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# Recent advances in using *Drosophila* to model neurodegenerative diseases

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## Abstract

Neurodegenerative diseases are progressive disorders of the nervous system that affect the function and maintenance of specific neuronal populations. Most disease cases are sporadic with no known cause. The identification of genes associated with familial cases of these diseases has enabled the development of animal models to study disease mechanisms. The model organism *Drosophila* has been successfully used to study pathogenic mechanisms of a wide range of neurodegenerative diseases. Recent genetic studies in the *Drosophila* models have provided new insights into disease mechanisms, emphasizing the roles played by mitochondrial dynamics, RNA (including miRNA) function, protein translation, and synaptic plasticity and differentiation. It is anticipated that *Drosophila* models will further our understanding of mechanisms of neurodegeneration and facilitate the development of novel and rational treatments for these debilitating neurodegenerative diseases.

### Keywords

Alzheimer's disease; Parkinson's disease; Tauopathies; Polyglutamine diseases; Prion disease; Amyotrophic lateral sclerosis; Spinal muscular atrophy; Batten disease; Mitochondrial dynamics; Synaptic dysfunction

## Building neurodegenerative disease models in *Drosophila*

Neurodegenerative diseases represent a subgroup of diseases sharing certain features. For example, these diseases are often of adult onset and disease pathogenesis usually involves the progressive loss of specific neuronal populations characteristic for each disease type. These diseases were once considered among the most obscure and intractable of all human illnesses. Thanks to the molecular cloning of genes associated with familial forms of such diseases as Alzheimer's disease (AD) and Parkinson's disease (PD), the pathogenic mechanisms of neurodegeneration can now be studied at the molecular level. One of the powerful approaches for studying disease mechanism is the establishment of animal models. *Drosophila* has proven to be an excellent tool for modeling human neurodegenerative diseases. A number of excellent reviews have been written on this topic recently [1–4]. This field is progressing rapidly. The aim of this review is to summarize recent developments in using *Drosophila* to study various human neurodegenerative diseases.

Animal models offers a number of advantages for studying human neurodegenerative diseases. For example, they allow genetic analysis of disease gene relationships and

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delineation of cellular pathways involved in disease pathogenesis. They also allow studies of molecular and cellular changes of the relevant brain cell populations at various stages of the disease process, from pre-symptomatic stage to full-blown disease manifestation.

Four approaches have been employed successfully to study neurodegeneration in *Drosophila*. First, forward genetic screens have been carried out to identify genes that when mutated can cause degeneration of the brain. Examples include the *Drosophila* mutants such as *bub-blegum*, *swisscheese*, and *drop-dead* [5,6]. The mammalian homologues of some of these genes also cause neurodegeneration when mutated, thus validating the usefulness of this approach in identifying conserved genes required to maintain nervous system integrity [7]. Second, transgenic overexpression approach has been used to model disease caused by a toxic gain-of-function (GOF) mechanism [8–11]. Third, a genetic inhibition of endogenous gene approach has been used to model the subset of familial diseases transmitted in a recessive fashion, which are likely caused by a loss-of-function (LOF) mechanism. Transposon-mediated mutagenesis, transgenic RNA interference (RNAi), and homologous recombination-based gene knockout can be used for this purpose [12–15]. Fourth, a pharmacological approach can be used to model neurodegenerative diseases and to test candidate therapeutics in animals [16,17]. Below we summarize the fly models established using the first three genetic approaches and lessons learned from them.

## PD models

PD is the most common movement disorder and the second most common neurodegenerative disease. The movement abnormality in PD is largely attributed to the deficiency of brain dopamine content caused by degeneration of dopaminergic neurons in the substantia nigra. A pathological hallmark of the disease is the formation of Lewy bodies, intracytoplasmic inclusions that are composed mainly of α-synuclein and ubiquitin. The most common forms of PD are sporadic with no known cause. Nevertheless, postmortem studies of sporadic cases have identified defective mitochondrial complex I function and oxidative damage in nigrostriatal dopaminergic neurons [18,19]. It is now well established that genetic factors contribute to PD pathogenesis. At least 10 distinct loci (PARK1-11) have been linked to rare familial forms of PD (FPD) [20,21]. To date, six FPD genes have been molecularly cloned, including α-synuclein (α-Syn), Parkin, Ubiquitin C-terminal hydrolase-1 (Uchl-1), DJ-1, PTEN induced kinase-1 (PINK-1), and Leucine-rich repeat kinase 2 (LRRK2) [22–28]. In the cases of α-Syn and LRRK2, disease modeling was achieved through transgenic overexpression of wild type or pathogenic forms of proteins, whereas for DJ-1, Parkin, and Pink1, disease models were created by genetic LOF approach.

