parvalbumin-immunoreactive interneurons in rodent neostriatum: an in vivo study with stereology. Neuroscience *104*, 93-103.

Luk, K.C., and Sadikot, A.F. (2004). Glutamate and regulation of proliferation in the developing mammalian telencephalon. Dev Neurosci *26*, 218-228.

Madeo, G., and Pisani, A. (2016). Pathophysiology of dystonia. In Handbook of Behavioral Neuroscience (Elsevier), pp. 929-950.

Maisonpierre, P.C., Belluscio, L., Friedman, B., Alderson, R.F., Wiegand, S.J., Furth, M.E., Lindsay, R.M., and Yancopoulos, G.D. (1990). NT-3, BDNF, and NGF in the developing rat nervous system: Parallel as well as reciprocal patterns of expression. Neuron *5*, 501-509.

Malach, R., and Graybiel, A.M. (1986). Mosaic architecture of the somatic sensory-recipient sector of the cat's striatum. J Neurosci *6*, 3436-3458.

Malkovska, I., Kernie, S.G., and Parada, L.F. (2006). Differential expression of the four untranslated BDNF exons in the adult mouse brain. Journal of neuroscience research *83*, 211-221.

Mamounas, L., Blue, M., Siuciak, J., and Altar, C. (1995). Brain-derived neurotrophic factor promotes the survival and sprouting of serotonergic axons in rat brain. The Journal of Neuroscience *15*, 7929-7939.

Mangiarini, L., Sathasivam, K., Seller, M., Cozens, B., Harper, A., Hetherington, C., Lawton, M., Trottier, Y., Lehrach, H., Davies, S.W.*, et al.* (1996). Exon 1 of the HD gene with an expanded CAG repeat is sufficient to cause a progressive neurological phenotype in transgenic mice. Cell *87*, 493-506.

Mao, L., Yang, L., Tang, Q., Samdani, S., Zhang, G., and Wang, J.Q. (2005). The Scaffold Protein Homer1b/c Links Metabotropic Glutamate Receptor 5 to Extracellular Signal-Regulated Protein Kinase Cascades in Neurons. The Journal of Neuroscience *25*, 2741-2752.

Marálek, M. (2000). Tardive Drug-Induced Extrapyramidal Syndromes. Pharmacopsychiatry *33*, 14-33.

Marín, O., Anderson, S.A., and Rubenstein, J.L. (2000). Origin and molecular specification of striatal interneurons. Journal of Neuroscience *20*, 6063-6076.

Marsden, C. (1976). Dystonia: the spectrum of the disease. Research publications-Association for Research in Nervous and Mental Disease *55*, 351-367.

Marsden, C.D., Obeso, J.A., Zarranz, J.J., and Lang, A.E. (1985). THE ANATOMICAL BASIS OF SYMPTOMATIC HEMIDYSTONIA. Brain *108*, 463-483.

Martella, G., Maltese, M., Nisticò, R., Schirinzi, T., Madeo, G., Sciamanna, G., Ponterio, G., Tassone, A., Mandolesi, G., Vanni, V.*, et al.* (2014). Regional specificity of synaptic plasticity deficits in a knock-in mouse model of DYT1 dystonia. Neurobiol Dis *65*, 124-132.

Martinez-Serrano, A., Hantzopoulos, P.A., and Bjorklund, A. (1996). Ex vivo gene transfer of brain-derived neurotrophic factor to the intact rat forebrain: neurotrophic effects on cholinergic neurons. Eur J Neurosci *8*, 727-735.

Massouh, M., Wallman, M.J., Pourcher, E., and Parent, A. (2008). The fate of the large striatal interneurons expressing calretinin in Huntington's disease. Neuroscience research *62*, 216-224.

Matsui, J.T., Vaidya, J.G., Johnson, H.J., Magnotta, V.A., Long, J.D., Mills, J.A., Lowe, M.J., Sakaie, K.E., Rao, S.M., Smith, M.M.*, et al.* (2014). Diffusion weighted imaging of prefrontal cortex in prodromal Huntington's disease. Hum Brain Mapp *35*, 1562-1573.

Matsumoto, N., Minamimoto, T., Graybiel, A.M., and Kimura, M. (2001). Neurons in the thalamic CM-Pf complex supply striatal neurons with information about behaviorally significant sensory events. Journal of neurophysiology *85*, 960-976.

McFarland, N.R., and Haber, S.N. (2000). Convergent inputs from thalamic motor nuclei and frontal cortical areas to the dorsal striatum in the primate. J Neurosci *20*, 3798-3813.

McFarland, N.R., and Haber, S.N. (2001). Organization of thalamostriatal terminals from the ventral motor nuclei in the macaque. J Comp Neurol *429*, 321-336.

McGeorge, A., and Faull, R. (1987). The organization and collateralization of corticostriate neurones in the motor and sensory cortex of the rat brain. Brain Res *423*, 318-324.

McGeorge, A., and Faull, R. (1989). The organization of the projection from the cerebral cortex to the striatum in the rat. Neuroscience *29*, 503-537.

McGregor, M.M., McKinsey, G.L., Girasole, A.E., Bair-Marshall, C.J., Rubenstein, J.L.R., and Nelson, A.B. (2019). Functionally Distinct Connectivity of Developmentally Targeted Striosome Neurons. Cell Reports *29*, 1419-1428.e1415.

Meade, C.A., Deng, Y.P., Fusco, F.R., Del Mar, N., Hersch, S., Goldowitz, D., and Reiner, A. (2002). Cellular localization and development of neuronal intranuclear inclusions in striatal and cortical neurons in R6/2 transgenic mice. J Comp Neurol *449*, 241-269.

Meissner, I., Sapir, S., Kokmen, E., and Stein, S.D. (1987). The paramedian diencephalic syndrome: a dynamic phenomenon. Stroke *18*, 380-385.

Melik-Musyan, A.B., and Fanardjyan, V.V. (1998). Projections of the central cerebellar nuclei to the intralaminar thalamic nuclei in cats. Neurophysiology *30*, 39-47.

Menalled, L., El-Khodor, B.F., Patry, M., Suarez-Farinas, M., Orenstein, S.J., Zahasky, B., Leahy, C., Wheeler, V., Yang, X.W., MacDonald, M.*, et al.* (2009). Systematic behavioral evaluation of Huntington's disease transgenic and knock-in mouse models. Neurobiol Dis *35*, 319-336.

Menalled, L.B. (2005). Knock-in mouse models of Huntington's disease. NeuroRx *2*, 465-470.

Menalled, L.B., Sison, J.D., Wu, Y., Olivieri, M., Li, X.-J., Li, H., Zeitlin, S., and Chesselet, M.-F. (2002a). Early motor dysfunction and striosomal distribution of huntingtin microaggregates in Huntington's disease knock-in mice. Journal of Neuroscience *22*, 8266-8276.

Menalled, L.B., Sison, J.D., Wu, Y., Olivieri, M., Li, X.J., Li, H., Zeitlin, S., and Chesselet, M.F. (2002b). Early motor dysfunction and striosomal distribution of huntingtin microaggregates in Huntington's disease knock-in mice. J Neurosci *22*, 8266-8276.

Messer, A., and Strominger, N.L. (1980). An allele of the mouse mutant dystonia musculorum exhibits lesions in red nucleus and striatum. Neuroscience *5*, 543-549.

Mesulam, M., Shaw, P., Mash, D., and Weintraub, S. (2004). Cholinergic nucleus basalis tauopathy emerges early in the aging-MCI-AD continuum. Ann Neurol *55*, 815-828.

Métin, C., and Godement, P. (1996). The ganglionic eminence may be an intermediate target for corticofugal and thalamocortical axons. Journal of Neuroscience *16*, 3219-3235.

Meunier, C., Merienne, N., Jolle, C., Deglon, N., and Pellerin, L. (2016). Astrocytes are key but indirect contributors to the development of the symptomatology and pathophysiology of Huntington's disease. Glia *64*, 1841-1856.

Milnerwood, A.J., Kaufman, A.M., Sepers, M.D., Gladding, C.M., Zhang, L., Wang, L., Fan, J., Coquinco, A., Qiao, J.Y., Lee, H.*, et al.* (2012). Mitigation of augmented extrasynaptic NMDAR signaling and apoptosis in cortico-striatal co-cultures from Huntington's disease mice. Neurobiol Dis *48*, 40-51.

Milnerwood, A.J., and Raymond, L.A. (2007). Corticostriatal synaptic function in mouse models of Huntington's disease: early effects of huntingtin repeat length and protein load. The Journal of physiology *585*, 817-831.

Milnerwood, A.J., and Raymond, L.A. (2010). Early synaptic pathophysiology in neurodegeneration: insights from Huntington's disease. Trends Neurosci *33*, 513-523.

Mink, J.W. (2018). Basal ganglia mechanisms in action selection, plasticity, and dystonia. European Journal of Paediatric Neurology *22*, 225-229.

Mohan, M. (2017). OPTOGENETICS-A BRIEF REVIEW. International Journal of Pharmaceutical Sciences and Research *8*, 440.

Moller, A., Strange, P., and Gundersen, H.J. (1990). Efficient estimation of cell volume and number using the nucleator and the disector. J Microsc *159*, 61-71.

Morse, J., Wiegand, S., Anderson, K., You, Y., Cai, N., Carnahan, J., Miller, J., DiStefano, P., Altar, C., and Lindsay, R. (1993). Brain-derived neurotrophic factor (BDNF) prevents the degeneration of medial septal cholinergic neurons following fimbria transection. The Journal of Neuroscience *13*, 4146-4156.

Morton, A.J., Nicholson, L.F., and Faull, R.L. (1993). Compartmental loss of NADPH diaphorase in the neuropil of the human striatum in Huntington's disease. Neuroscience *53*, 159-168.

Moya-Alvarado, G., Gonzalez, A., Stuardo, N., and Bronfman, F.C. (2018). Brain-Derived Neurotrophic Factor (BDNF) Regulates Rab5-Positive Early Endosomes in Hippocampal Neurons to Induce Dendritic Branching. Frontiers in cellular neuroscience *12*.

Myers, R.H., Leavitt, J., Farrer, L.A., Jagadeesh, J., McFarlane, H., Mastromauro, C.A., Mark, R.J., and Gusella, J.F. (1989). Homozygote for Huntington disease. Am J Hum Genet *45*, 615-618.

Myers, R.H., Sax, D.S., Schoenfeld, M., Bird, E.D., Wolf, P.A., Vonsattel, J.P., White, R.F., and Martin, J.B. (1985). Late onset of Huntington's disease. J Neurol Neurosurg Psychiatry *48*, 530-534.

Nakamura, K.C., Fujiyama, F., Furuta, T., Hioki, H., and Kaneko, T. (2009). Afferent islands are larger than mu-opioid receptor patch in striatum of rat pups. Neuroreport *20*, 584-588.

Nakano, K., Hasegawa, Y., Tokushige, A., Nakagawa, S., Kayahara, T., and Mizuno, N. (1990). Topographical projections from the thalamus, subthalamic nucleus and pedunculopontine tegmental nucleus to the striatum in the Japanese monkey, Macaca fuscata. Brain Res *537*, 54-68.

Nakano, Y., Karube, F., Hirai, Y., Kobayashi, K., Hioki, H., Okamoto, S., Kameda, H., and Fujiyama, F. Parvalbumin-producing striatal interneurons receive excitatory inputs onto proximal dendrites from the motor thalamus in male mice. Journal of neuroscience research, n/a-n/a.

Nanda, B., Galvan, A., Smith, Y., and Wichmann, T. (2009). Effects of stimulation of the centromedian nucleus of the thalamus on the activity of striatal cells in awake rhesus monkeys. Eur J Neurosci *29*, 588-598.

Naumann, M., Pirker, W., Reiners, K., Lange, K.W., Becker, G., and Brücke, T. (1998). Imaging the pre- and postsynaptic side of striatal dopaminergic synapses in idiopathic cervical dystonia: A SPECT STUDY Using [123I] epidepride and [123I] β-CIT. Movement Disorders *13*, 319-323.

Neumann, W.J., Jha, A., Bock, A., Huebl, J., Horn, A., Schneider, G.H., Sander, T.H., Litvak, V., and Kuhn, A.A. (2015). Cortico-pallidal oscillatory connectivity in patients with dystonia. Brain.

Neychev, V.K., Gross, R.E., Lehéricy, S., Hess, E.J., and Jinnah, H.A. (2011). The functional neuroanatomy of dystonia. Neurobiol Dis *42*, 185-201.

Nguyen, K.Q., Rymar, V.V., and Sadikot, A.F. (2016). Impaired TrkB Signaling Underlies Reduced BDNF-Mediated Trophic Support of Striatal Neurons in the R6/2 Mouse Model of Huntington's Disease. Frontiers in cellular neuroscience *10*, 37.

Niethammer, M., Carbon, M., Argyelan, M., and Eidelberg, D. (2011). Hereditary Dystonia as a Neurodevelopmental Circuit Disorder: Evidence from Neuroimaging. Neurobiol Dis *42*, 202-209.

Numakawa, T., Suzuki, S., Kumamaru, E., Adachi, N., Richards, M., and Kunugi, H. (2010). BDNF function and intracellular signaling in neurons. Histol Histopathol *25*, 237-258.

O'Dwyer, J.P., O'Riordan, S., Saunders-Pullman, R., Bressman, S.B., Molloy, F., Lynch, T., and Hutchinson, M. (2005). Sensory abnormalities in unaffected relatives in familial adult-onset dystonia. Neurology *65*, 938.

Obrietan, K., Gao, X.-B., and Pol, A.N.v.d. (2002). Excitatory Actions of GABA Increase BDNF Expression via a MAPK-CREB–Dependent Mechanism—A Positive Feedback Circuit in Developing Neurons. Journal of neurophysiology *88*, 1005-1015.

Ohtani, N., Goto, T., Waeber, C., and Bhide, P.G. (2003). Dopamine modulates cell cycle in the lateral ganglionic eminence. J Neurosci *23*, 2840-2850.

Olsson, M., Björklund, A., and Campbell, K. (1998). Early specification of striatal projection neurons and interneuronal subtypes in the lateral and medial ganglionic eminence. Neuroscience *84*, 867-876.

Ostrem, J.L., and Starr, P.A. (2008). Treatment of Dystonia with Deep Brain Stimulation. Neurotherapeutics : the journal of the American Society for Experimental NeuroTherapeutics *5*, 320-330.

Ozelius, L.J., Hewett, J.W., Page, C.E., Bressman, S.B., Kramer, P.L., Shalish, C., de Leon, D., Brin, M.F., Raymond, D., Corey, D.P*., et al.* (1997). The early-onset torsion dystonia gene (DYT1) encodes an ATP-binding protein. Nat Genet *17*, 40.

Pappas, S.S., Darr, K., Holley, S.M., Cepeda, C., Mabrouk, O.S., Wong, J.M., LeWitt, T.M., Paudel, R., Houlden, H., Kennedy, R.T*., et al.* (2015). Forebrain deletion of the dystonia protein torsinA causes dystonic-like movements and loss of striatal cholinergic neurons. Elife *4*, e08352.

Parent, A. (1990). Extrinsic connections of the basal ganglia. Trends in Neurosciences *13*, 254-258.

Parent, A., and De Bellefeuille, L. (1982). Organization of efferent projections from the internal segment of globus pallidus in primate as revealed by flourescence retrograde labeling method. Brain Res *245*, 201-213.

Parent, A., Mackey, A., and De Bellefeuille, L. (1983). The subcortical afferents to caudate nucleus and putamen in primate: A fluorescence retrograde double labeling study. Neuroscience *10*, 1137-1150.