### α-Syn models

α-Syn encodes a small protein whose normal function is likely involved in synaptic transmission. It is a main component of Lewy bodies and its aggregation is frequently found in PD. Although *Drosophila* does not have a clear α-Syn homologue, overexpression of wild-type and FPD-associated mutant α-Syn in *Drosophila* has recapitulated key features of PD, including Lewy body–like aggregate formation, selective degeneration of dopaminergic neurons, and locomotor behavior abnormality [10,29]. The fact that both mutant and wild-type α-Syn form aggregates in fly neurons and cause disease phenotypes supports the notion that improper disposal of aggregation-prone abnormal proteins can result in neurodegeneration. This is also consistent with the finding that genomic triplication of α-Syn gene can cause PD [30].

Consistent with a role for aberrant protein quality control in α-Syn pathogenesis, directed expression of the molecular chaperone HSP70, prevented dopaminergic neuronal loss associated with α-Syn expression in *Drosophila* [29]. A recent human genetics study

indicated that polymorphisms in Hsp70 genes may affect susceptibility to PD [31]. The potential of Hsp70 gene therapy in a mouse model of MPTP-induced PD was also tested. Viral vector-mediated Hsp70 gene transfer to dopamine neurons significantly protected mouse dopaminergic system against MPTP-induced dopaminergic toxicity [32]. Increasing chaperone activity may offer a new avenue for the treatment of PD.

Mammalian studies have previously shown that α-Syn present in Lewy bodies is phosphorylated at serine 129 (Ser129) [33]. Chen and Feany showed that mutating Ser129 to non-phosphorylatable alanine completely suppressed α-Syn toxicity, whereas changing Ser129 to the phosphomimetic aspartate significantly enhanced α-Syn toxicity in the fly model. Interestingly, blocking phosphorylation at Ser129 substantially increased aggregate formation while preventing neurotoxicity [34]. In another study using deletion constructs of α-Syn, however, a positive role for aggregation of α-Syn in mediating dopaminergic neuron toxicity in vivo was proposed [35]. The precise mechanism by which the different aggregated form of α-Syn may exert differential effects on neurotoxicity remains to be determined.

The fly α-Syn model has been used to test candidate factors suspected of playing a role in the pathogenesis of PD. Aging has been firmly established as a risk factor for most neurodegenerative diseases. In one study the role of sirtuins, members of the histone deacetylase family of proteins that participate in aging, in regulating α-Syn toxicity was tested. Pharmacological application of a potent inhibitor of sirtuin 2 (SIRT2) or genetic inhibition of SIRT2 via small interfering RNA rescued α-Syn toxicity in a cellular model. The inhibitors also protected against dopaminergic cell death in a *Drosophila* α-Syn model of PD [36]. In another study, the role of oxidative stress in α-Syn pathogenesis was tested. It was found that dopaminergic neurons are specifically sensitive to hyperoxia induced oxidative stress and that mutant forms of α-Syn show an enhanced toxicity under these conditions. The co-expression of Cu/Zn superoxide dismutase protects against the dopaminergic neuronal loss induced by mutant α-Syn, thus identifying oxidative stress as an important causative factor in α-Syn pathogenesis [37].