Parent, M., and Parent, A. (2016). Chapter 2 - The Primate Basal Ganglia Connectome As Revealed By Single-Axon Tracing. In Axons and Brain Architecture, K.S. Rockland, ed. (San Diego: Academic Press), pp. 27-46.

Parievsky, A., Moore, C., Kamdjou, T., Cepeda, C., Meshul, C.K., and Levine, M.S. (2017). Differential electrophysiological and morphological alterations of thalamostriatal and corticostriatal projections in the R6/2 mouse model of Huntington's disease. Neurobiol Dis *108*, 29-44.

Parr-Brownlie, L.C., Poloskey, S.L., Bergstrom, D.A., and Walters, J.R. (2009). Parafascicular thalamic nucleus activity in a rat model of Parkinson's disease. Exp Neurol *217*, 269-281.

Paulsen, J.S. (2011). Cognitive impairment in Huntington disease: diagnosis and treatment. Curr Neurol Neurosci Rep *11*, 474-483.

Pavese, N., Gerhard, A., Tai, Y.F., Ho, A.K., Turkheimer, F., Barker, R.A., Brooks, D.J., and Piccini, P. (2006). Microglial activation correlates with severity in Huntington disease: a clinical and PET study. Neurology *66*, 1638-1643.

Pearce, M.M.P., Spartz, E.J., Hong, W., Luo, L., and Kopito, R.R. (2015). Prion-like transmission of neuronal huntingtin aggregates to phagocytic glia in the Drosophila brain. Nat Commun *6*, 6768.

Pelzer, E.A., Melzer, C., Timmermann, L., von Cramon, D.Y., and Tittgemeyer, M. (2017). Basal ganglia and cerebellar interconnectivity within the human thalamus. Brain structure & function *222*, 381-392.

Penny, G.R., Wilson, C.J., and Kitai, S.T. (1988). Relationship of the axonal and dendritic geometry of spiny projection neurons to the compartmental organization of the neostriatum. J Comp Neurol *269*, 275-289.

Petrasch-Parwez, E., Nguyen, H.P., Lobbecke-Schumacher, M., Habbes, H.W., Wieczorek, S., Riess, O., Andres, K.H., Dermietzel, R., and Von Horsten, S. (2007). Cellular and subcellular localization of Huntingtin [corrected] aggregates in the brain of a rat transgenic for Huntington disease. J Comp Neurol *501*, 716-730.

Petryszyn, S., Parent, A., and Parent, M. (2017). The calretinin interneurons of the striatum: comparisons between rodents and primates under normal and pathological conditions. Journal of Neural Transmission, 1-12.

Phillips, W., Morton, A.J., and Barker, R.A. (2005). Abnormalities of neurogenesis in the R6/2 mouse model of Huntington's disease are attributable to the in vivo microenvironment. J Neurosci *25*, 11564-11576.

Phukan, J., Albanese, A., Gasser, T., and Warner, T. (2011). Primary dystonia and dystonia-plus syndromes: clinical characteristics, diagnosis, and pathogenesis. Lancet Neurol *10*, 1074-1085.

Picconi, B., Passino, E., Sgobio, C., Bonsi, P., Barone, I., Ghiglieri, V., Pisani, A., Bernardi, G., Ammassari-Teule, M., and Calabresi, P. (2006). Plastic and behavioral abnormalities in experimental Huntington's disease: a crucial role for cholinergic interneurons. Neurobiol Dis *22*, 143-152.

Pisani, A., Bernardi, G., Ding, J., and Surmeier, D.J. (2007). Re-emergence of striatal cholinergic interneurons in movement disorders. Trends in neurosciences *30*, 545-553.

Pisani, A., Bonsi, P., Centonze, D., Calabresi, P., and Bernardi, G. (2000). Activation of D2-like dopamine receptors reduces synaptic inputs to striatal cholinergic interneurons. J Neurosci *20*, RC69.

Pisani, A., Martella, G., Tscherter, A., Bonsi, P., Sharma, N., Bernardi, G., and Standaert, D.G. (2006). Altered responses to dopaminergic D2 receptor activation and N-type calcium currents in striatal cholinergic interneurons in a mouse model of DYT1 dystonia. Neurobiol Dis *24*, 318-325.

Pizoli, C.E., Jinnah, H.A., Billingsley, M.L., and Hess, E.J. (2002). Abnormal cerebellar signaling induces dystonia in mice. J Neurosci *22*, 7825-7833.

Politis, M., Pavese, N., Tai, Y.F., Kiferle, L., Mason, S.L., Brooks, D.J., Tabrizi, S.J., Barker, R.A., and Piccini, P. (2011). Microglial activation in regions related to cognitive function predicts disease onset in Huntington's disease: a multimodal imaging study. Hum Brain Mapp *32*, 258-270.

Polleux, F., Dehay, C., and Kennedy, H. (1996). Gradients and timing of the arrival of thalamic axons in the mouse neocortex. Paper presented at: Soc Neurosci Abstr.

Popolo, M., McCarthy, D.M., and Bhide, P.G. (2004). Influence of dopamine on precursor cell proliferation and differentiation in the embryonic mouse telencephalon. Dev Neurosci *26*, 229-244.

Pouladi, M.A., Morton, A.J., and Hayden, M.R. (2013). Choosing an animal model for the study of Huntington's disease. Nature Reviews Neuroscience *14*, 708.

Pouladi, M.A., Stanek, L.M., Xie, Y., Franciosi, S., Southwell, A.L., Deng, Y., Butland, S., Zhang, W., Cheng, S.H., and Shihabuddin, L.S. (2012). Marked differences in neurochemistry and aggregates despite similar behavioural and neuropathological features of Huntington disease in the full-length BACHD and YAC128 mice. Hum Mol Genet *21*, 2219-2232.

Prensa, L., Parent, A., and Gimenez-Amaya, J.M. (1999). [Compartmentalized organization of human corpus striatum]. Revista de neurologia *28*, 512-519.

Priestley, J.V., Somogyi, P., and Cuello, A.C. (1981). Neurotransmitter-specific projection neurons revealed by combining PAP immunohistochemistry with retrograde transport of HRP. Brain Res *220*, 231-240.

Quartarone, A. (2013). Transcranial magnetic stimulation in dystonia. Handb Clin Neurol *116*, 543-553.

Quartarone, A., Bagnato, S., Rizzo, V., Morgante, F., Sant'Angelo, A., Crupi, D., Romano, M., Messina, C., Berardelli, A., and Girlanda, P. (2005). Corticospinal excitability during motor imagery of a simple tonic finger movement in patients with writer's cramp. Mov Disord *20*, 1488-1495.

Quartarone, A., Morgante, F., Sant'angelo, A., Rizzo, V., Bagnato, S., Terranova, C., Siebner, H.R., Berardelli, A., and Girlanda, P. (2008). Abnormal plasticity of sensorimotor circuits extends beyond the affected body part in focal dystonia. J Neurol Neurosurg Psychiatry *79*, 985-990.

Quartarone, A., and Pisani, A. (2011). Abnormal plasticity in dystonia: Disruption of synaptic homeostasis. Neurobiol Dis *42*, 162-170.

Quartarone, A., Rizzo, V., Terranova, C., Morgante, F., Schneider, S., Ibrahim, N., Girlanda, P., Bhatia, K.P., and Rothwell, J.C. (2009). Abnormal sensorimotor plasticity in organic but not in psychogenic dystonia. Brain *132*, 2871-2877.

Raju, D.V., Shah, D.J., Wright, T.M., Hall, R.A., and Smith, Y. (2006). Differential synaptology of vGluT2-containing thalamostriatal afferents between the patch and matrix compartments in rats. J Comp Neurol *499*, 231-243.

Ramanathan, S., Hanley, J.J., Deniau, J.-M., and Bolam, J.P. (2002a). Synaptic Convergence of Motor and Somatosensory Cortical Afferents onto GABAergic Interneurons in the Rat Striatum. The Journal of Neuroscience *22*, 8158.

Ramanathan, S., Hanley, J.J., Deniau, J.M., and Bolam, J.P. (2002b). Synaptic convergence of motor and somatosensory cortical afferents onto GABAergic interneurons in the rat striatum. J Neurosci *22*, 8158-8169.

Ramdhani, R.A., Kumar, V., Velickovic, M., Frucht, S.J., Tagliati, M., and Simonyan, K. (2014). What's special about task in dystonia? A voxel-based morphometry and diffusion weighted imaging study. Mov Disord *29*, 1141-1150.

Rantamäki, M., Krahe, R., Paetau, A., Cormand, B., Mononen, I., and Udd, B. (2001). Adult-onset autosomal recessive ataxia with thalamic lesions in a Finnish family. Neurology *57*, 1043-1049.

Rattray, I., Smith, E., Gale, R., Matsumoto, K., Bates, G.P., and Modo, M. (2013). Correlations of behavioral deficits with brain pathology assessed through longitudinal MRI and histopathology in the R6/2 mouse model of HD. PLoS One *8*, e60012.

Raymond, L.A., Andre, V.M., Cepeda, C., Gladding, C.M., Milnerwood, A.J., and Levine, M.S. (2011). Pathophysiology of Huntington's disease: time-dependent alterations in synaptic and receptor function. Neuroscience *198*, 252-273.

Rees, E.M., Farmer, R., Cole, J.H., Haider, S., Durr, A., Landwehrmeyer, B., Scahill, R.I., Tabrizi, S.J., and Hobbs, N.Z. (2014). Cerebellar abnormalities in Huntington's disease: a role in motor and psychiatric impairment? Mov Disord *29*, 1648-1654.

Reilly, C.E. (2001). Wild-type huntingtin up-regulates BDNF transcription in Huntington's disease. Journal of neurology *248*, 920-922.

Reiner, A., Albin, R.L., Anderson, K.D., D'Amato, C.J., Penney, J.B., and Young, A.B. (1988). Differential loss of striatal projection neurons in Huntington disease. Proc Natl Acad Sci U S A *85*, 5733-5737.

Reiner, A., Dragatsis, I., and Dietrich, P. (2011). GENETICS AND NEUROPATHOLOGY OF HUNTINGTON'S DISEASE. International review of neurobiology *98*, 325-372.

Reiner, A., Shelby, E., Wang, H., Demarch, Z., Deng, Y., Guley, N.H., Hogg, V., Roxburgh, R., Tippett, L.J., Waldvogel, H.J*., et al.* (2013). Striatal parvalbuminergic neurons are lost in Huntington's disease: implications for dystonia. Mov Disord *28*, 1691-1699.

Reiprich, P., Kilb, W., and Luhmann, H.J. (2005). Neonatal NMDA receptor blockade disturbs neuronal migration in rat somatosensory cortex in vivo. Cerebral cortex *15*, 349-358.

Ribeiro, F.M., Paquet, M., Ferreira, L.T., Cregan, T., Swan, P., Cregan, S.P., and Ferguson, S.S.G. (2010). Metabotropic Glutamate Receptor-Mediated Cell Signaling Pathways Are Altered in a Mouse Model of Huntington's Disease. The Journal of Neuroscience *30*, 316-324.

Rice, M.E., and Cragg, S.J. (2004). Nicotine amplifies reward-related dopamine signals in striatum. Nature neuroscience *7*, 583.

Richardson, P.J., Dixon, A.K., Lee, K., Bell, M.I., Cox, P.J., Williams, R., Pinnock, R.D., and Freeman, T.C. (2000). Correlating physiology with gene expression in striatal cholinergic neurones. J Neurochem *74*, 839-846.

Richter, F., and Richter, A. (2014). Genetic animal models of dystonia: Common features and diversities. Progress in neurobiology *121*, 91-113.

Rong, R., Ahn, J.-Y., Huang, H., Nagata, E., Kalman, D., Kapp, J.A., Tu, J., Worley, P.F., Snyder, S.H., and Ye, K. (2003). PI3 kinase enhancer–Homer complex couples mGluRI to PI3 kinase, preventing neuronal apoptosis. Nature neuroscience *6*, 1153.

Rosas, H.D., Salat, D.H., Lee, S.Y., Zaleta, A.K., Pappu, V., Fischl, B., Greve, D., Hevelone, N., and Hersch, S.M. (2008). Cerebral cortex and the clinical expression of Huntington's disease: complexity and heterogeneity. Brain *131*, 1057-1068.

Rub, U., Hoche, F., Brunt, E.R., Heinsen, H., Seidel, K., Del Turco, D., Paulson, H.L., Bohl, J., von Gall, C., Vonsattel, J.P*., et al.* (2013). Degeneration of the cerebellum in Huntington's disease (HD): possible relevance for the clinical picture and potential gateway to pathological mechanisms of the disease process. Brain Pathol *23*, 165-177.

Rub, U., Seidel, K., Heinsen, H., Vonsattel, J.P., den Dunnen, W.F., and Korf, H.W. (2016). Huntington's disease (HD): the neuropathology of a multisystem neurodegenerative disorder of the human brain. Brain Pathol *26*, 726-740.

Rub, U., Vonsattel, J.P., Heinsen, H., and Korf, H.W. (2015). Degeneration of Select Motor and Limbic Nuclei of the Thalamus in Huntington's Disease (HD). Adv Anat Embryol Cell Biol *217*, 41-53.

Rudkin, T.M., and Sadikot, A.F. (1999). Thalamic input to parvalbumin-immunoreactive GABAergic interneurons: organization in normal striatum and effect of neonatal decortication. Neuroscience *88*, 1165-1175.

Rumbach, L., Barth, P., Costaz, A., and Mas, J. (1995). Hemidystonia consequent upon ipsilateral vertebral artery occlusion and cerebellar infarction. Mov Disord *10*, 522-525.

Rushlow, W., Naus, C.C., and Flumerfelt, B.A. (1996). Somatostatin and the patch/matrix compartments of the rat caudate-putamen. J Comp Neurol *364*, 184-190.

Rymar, V.V., and Sadikot, A.F. (2007). Laminar fate of cortical GABAergic interneurons is dependent on both birthdate and phenotype. J Comp Neurol *501*, 369-380.

Rymar, V.V., Sasseville, R., Luk, K.C., and Sadikot, A.F. (2004). Neurogenesis and stereological morphometry of calretinin-immunoreactive GABAergic interneurons of the neostriatum. J Comp Neurol *469*, 325-339.

Sadikot, A.F., Burhan, A.M., Belanger, M.C., and Sasseville, R. (1998). NMDA receptor antagonists influence early development of GABAergic interneurons in the mammalian striatum. Brain Res Dev Brain Res *105*, 35-42.

Sadikot, A.F., Leung, K., Mittal, S., Rymar, V.V., Alonso-Vanegas, M., and Luk, K.C. (2005). Anterograde trophic mechanisms participate in pattern formation in the striatum: a role for BDNF in glutamatergic afferents. In The Basal Ganglia VIII (Springer), pp. 219-228.

Sadikot, A.F., Parent, A., and Francois, C. (1992a). Efferent connections of the centromedian and parafascicular thalamic nuclei in the squirrel monkey: a PHA-L study of subcortical projections. J Comp Neurol *315*, 137-159.

Sadikot, A.F., Parent, A., Smith, Y., and Bolam, J.P. (1992b). Efferent connections of the centromedian and parafascicular thalamic nuclei in the squirrel monkey: a light and electron microscopic study of the thalamostriatal projection in relation to striatal heterogeneity. J Comp Neurol *320*, 228-242.

Sadikot, A.F., and Sasseville, R. (1997). Neurogenesis in the mammalian neostriatum and nucleus accumbens: parvalbumin-immunoreactive GABAergic interneurons. J Comp Neurol *389*, 193-211.

Sako, W., Morigaki, R., Kaji, R., Tooyama, I., Okita, S., Kitazato, K., Nagahiro, S., Graybiel, A.M., and Goto, S. (2011). Identification and localization of a neuron-specific isoform of TAF1 in rat brain: implications for neuropathology of DYT3 dystonia. Neuroscience *189*, 100-107.