Despite these studies, the exact mechanism by which accumulation of wild type α-Syn or expression of mutant α-Syn causes selective dopaminergic neuron degeneration remains elusive. Systematic genetic studies promise to address this question. In one study initially carried out in yeast, α-Syn was shown to cause a block in endoplasmic reticulum (ER)-to-Golgi vesicular trafficking. A genome-wide screen identified toxicity modifiers, including the Rab guanosine triphosphatase Ypt1p, which associated with cytoplasmic α-Syn inclusions. Elevated expression of Rab1, the metazoan *YPT1* homolog, protected against α-Syninduced dopaminergic neuron loss in a fly PD model [38]. This study is beginning to reveal basic cellular functions disrupted by toxic α-Syn expression that may contribute to synucleinopathies. Other genomewide approaches such as DNA microarray analysis of transcriptional profiles or proteomic analysis of protein expression changes in animals ubiquitously expressing α-Syn in the nervous system is beginning to yield useful information [39,40]. Further refinement of the approaches, such as analyzing purified dopaminergic neurons, the disease-relevant population, at different stages of the disease process will provide more insights into mechanisms of α-Syn pathogenesis.

## Parkin and Pink1 models

Parkin encodes a ring-finger domain containing E3 ubiquitin ligase. Its inactivation causes autosomal recessive juvenile parkinsonism. Three labs independently generated *parkin* null mutant flies [12,13,41]. A prominent feature of *parkin* mutant flies is mitochondrial pathology and apoptotic muscle degeneration. In addition, these mutant flies exhibit sterility, reduced lifespan, reduced cell number and size, and hypersensitivity to oxidative stress.

Whole mount confocal analysis also detected notable dopaminergic neuron loss in the protocerebral posterior lateral (PPL) 1 cluster in *parkin* null flies [42]. Using a different approach, *parkin* function was inhibited by transgenic RNAi [14]. Parkin RNAi flies exhibited genetic interaction with transgenic flies overexpressing human Pael-R, a Parkin substrate protein [43], in promoting dopaminergic degeneration, consistent with Pael-R being a Parkin substrate that mediates Parkin deficiency-induced PD. Recent studies in mice also showed that Pael-R induces dopaminergic neurons degeneration in the substantia nigra, and that Pael-R toxicity is enhanced under Parkin inactivation condition [44]. In an interesting twist, overexpression of certain mutant forms of human Parkin in *Drosophila* also led to dopaminergic neurodegeneration and mitochondrial abnormality in indirect flight muscle [45,46], suggesting that at least some of the FPD mutations in Parkin may have gained certain toxic property or act in a dominant-negative fashion.

Pink1 encodes a Ser/Thr kinase with an N-terminal mitochondrial targeting domain. *Drosophila* Pink1 models of PD were generated by transposon-mediated mutagenesis and RNAi approaches. Inhibition of *Drosophila* Pink1 (dPink1) function results in phenotypes strikingly similar to that seen in Parkin mutant or RNAi flies, including male sterility, apoptotic muscle degeneration, dopaminergic neuron loss, defective mitochondrial morphology, and increased sensitivity to multiple stresses including oxidative stress [47–50]. The similar phenotypes observed in Parkin and Pink1 mutant flies prompted genetic epistasis studies to investigate the relationship between these two PD-associated genes. Overexpression of Parkin largely rescued the *Pink1* mutant phenotypes, whereas overexpression of Pink1 had no effect on *Parkin* mutant phenotypes. Further, *pink1 parkin* double mutants showed phenotypes identical to those observed in either single mutant alone. These observations suggest that Pink1 and Parkin function in the same pathway, with Pink1 acting upstream of Parkin [47–49]. Pink1 has recently been shown to directly phosphorylate Parkin and regulate its mitochondrial localization [51], providing a possible molecular explanation for their genetic relationship.

A breakthrough in understanding the exact mitochondrial process regulated by Pink1 and Parkin is provided by the finding that Pink1 and Parkin interact with the mitochondrial fission and fusion pathway genes [52–55]. Mitochondrial fission and fusion are evolutionarily conserved membrane remodeling processes that regulate the morphology and subcellular distribution of mitochondrial units [56]. Mitochondrial fission/fusion has recently been shown to be important for regulating synaptic structure and plasticity [57], with imbalance of mitochondrial fission/ fusion leading to neurodegeneration [58]. Genetic studies strongly suggest that the mitochondrial morphology defects in Pink1 and Parkin mutant are due to decreased fission or increased fusion. Two dynamin-like GTPases, Drp1 and Opa1, are critical components of the machinery that regulate mitochondrial fission and fusion, respectively. Enhancing mitochondrial fission activity by the overexpression of Drp1 or reducing fusion activity by the loss of one copy of *Opa1* can rescue *Pink1* mutant phenotypes in dopaminergic neurons [52]. The biochemical mechanisms linking Pink1/ Parkin to mitochondrial fission/fusion machinery remain to be determined. Parkin overexpression has been shown to protect against dopaminergic neurodegeneration caused by $\alpha$-Syn overexpression in *Drosophila* and mammals [14,59]. It will be interesting to test whether $\alpha$-Syn overexpression also affects mitochondrial morphology or dynamics.

It is intriguing that the indirect flight muscle and dopaminergic neurons are particularly vulnerable to Pink1 and Parkin mutations. It is possible that high endogenous levels of oxidative stress in these tissues may confer this vulnerability. Being one of the most metabolically active tissue, fly indirect flight muscle probably produces more ROS than any other tissues and is presumably under constant oxidative stress [60]. Dopaminergic neurons are also constantly under oxidative stress due to the oxidative metabolism of dopamine. In

fact, a link between dopamine metabolism and α-Syn toxicity has been observed in *Drosophila* primary neuronal culture [61]. Together, these conditions may confer particular vulnerability of these tissues to additional oxidative insults caused by mitochondrial dysfunction in Pink1 or Parkin mutants.

## DJ-1 models

DJ-1 encodes a highly conserved protein belonging to the ThiJ/PfpI superfamily. Its identified molecular functions include molecular chaperone, RNA binding, and anti-oxidant. There are two DJ-1 homologues in *Drosophila*, named DJ-1A and DJ-1B. DJ-1A appears to be a closer homologue of human DJ-1 than DJ-1B based on sequence conservation. DJ-1A and DJ-1B knockout flies display altered sensitivity to environmental toxins such as paraquat, $H_2O_2$, and rotenone [62,63]. Knock down of DJ-1A by transgenic RNAi in a cell type-specific fashion resulted in accumulation of reactive oxygen species, hypersensitivity to oxidative stress, and dysfunction and degeneration of dopaminergic and photoreceptor neurons [64]. It is unclear at this point whether the differential effects between the genetic deletion mutants and the RNAi flies is due to “off-target” effects of RNAi or that certain non-cell autonomous effects of DJ-1A inactivation may have contributed to the tissue-specific RNAi effect.

Using DJ-1A RNAi-induced cell death as an assay, a systematic genetic interaction study was carried out to identify genetic modifiers. This led to the isolation of components of the PI3 K/Akt signaling pathway as specific genetic modifiers [64]. An independent study also found DJ-1A as a genetic modifier of PTEN-induced small-eye phenotype in *Drosophila*, and that DJ-1 RNAi or overexpression affects the phosphorylation of PKB/Akt. Interestingly, in primary breast cancer samples, DJ-1 expression correlates negatively with PTEN immunoreactivity and positively with Akt hyper-phosphorylation [65]. DJ-1 thus appears to be a key negative regulator of PTEN, and may serve as a useful prognostic marker for cancer. These studies raise the interesting possibility that cancer and Parkinson’s disease, two seemingly disparate diseases, may share certain underlying biochemical pathways. The specific molecular function of DJ-1 in these processes is unknown. A recent study showed that DJ-1 associates with RNA targets in cells and brain tissues in an oxidation-dependent manner. RNA targets of DJ-1 include mitochondrial genes, genes involved in glutathione metabolism, and members of the PTEN/PI3K cascade [66]. One possibility is that DJ-1 may regulate the translation of these RNA targets.