Sako, W., Murakami, N., Izumi, Y., and Kaji, R. Val66Met polymorphism of brain-derived neurotrophic factor is associated with idiopathic dystonia. Journal of Clinical Neuroscience *22*, 575-577.

Saleh, N., Moutereau, S., Durr, A., Krystkowiak, P., Azulay, J.-P., Tranchant, C., Broussolle, E., Morin, F., Bachoud-Lévi, A.-C., and Maison, P. (2009). Neuroendocrine Disturbances in Huntington's Disease. PLoS One *4*, e4962.

Samadi, P., Boutet, A., Rymar, V.V., Rawal, K., Maheux, J., Kvann, J.C., Tomaszewski, M., Beaubien, F., Cloutier, J.F., Levesque, D.*, et al.* (2013). Relationship between BDNF expression in major striatal afferents, striatum morphology and motor behavior in the R6/2 mouse model of Huntington's disease. Genes, brain, and behavior *12*, 108-124.

Sapp, E., Kegel, K.B., Aronin, N., Hashikawa, T., Uchiyama, Y., Tohyama, K., Bhide, P.G., Vonsattel, J.P., and DiFiglia, M. (2001). Early and progressive accumulation of reactive microglia in the Huntington disease brain. J Neuropathol Exp Neurol *60*, 161-172.

Sasi, M., Vignoli, B., Canossa, M., and Blum, R. (2017). Neurobiology of local and intercellular BDNF signaling. Pflügers Archiv - European Journal of Physiology *469*, 593-610.

Sawiak, S.J., Wood, N.I., and Morton, A.J. (2016). Similar Progression of Morphological and Metabolic Phenotype in R6/2 Mice with Different CAG Repeats Revealed by In Vivo Magnetic Resonance Imaging and Spectroscopy. J Huntingtons Dis *5*, 271-283.

Sawiak, S.J., Wood, N.I., Williams, G.B., Morton, A.J., and Carpenter, T.A. (2009). Voxel-based morphometry in the R6/2 transgenic mouse reveals differences between genotypes not seen with manual 2D morphometry. Neurobiol Dis *33*, 20-27.

Schilling, G., Becher, M.W., Sharp, A.H., Jinnah, H.A., Duan, K., Kotzuk, J.A., Slunt, H.H., Ratovitski, T., Cooper, J.K., Jenkins, N.A.*, et al.* (1999). Intranuclear Inclusions and Neuritic Aggregates in Transgenic Mice Expressing a Mutant N-Terminal Fragment of Huntingtin. Hum Mol Genet *8*, 397-407.

Schmanke, T.D., Villablanca, J.R., Lekht, V., and Patel, H.M. (1998). A critical period for reduced brain vulnerability to developmental injury. II. Volumetric study of the neocortex and thalamus in cats. Brain Res Dev Brain Res *105*, 325-337.

Schmidt-Kastner, R., Wetmore, C., and Olson, L. (1996). COMPARATIVE STUDY OF BRAIN-DERIVED NEUROTROPHIC FACTOR MESSENGER RNA AND PROTEIN AT THE CELLULAR LEVEL SUGGESTS MULTIPLE ROLES IN HIPPOCAMPUS, STRIATUM AND CORTEX. Neuroscience *74*, 161-183.

Schmitz, T.W., Nathan Spreng, R., The Alzheimer's Disease Neuroimaging, I., Weiner, M.W., Aisen, P., Petersen, R., Jack, C.R., Jagust, W., Trojanowki, J.Q., Toga, A.W.*, et al.* (2016). Basal forebrain degeneration precedes and predicts the cortical spread of Alzheimer's pathology. Nat Commun *7*, 13249.

Sciamanna, G., Hollis, R., Ball, C., Martella, G., Tassone, A., Marshall, A., Parsons, D., Li, X., Yokoi, F., Zhang, L.*, et al.* (2012a). Cholinergic dysregulation produced by selective inactivation of the dystonia-associated protein torsinA. Neurobiol Dis *47*, 416-427.

Sciamanna, G., Tassone, A., Mandolesi, G., Puglisi, F., Ponterio, G., Martella, G., Madeo, G., Bernardi, G., Standaert, D.G., Bonsi, P.*, et al.* (2012b). Cholinergic dysfunction alters synaptic integration between thalamostriatal and corticostriatal inputs in DYT1 dystonia. J Neurosci *32*, 11991-12004.

Scontrini, A., Conte, A., Defazio, G., Fiorio, M., Fabbrini, G., Suppa, A., Tinazzi, M., and Berardelli, A. (2009). Somatosensory temporal discrimination in patients with primary focal dystonia. Journal of Neurology, Neurosurgery & Psychiatry *80*, 1315.

Semenova, U., Raeva, S., and Sedov, A. (2016). Participation of the thalamic CM-Pf complex in movement performance in patients with dystonia. Mov Disord *31*, 1398-1404.

Sepers, M.D., and Raymond, L.A. (2014). Mechanisms of synaptic dysfunction and excitotoxicity in Huntington's disease. Drug Discov Today *19*, 990-996.

Seto-Ohshima, A., Emson, P.C., Lawson, E., Mountjoy, C.Q., and Carrasco, L.H. (1988). Loss of matrix calcium-binding protein-containing neurons in Huntington's disease. Lancet *1*, 1252-1255.

Shashidharan, P., Sandu, D., Potla, U., Armata, I.A., Walker, R.H., McNaught, K.S., Weisz, D., Sreenath, T., Brin, M.F., and Olanow, C.W. (2005). Transgenic mouse model of early-onset DYT1 dystonia. Hum Mol Genet *14*, 125-133.

Shehadeh, J., Fernandes, H.B., Zeron Mullins, M.M., Graham, R.K., Leavitt, B.R., Hayden, M.R., and Raymond, L.A. (2006). Striatal neuronal apoptosis is preferentially enhanced by NMDA receptor activation in YAC transgenic mouse model of Huntington disease. Neurobiol Dis *21*, 392-403.

Shulga, A., Blaesse, A., Kysenius, K., Huttunen, H.J., Tanhuanpää, K., Saarma, M., and Rivera, C. (2009). Thyroxin regulates BDNF expression to promote survival of injured neurons. Molecular and Cellular Neuroscience *42*, 408-418.

Sidibé, M., Bevan, M.D., Bolam, J.P., and Smith, Y. (1997). Efferent connections of the internal globus pallidus in the squirrel monkey: I. topography and synaptic organization of the pallidothalamic projection. Journal of Comparative Neurology *382*, 323-347.

Sidibe, M., and Smith, Y. (1999). Thalamic inputs to striatal interneurons in monkeys: synaptic organization and co-localization of calcium binding proteins. Neuroscience *89*, 1189-1208.

Silva, A., Naia, L., Dominguez, A., Ribeiro, M., Rodrigues, J., Vieira, O.V., Lessmann, V., and Rego, A.C. (2015). Overexpression of BDNF and Full-Length TrkB Receptor Ameliorate Striatal Neural Survival in Huntington's Disease. Neurodegenerative Diseases *15*, 207-218.

Simmons, D.A., Belichenko, N.P., Yang, T., Condon, C., Monbureau, M., Shamloo, M., Jing, D., Massa, S.M., and Longo, F.M. (2013). A Small Molecule TrkB Ligand Reduces Motor Impairment and Neuropathology in R6/2 and BACHD Mouse Models of Huntington's Disease. The Journal of Neuroscience *33*, 18712.

Slow, E.J., Van Raamsdonk, J., Rogers, D., Coleman, S.H., Graham, R.K., Deng, Y., Oh, R., Bissada, N., Hossain, S.M., and Yang, Y.-Z. (2003). Selective striatal neuronal loss in a YAC128 mouse model of Huntington disease. Hum Mol Genet *12*, 1555-1567.