## LRRK2 models

LRRK2 encodes a large protein that contains a leucine rich repeat (LRR) domain, a kinase domain, a RAS-like GTPase domain, and a WD-40 domain. Mutations in LRRK2 are frequently found in familial as well as sporadic PD cases, indicating a general role for LRRK2 dysfunction in disease pathogenesis. Although an initial report claimed no dopaminergic degeneration in transgenic flies overexpressing the wild type or R1069C pathogenic forms of the *Drosophila* LRRK2 homologue (dLRRK) [67], later studies expressing the I2020T mutant form of dLRRK or the wild type and the G2019S mutant forms of hLRRK2 did observe dopaminergic neurodegeneration [68,69]. Moreover, both human LRRK2 and dLRRK phosphorylate eukaryotic initiation factor 4E (eIF4E)-binding protein (4E-BP), a negative regulator of eIF4E-mediated protein translation and a key mediator of various stress responses. Modulation of the eIF4E/4E-BP pathway by LRRK2 stimulates eIF4E-mediated protein translation, and attenuates resistance to oxidative stress and survival of DA neuron in *Drosophila*, suggesting that chronic inactivation of 4E-BP by LRRK2 with pathogenic mutations deregulates protein translation and cause age-dependent loss of DA neurons [69]. This study for the first time directly link deregulation of protein translation to PD pathogenesis.

## AD and related tauopathy models

AD is the most common neurodegenerative diseases and the leading cause of dementia. Clinically, it is manifested as a gradual decline of cognitive functions. Two microscopic brain lesions, the amyloid plaque and neurofibrillary tangles (NFT), are diagnostic of AD in the demented patient when present in abnormally high densities for a given age. The main components of the amyloid plaque are the A$\beta$-40 and A$\beta$-42 peptides generated by endoproteolysis of the amyloid precursor protein (APP) via the sequential action of $\beta$- and $\gamma$-secretases. $\beta$-secretase activity is provided by the $\beta$-site APP-cleaving enzyme (BACE), whereas $\gamma$-secretase activity depends on a protein complex consisting of presenilin (Psn), nicastrin, aph-1, and pen-2 [70]. Autosomal dominant mutations in APP, Psn-1, and Psn-2 can accelerate disease onset and progression in familial AD cases. The major component of the NFT is tau protein, a highly soluble, microtubule-binding protein normally enriched in axons [71]. Tau becomes abnormally phosphorylated and insoluble in the NFT [72–74]. Direct evidence of the pathogenicity of tau was provided by the discovery that mutations in tau are associated with frontotemporal dementia with parkinsonism linked to chromosome 17 (FTDP-17) [75–78].

Most of the genes implicated in AD pathogenesis have clear fly homologues. There is a fly homologue of APP (called APP-like or APPL). Flies deficient for APPL exhibit behavioral abnormality, which can be rescued by a human APP transgene, indicating functional conservation between fly APPL and human APP [79]. A recent study found that fly APPL can be processed into A$\beta$ fragments that aggregate into intracellular fibrils, amyloid deposits, and cause age-dependent behavioral deficits and neurodegeneration [80], suggesting that the proteolytic processing and A$\beta$-induced neurotoxicity are conserved for APP proteins. Fly models of human A$\beta$ peptide-induced amyloid formation and neurodegeneration have also been generated. This was achieved either by constructing triple transgenics co-expressing a wild type human BACE transgene, and human APP and fly Psn transgenes containing FAD-linked mutations, or by overexpressing A$\beta$-40 and A$\beta$-42 peptides fused to signal peptides [81,82]. Overexpression studies in *Drosophila* have shown that pathogenic APP can cause axonopathy and neurodegeneration, but this effect of full-length APP is independent of A$\beta$ peptides [83], suggesting that other regions of APP protein exert pathogenic roles as well.

The fly A$\beta$-42 model has been used in genetic modifier screens to identify other genes that act in the same cellular pathway. In one study, the Toll/NFkB signaling pathway was found to mediate the neuropathological effects of human A$\beta$-42 toxicity in *Drosophila*, suggesting the involvement of innate immunity/inflammatory pathway in A$\beta$-42 pathogenesis [84]. In another study, the effect of manipulating Neprilysin on A$\beta$-42 toxicity in *Drosophila* was tested. Neprilysin is one of the rate-limiting A$\beta$-degrading enzymes first found in mammals. In *Drosophila*, overexpression of Neprilysin reduces A$\beta$-42-induced neuronal loss and intraneuronal A$\beta$-42 deposits. However, this manipulation caused a reduction in cAMP-responsive element-binding protein (CEBP)-mediated transcription, age-dependent axonal pathology, and premature death [85]. This finding has therapeutic implications when strategies aimed at enhancing Neprilysin activity are considered.

Tau-induced neurodegeneration has been successfully modeled in *Drosophila* by either expression of a bovine tau-GFP reporter protein [86], or expression of wild type or pathogenic forms of human tau [87,88]. Overexpression of a fly homologue of tau (dtau) also compromised neuronal function [89]. Tau phosphorylation/dephosphorylation has attracted considerable interest in tauopathy research, due to the identification of abnormally phosphorylated tau in NFT. A number of protein kinases and phosphatases have been found to regulate tau phosphorylation in vitro [90]; however, surprisingly little is known about the

in vivo roles of these enzymes in determining tau toxicity. Through LOF and overexpression genetic studies and biochemical analysis, *Drosophila* Partioning defective-1 (PAR-1) was found to be a physiological tau kinase that plays a central role in determining tau phosphorylation and toxicity in *Drosophila* [91]. PAR-1 is a Ser/Thr kinase originally identified in *C. elegans* for its role in regulating cell polarity and asymmetric cell division [92]. MARK kinases, the mammalian homologues of PAR-1, regulate microtubule dynamics, epithelial cell polarity, and neuronal differentiation under normal conditions and bind to NFT in AD brain [93–95]. In the fly tauopathy model, PAR-1 was found to initiate a multisite phosphorylation process that generates pathogenic tau [91]. These results, together with the findings from a genetic screen that uncovered kinases and phosphatases as major modifiers of tau toxicity [96], established the importance of phosphorylation in determining tau toxicity in vivo.

The importance of individual phosphorylation sites in determining tau toxicity in vivo has not been clearly defined. Post-mortem analysis indicated that in the AD patient brain tau is hyperphosphorylated in more than 20 Ser/Thr sites [97]. In theory, these phosphorylation sites could act in concert to confer tau toxicity. Alternatively, a small subset of sites might play a dominant role in determining tau toxicity. Mutagenesis studies has emphasized the importance of a small subset of phosphorylation events controlled by PAR-1/MARK in determining tau toxicity [91,98], whereas a large number of SP/TP sites phosphorylated by proline-directed kinases appear to work in concert to promote tau neurotoxicity in vivo [99].

The mechanism by which tau phosphorylation affects its toxicity remains poorly understood. Phosphorylation could affect the protein-protein interaction between tau and its effector proteins, or regulate the turnover, solubility, or proteolytic cleavage of tau. A recent cross species functional genomic study identified the highly conserved puromycin-sensitive aminopeptidase (PSA/Npepps) as a tau modifier. PSA protected against tau-induced neurodegeneration in vivo, whereas PSA loss of function exacerbated neurodegeneration. Further, human PSA was found to directly proteolyze tau [100]. It would be interesting to test whether the cleavage of tau by PSA is regulated by phosphorylation.

The upstream signalling mechanisms that regulate tau phosphorylation under normal and disease conditions are beginning to be defined. The identification of PAR-1 as a physiological tau kinase offers an excellent entry point to dissect the signalling mechanisms. Recent studies show that phosphorylation of PAR-1 by the tumor suppressor protein LKB1 is required for its activation, which in turn promotes tau phosphorylation in *Drosophila*. Diverse stress stimuli, such as human APP-induced neurotoxicity, can promote PAR-1 activation and tau phosphorylation in an LKB1-dependent manner [101]. These results reveal a new function for the tumor suppressor protein LKB1 in a signalling cascade through which the phosphorylation and function of tau is regulated by AD-related toxic stimuli.

Loss of synaptic function has been recognized as one of the earliest signs of the disease process in AD [102]. Studies in animal models have further supported the role of synaptic dysfunction in disease pathogenesis. APP and APP-like proteins are known to exert normal physiological function at the synapse [79,103]. The mouse transgenic APP model exhibits learning and memory deficits in the absence of obvious neuronal degeneration [104]. Interestingly, removal of endogenous tau can suppress most of the cognitive deficits in the APP model [105], indicating a critical role of tau in mediating the toxic effects of APP at the synapse. This is consistent with the findings that synaptic toxicity is also observed in *Drosophila* tau models [106,107], and that genes involved in synaptogenesis are recovered as tau modifiers in an unbiased genetic screen [108].