Smith, G.A., Rocha, E.M., McLean, J.R., Hayes, M.A., Izen, S.C., Isacson, O., and Hallett, P.J. (2014a). Progressive axonal transport and synaptic protein changes correlate with behavioral and neuropathological abnormalities in the heterozygous Q175 KI mouse model of Huntington's disease. Hum Mol Genet *23*, 4510-4527.

Smith, R., Chung, H., Rundquist, S., Maat-Schieman, M.L.C., Colgan, L., Englund, E., Liu, Y.-J., Roos, R.A.C., Faull, R.L.M., Brundin, P.*, et al.* (2006). Cholinergic neuronal defect without cell loss in Huntington's disease. Hum Mol Genet *15*, 3119-3131.

Smith, Y., Bevan, M.D., Shink, E., and Bolam, J.P. (1998). Microcircuitry of the direct and indirect pathways of the basal ganglia. Neuroscience *86*, 353-387.

Smith, Y., and Bolam, J.P. (1989). Neurons of the substantia nigra reticulata receive a dense GABA-containing input from the globus pallidus in the rat. Brain Res *493*, 160-167.

Smith, Y., Galvan, A., Ellender, T.J., Doig, N., Villalba, R.M., Huerta-Ocampo, I., Wichmann, T., and Bolam, J.P. (2014b). The thalamostriatal system in normal and diseased states. Front Syst Neurosci *8*, 5.

Smith, Y., and Parent, A. (1986). Differential connections of caudate nucleus and putamen in the squirrel monkey (Saimiri sciureus). Neuroscience *18*, 347-371.

Sohur, U.S., Padmanabhan, H.K., Kotchetkov, I.S., Menezes, J.R.L., and Macklis, J.D. (2014). Anatomic and Molecular Development of Corticostriatal Projection Neurons in Mice. Cerebral cortex *24*, 293-303.

Song, C.-H., Fan, X., Exeter, C.J., Hess, E.J., and Jinnah, H.A. (2012). Functional analysis of dopaminergic systems in a DYT1 knock-in mouse model of dystonia. Neurobiol Dis *48*, 66-78.

Song, C., Zhang, Y., Parsons, C.G., and Liu, Y.F. (2003). Expression of polyglutamine-expanded huntingtin induces tyrosine phosphorylation of N-methyl-D-aspartate receptors. J Biol Chem *278*, 33364-33369.

Spokes, E.G. (1980). Neurochemical alterations in Huntington's chorea: a study of post-mortem brain tissue. Brain *103*, 179-210.

Squitieri, F., Gellera, C., Cannella, M., Mariotti, C., Cislaghi, G., Rubinsztein, D.C., Almqvist, E.W., Turner, D., Bachoud-Levi, A.C., Simpson, S.A.*, et al.* (2003). Homozygosity for CAG mutation in Huntington disease is associated with a more severe clinical course. Brain *126*, 946-955.

Stack, E.C., Dedeoglu, A., Smith, K.M., Cormier, K., Kubilus, J.K., Bogdanov, M., Matson, W.R., Yang, L., Jenkins, B.G., Luthi-Carter, R.*, et al.* (2007). Neuroprotective effects of synaptic modulation in Huntington's disease R6/2 mice. The Journal of neuroscience : the official journal of the Society for Neuroscience *27*, 12908-12915.

Stack, E.C., Kubilus, J.K., Smith, K., Cormier, K., Del Signore, S.J., Guelin, E., Ryu, H., Hersch, S.M., and Ferrante, R.J. (2005). Chronology of behavioral symptoms and neuropathological sequela in R6/2 Huntington's disease transgenic mice. J Comp Neurol *490*, 354-370.

Stack, E.C., Smith, K.M., Ryu, H., Cormier, K., Chen, M., Hagerty, S.W., Del Signore, S.J., Cudkowicz, M.E., Friedlander, R.M., and Ferrante, R.J. (2006). Combination therapy using minocycline and coenzyme Q10 in R6/2 transgenic Huntington's disease mice. Biochim Biophys Acta *1762*, 373-380.

Standaert, D.G., Friberg, I.K., Landwehrmeyer, G.B., Young, A.B., and Penney, J.B., Jr. (1999). Expression of NMDA glutamate receptor subunit mRNAs in neurochemically identified projection and interneurons in the striatum of the rat. Brain Res Mol Brain Res *64*, 11-23.

Steele, J.C., Richardson, J., and Olszewski, J. (1964). Progressive supranuclear palsy: A heterogeneous degeneration involving the brain stem, basal ganglia and cerebellum with vertical gaze and pseudobulbar palsy, nuchal dystonia and dementia. Arch Neurol *10*, 333-359.

Steinbusch, H.W., van der Kooy, D., Verhofstad, A.A., and Pellegrino, A. (1980). Serotonergic and non-serotonergic projections from the nucleus raphe dorsalis to the caudate-putamen complex in the rat, studied by a combined immunofluorescence and fluorescent retrograde axonal labeling technique. Neurosci Lett *19*, 137-142.

Steininger, T.L., Wainer, B.H., Klein, R., Barbacid, M., and Palfrey, H.C. (1993). High-affinity nerve growth factor receptor (Trk) immunoreactivity is localized in cholinergic neurons of the basal forebrain and striatum in the adult rat brain. Brain Res *612*, 330-335.

Surmeier, D.J., Ding, J., Day, M., Wang, Z., and Shen, W. (2007). D1 and D2 dopamine-receptor modulation of striatal glutamatergic signaling in striatal medium spiny neurons. Trends Neurosci *30*, 228-235.

Suzuki, M., Desmond, T.J., Albin, R.L., and Frey, K.A. (2001). Vesicular neurotransmitter transporters in Huntington's disease: initial observations and comparison with traditional synaptic markers. Synapse *41*, 329-336.

Suzuki, M., Nelson, A.D., Eickstaedt, J.B., Wallace, K., Wright, L.S., and Svendsen, C.N. (2006). Glutamate enhances proliferation and neurogenesis in human neural progenitor cell cultures derived from the fetal cortex. Eur J Neurosci *24*, 645-653.

Szabo, J. (1980). Distribution of striatal afferents from the mesencephalon in the cat. Brain Res *188*, 3-21.

Tai, Y.F., Pavese, N., Gerhard, A., Tabrizi, S.J., Barker, R.A., Brooks, D.J., and Piccini, P. (2007). Imaging microglial activation in Huntington's disease. Brain Res Bull *72*, 148-151.

Tallaksen-Greene, S.J., Crouse, A.B., Hunter, J.M., Detloff, P.J., and Albin, R.L. (2005). Neuronal intranuclear inclusions and neuropil aggregates in HdhCAG(150) knockin mice. Neuroscience *131*, 843-852.

Tallaksen-Greene, S.J., Kaatz, K.W., Romano, C., and Albin, R.L. (1998). Localization of mGluR1a-like immunoreactivity and mGluR5-like immunoreactivity in identified populations of striatal neurons. Brain Res *780*, 210-217.

Tang, B., Seredenina, T., Coppola, G., Kuhn, A., Geschwind, D.H., Luthi-Carter, R., and Thomas, E.A. (2011). Gene expression profiling of R6/2 transgenic mice with different CAG repeat lengths reveals genes associated with disease onset and progression in Huntington's disease. Neurobiol Dis *42*, 459-467.

Tanimura, A., Du, Y., Kondapalli, J., Wokosin, D.L., and Surmeier, D.J. (2019). Cholinergic Interneurons Amplify Thalamostriatal Excitation of Striatal Indirect Pathway Neurons in Parkinson's Disease Models. Neuron *101*, 444-458.e446.

Tanimura, A., Lim, S.A., Aceves Buendia, J.J., Goldberg, J.A., and Surmeier, D.J. (2016). Cholinergic Interneurons Amplify Corticostriatal Synaptic Responses in the Q175 Model of Huntington's Disease. Front Syst Neurosci *10*, 102.

Team, R.C. (2018). R: A language and environment for statistical computing; 2015.

Teipel, S.J., Cavedo, E., Hampel, H., and Grothe, M.J. (2018). Basal Forebrain Volume, but Not Hippocampal Volume, Is a Predictor of Global Cognitive Decline in Patients With Alzheimer's Disease Treated With Cholinesterase Inhibitors. Front Neurol *9*, 642.

Teo, R.T., Hong, X., Yu-Taeger, L., Huang, Y., Tan, L.J., Xie, Y., To, X.V., Guo, L., Rajendran, R., Novati, A*., et al.* (2016). Structural and molecular myelination deficits occur prior to neuronal loss in the YAC128 and BACHD models of Huntington disease. Hum Mol Genet *25*, 2621-2632.

Tepper, J.M., and Bolam, J.P. (2004). Functional diversity and specificity of neostriatal interneurons. Curr Opin Neurobiol *14*, 685-692.

Threlfell, S., Lalic, T., Platt, N.J., Jennings, K.A., Deisseroth, K., and Cragg, S.J. (2012). Striatal dopamine release is triggered by synchronized activity in cholinergic interneurons. Neuron *75*, 58-64.

Tinazzi, M., Rosso, T., and Fiaschi, A. (2003). Role of the somatosensory system in primary dystonia. Movement Disorders *18*, 605-622.

Tippett, L.J., Waldvogel, H.J., Thomas, S.J., Hogg, V.M., van Roon-Mom, W., Synek, B.J., Graybiel, A.M., and Faull, R.L. (2007). Striosomes and mood dysfunction in Huntington's disease. Brain *130*, 206-221.

Tolosa, E., and Compta, Y. (2006). Dystonia in Parkinson’s disease. Journal of neurology *253*, vii7-vii13.

Uluğ, A.M., Vo, A., Argyelan, M., Tanabe, L., Schiffer, W.K., Dewey, S., Dauer, W.T., and Eidelberg, D. (2011). Cerebellothalamocortical pathway abnormalities in torsinA DYT1 knock-in mice. Proceedings of the National Academy of Sciences *108*, 6638-6643.

Van Der Kooy, D., and Carter, D.A. (1981). The organization of the efferent projections and striatal afferents of the entopeduncular nucleus and adjacent areas in the rat. Brain Res *211*, 15-36.

van Harten, P.N., Hoek, H.W., Matroos, G.E., Koeter, M., and Kahn, R.S. (1997). The inter-relationships of tardive dyskinesia, parkinsonism, akathisia and tardive dystonia: the Curaçao Extrapyramidal Syndromes Study II. Schizophrenia Research *26*, 235-242.

Van Raamsdonk, J.M., Murphy, Z., Slow, E.J., Leavitt, B.R., and Hayden, M.R. (2005). Selective degeneration and nuclear localization of mutant huntingtin in the YAC128 mouse model of Huntington disease. Hum Mol Genet *14*, 3823-3835.

VanDongen, A.M. (2008). Biology of the NMDA Receptor (CRC Press).

Vanhoutte, P., and Bading, H. (2003). Opposing roles of synaptic and extrasynaptic NMDA receptors in neuronal calcium signalling and BDNF gene regulation. Current Opinion in Neurobiology *13*, 366-371.

Velier, J., Kim, M., Schwarz, C., Kim, T.W., Sapp, E., Chase, K., Aronin, N., and DiFiglia, M. (1998). Wild-type and mutant huntingtins function in vesicle trafficking in the secretory and endocytic pathways. Exp Neurol *152*, 34-40.

Vetter, J.M., Jehle, T., Heinemeyer, J., Franz, P., Behrens, P.F., Jackisch, R., Landwehrmeyer, G.B., and Feuerstein, T.J. (2003). Mice transgenic for exon 1 of Huntington's disease: properties of cholinergic and dopaminergic pre-synaptic function in the striatum. J Neurochem *85*, 1054-1063.

Villablance, J.R., Burgess, J.W., and Olmstead, C.E. (1986). Recovery of function after neonatal or adult hemispherectomy in cats: I. Time course, movement, posture and sensorimotor tests. Behavioural brain research *19*, 205-226.

Villalba, R.M., Wichmann, T., and Smith, Y. (2014). Neuronal loss in the caudal intralaminar thalamic nuclei in a primate model of Parkinson's disease. Brain structure & function *219*, 381-394.

Vitalis, T., and Parnavelas, J.G. (2003). The Role of Serotonin in Early Cortical Development. Dev Neurosci *25*, 245-256.

Vonsattel, J.P., and DiFiglia, M. (1998). Huntington disease. J Neuropathol Exp Neurol *57*, 369-384.

Vonsattel, J.P., Myers, R.H., Stevens, T.J., Ferrante, R.J., Bird, E.D., and Richardson, E.P., Jr. (1985). Neuropathological classification of Huntington's disease. J Neuropathol Exp Neurol *44*, 559-577.

Voorn, P., Vanderschuren, L.J.M.J., Groenewegen, H.J., Robbins, T.W., and Pennartz, C.M.A. (2004). Putting a spin on the dorsal–ventral divide of the striatum. Trends in Neurosciences *27*, 468-474.

Wahlsten, D. (2011). Chapter 4 - Designs. In Mouse Behavioral Testing (London: Academic Press), pp. 53-73.

Waldvogel, H.J., Thu, D., Hogg, V., Tippett, L., and Faull, R.L. (2012). Selective neurodegeneration, neuropathology and symptom profiles in Huntington's disease. Adv Exp Med Biol *769*, 141-152.

Walker, R.H., Arbuthnott, G.W., Baughman, R.W., and Graybiel, A.M. (1993). Dendritic domains of medium spiny neurons in the primate striatum: relationships to striosomal borders. J Comp Neurol *337*, 614-628.

Walker, R.H., and Graybiel, A.M. (1993). Dendritic arbors of spiny neurons in the primate striatum are directionally polarized. J Comp Neurol *337*, 629-639.

Wall, N.R., De La Parra, M., Callaway, E.M., and Kreitzer, A.C. (2013). Differential innervation of direct- and indirect-pathway striatal projection neurons. Neuron *79*, 347-360.

Wallace, J.A., and Lauder, J.M. (1983). Development of the serotonergic system in the rat embryo: an immunocytochemical study. Brain Res Bull *10*, 459-479.

Watabe-Uchida, M., Zhu, L., Ogawa, Sachie K., Vamanrao, A., and Uchida, N. (2012). Whole-Brain Mapping of Direct Inputs to Midbrain Dopamine Neurons. Neuron *74*, 858-873.

Weiss, K.R., and Littleton, J.T. (2016). Characterization of axonal transport defects in Drosophila Huntingtin mutants. Journal of neurogenetics *30*, 212-221.

Wexler, N.S., Young, A.B., Tanzi, R.E., Travers, H., Starosta-Rubinstein, S., Penney, J.B., Snodgrass, S.R., Shoulson, I., Gomez, F., Ramos Arroyo, M.A.*, et al.* (1987). Homozygotes for Huntington's disease. Nature *326*, 194-197.

Wheeler, V.C., White, J.K., Gutekunst, C.-A., Vrbanac, V., Weaver, M., Li, X.-J., Li, S.-H., Yi, H., Vonsattel, J.-P., and Gusella, J.F. (2000). Long glutamine tracts cause nuclear localization of a novel form of huntingtin in medium spiny striatal neurons in Hdh Q92 and Hdh Q111 knock-in mice. Hum Mol Genet *9*, 503-513.

Wilson, B.K., and Hess, E.J. (2013). Animal models for dystonia. Mov Disord *28*, 982-989.

Wilson, C.J., Chang, H.T., and Kitai, S.T. (1990). Firing patterns and synaptic potentials of identified giant aspiny interneurons in the rat neostriatum. The Journal of neuroscience : the official journal of the Society for Neuroscience *10*, 508-519.

Wobbrock, J.O., Findlater, L., Gergle, D., and Higgins, J.J. (2011). The aligned rank transform for nonparametric factorial analyses using only anova procedures. In Proceedings of the SIGCHI Conference on Human Factors in Computing Systems (Vancouver, BC, Canada: ACM), pp. 143-146.

Wood, N.I., Carta, V., Milde, S., Skillings, E.A., McAllister, C.J., Ang, Y.L., Duguid, A., Wijesuriya, N., Afzal, S.M., Fernandes, J.X.*, et al.* (2010). Responses to environmental enrichment differ with sex and genotype in a transgenic mouse model of Huntington's disease. PLoS One *5*, e9077.

Wood, N.I., Glynn, D., and Morton, A.J. (2011). "Brain training" improves cognitive performance and survival in a transgenic mouse model of Huntington's disease. Neurobiol Dis *42*, 427-437.

Wood, N.I., Goodman, A.O., van der Burg, J.M., Gazeau, V., Brundin, P., Bjorkqvist, M., Petersen, A., Tabrizi, S.J., Barker, R.A., and Morton, A.J. (2008). Increased thirst and drinking in Huntington's disease and the R6/2 mouse. Brain Res Bull *76*, 70-79.

Wood, N.I., and Morton, A.J. (2015). Social Behaviour is Impaired in the R6/2 Mouse Model of Huntington's Disease. J Huntingtons Dis *4*, 61-73.

Wood, N.I., Pallier, P.N., Wanderer, J., and Morton, A.J. (2007). Systemic administration of Congo red does not improve motor or cognitive function in R6/2 mice. Neurobiol Dis *25*, 342-353.

Xie, Y., Hayden, M.R., and Xu, B. (2010). BDNF Overexpression in the Forebrain Rescues Huntington's Disease Phenotypes in YAC128 Mice. The Journal of Neuroscience *30*, 14708-14718.

Xu, B., Zang, K., Ruff, N.L., Zhang, Y.A., McConnell, S.K., Stryker, M.P., and Reichardt, L.F. (2000). Cortical Degeneration in the Absence of Neurotrophin Signaling: Dendritic Retraction and Neuronal Loss after Removal of the Receptor TrkB. Neuron *26*, 233-245.

Yang, D., Wang, C.-E., Zhao, B., Li, W., Ouyang, Z., Liu, Z., Yang, H., Fan, P., O'neill, A., and Gu, W. (2010). Expression of Huntington's disease protein results in apoptotic neurons in the brains of cloned transgenic pigs. Hum Mol Genet *19*, 3983-3994.

Yang, J., Luo, C., Song, W., Guo, X., Zhao, B., Chen, X., Huang, X., Gong, Q., and Shang, H.F. (2014). Diffusion tensor imaging in blepharospasm and blepharospasm-oromandibular dystonia. Journal of neurology *261*, 1413-1424.

Yang, S.-H., Cheng, P.-H., Banta, H., Piotrowska-Nitsche, K., Yang, J.-J., Cheng, E.C.H., Snyder, B., Larkin, K., Liu, J., Orkin, J*., et al.* (2008). Towards a transgenic model of Huntington's disease in a non-human primate. Nature *453*, 921.

Young, A.B., Greenamyre, J.T., Hollingsworth, Z., Albin, R., D'Amato, C., Shoulson, I., and Penney, J.B. (1988). NMDA receptor losses in putamen from patients with Huntington's disease. Science *241*, 981-983.

Yu-Taeger, L., Petrasch-Parwez, E., Osmand, A.P., Redensek, A., Metzger, S., Clemens, L.E., Park, L., Howland, D., Calaminus, C., Gu, X*., et al.* (2012). A novel BACHD transgenic rat exhibits characteristic neuropathological features of Huntington disease. J Neurosci *32*, 15426-15438.

Zadro, I., Brinar, V.V., Barun, B., Ozretic, D., and Habek, M. (2008). Cervical dystonia due to cerebellar stroke. Mov Disord *23*, 919-920.

Zaja-Milatovic, S., Milatovic, D., Schantz, A.M., Zhang, J., Montine, K.S., Samii, A., Deutch, A.Y., and Montine, T.J. (2005). Dendritic degeneration in neostriatal medium spiny neurons in Parkinson disease. Neurology *64*, 545.

Zeron, M.M., Hansson, O., Chen, N., Wellington, C.L., Leavitt, B.R., Brundin, P., Hayden, M.R., and Raymond, L.A. (2002). Increased sensitivity to N-methyl-D-aspartate receptor-mediated excitotoxicity in a mouse model of Huntington's disease. Neuron *33*, 849-860.

Zeuner, K.E., Knutzen, A., Granert, O., Gotz, J., Wolff, S., Jansen, O., Dressler, D., Hefter, H., Hallett, M., Deuschl, G*., et al.* (2015). Increased volume and impaired function: the role of the basal ganglia in writer's cramp. Brain Behav *5*, e00301.

Zhang, H., Das, S., Li, Q.Z., Dragatsis, I., Repa, J., Zeitlin, S., Hajnoczky, G., and Bezprozvanny, I. (2008a). Elucidating a normal function of huntingtin by functional and microarray analysis of huntingtin-null mouse embryonic fibroblasts. BMC neuroscience *9*, 38.

Zhang, H., Li, Q., Graham, R.K., Slow, E., Hayden, M.R., and Bezprozvanny, I. (2008b). Full length mutant huntingtin is required for altered Ca2+ signaling and apoptosis of striatal neurons in the YAC mouse model of Huntington's disease. Neurobiol Dis *31*, 80-88.

Zhang, W., Basile, A.S., Gomeza, J., Volpicelli, L.A., Levey, A.I., and Wess, J. (2002). Characterization of Central Inhibitory Muscarinic Autoreceptors by the Use of Muscarinic Acetylcholine Receptor Knock-Out Mice. The Journal of Neuroscience *22*, 1709-1717.

Zheng, J., Shen, W.H., Lu, T.J., Zhou, Y., Chen, Q., Wang, Z., Xiang, T., Zhu, Y.C., Zhang, C., Duan, S*., et al.* (2008). Clathrin-dependent endocytosis is required for TrkB-dependent Akt-mediated neuronal protection and dendritic growth. J Biol Chem *283*, 13280-13288.

Zuccato, C., Belyaev, N., Conforti, P., Ooi, L., Tartari, M., Papadimou, E., MacDonald, M., Fossale, E., Zeitlin, S., Buckley, N*., et al.* (2007). Widespread Disruption of Repressor Element-1 Silencing

Transcription Factor/Neuron-Restrictive Silencer Factor Occupancy at Its Target Genes in Huntington's Disease. The Journal of Neuroscience *27*, 6972-6983.

Zuccato, C., and Cattaneo, E. (2007). Role of brain-derived neurotrophic factor in Huntington's disease. Progress in neurobiology *81*, 294-330.

Zuccato, C., and Cattaneo, E. (2009). Brain-derived neurotrophic factor in neurodegenerative diseases. Nature Reviews Neurology *5*, 311.

Zuccato, C., Ciammola, A., Rigamonti, D., Leavitt, B.R., Goffredo, D., Conti, L., MacDonald, M.E., Friedlander, R.M., Silani, V., Hayden, M.R.*, et al.* (2001). Loss of Huntingtin-Mediated BDNF Gene Transcription in Huntington's Disease. Science *293*, 493-498.

Zuccato, C., Liber, D., Ramos, C., Tarditi, A., Rigamonti, D., Tartari, M., Valenza, M., and Cattaneo, E. (2005). Progressive loss of BDNF in a mouse model of Huntington's disease and rescue by BDNF delivery. Pharmacological Research *52*, 133-139.

Zuccato, C., Marullo, M., Conforti, P., MacDonald, M.E., Tartari, M., and Cattaneo, E. (2008). RESEARCH ARTICLE: Systematic Assessment of BDNF and Its Receptor Levels in Human Cortices Affected by Huntington's Disease. Brain Pathology *18*, 225-238.

Zuccato, C., Tartari, M., Crotti, A., Goffredo, D., Valenza, M., Conti, L., Cataudella, T., Leavitt, B.R., Hayden, M.R., Timmusk, T.*, et al.* (2003). Huntingtin interacts with REST/NRSF to modulate the transcription of NRSE-controlled neuronal genes. Nat Genet *35*, 76.