It appears that tau is not the only synaptic protein whose abnormality causes synaptic dysfunction in AD. A recent study found that overactivation of PAR-1 in *Drosophila* causes the delocalization of Dlg from the postsynaptic membrane at the neuromuscular junction (NMJ). This is mediated through the direct phosphorylation of Dlg by PAR-1 [109]. Dlg is the fly ortholog of the key postsynaptic scaffold protein PSD-95, whose synaptic localization has been found to be decreased in AD [110–112]. Given that aberrant activation of PAR-1 like kinases has been implicated in AD conditions, this study offers a new direction for understanding the molecular basis of synaptic dysfunction in AD. Further supporting the role of synaptic dysfunction in AD pathogenesis, a recent study in *Drosophila* provided evidence of learning and synaptic defects in a *Drosophila psn* mutant and suggested a presynaptic role for presenilin in normal neuronal function [113].

Compared to early-onset AD, relatively little is known about the genetic control of late-onset AD. Ubiquilin 1 variants have been associated with increased risk for late-onset AD. In *Drosophila*, silencing of endogenous dUbqln in the central nervous system led to age-dependent neurodegeneration. The dUbqln exhibited strong in vivo functional interactions with the AD-associated genes presenilin and APP, providing insights into the potential role of Ubiquilin 1 in AD pathogenesis [114].

## Polyglutamine (polyQ) diseases

A diverse group of dominantly inherited neurodegenerative diseases are caused by CAG trinucleotide repeat expansion within disease-specific proteins. These include Hunting-ton's disease (HD), spinobulbar muscular strophy (SBMA), spinocerebellar ataxia (SCA) 1, 2, 3, 6, 7, and 17; and dentatorubralpallidoluysian atrophy. These polyQ expansions confer toxic properties to the resident proteins. There is a good correlation between repeat length and disease severity [115]. These polyQ repeats form intranuclear inclusions in neurons and cultured cells and are intrinsically toxic. The *Drosophila* compound eye has been an excellent system for modeling these polyQ diseases. In almost all cases, expression of truncated, expanded polyQ-containing protein fragments leads to retinal degeneration. In the case of SCA-1, overexpression of the non-expanded wild type ataxin-1 (Atx-1) protein also resulted in retinal degeneration, indicating that the ataxin-1 protein is intrinsically toxic when present at high levels [11]. Studies in *Drosophila* also revealed that expression of expanded polyQ-containing proteins in glial cells also confers toxicity [116–118], suggesting that degeneration or dysfunction of these cells may also contribute to the disease process.

Significant insights into the pathogenic pathways of polyQ diseases were obtained from comprehensive genetic screens using the SCA models. Loss-of-function and overexpression screens identified genes involved in a number of biological processes, some of which were expected, whereas other are unanticipated, including genes involved in RNA processing, nuclear transport, transcriptional regulation (histone deacetylation), and cellular detoxification [11]. A similar approach applied to the SCA-3 model identified genes involved in protein misfolding and autophagy as modifiers [119]. One interesting finding from the SCA-3 screen is the uncovering of muscleblind (mbl), a gene implicated in the RNA toxicity of CUG expansion diseases, as a SCA-3 modifier, suggesting a toxic role of RNA in polyQ-induced degeneration. Further studies validated this notion. These studies highlight common mechanisms in RNA-based and polyQ-protein-based trinucleotide repeat expansion diseases [120]. Using a candidate gene approach, the miRNA pathway was shown to modulate the neurodegeneration phenotypes of the SCA-3 model [121]. The detailed biochemical mechanism involved in miRNA modulation of SCA-3 toxicity remains to be determined. Given the well established role of the miRNA pathway in regulating protein

translation, it is possible that deregulated synthesis of certain key proteins is involved in SCA-3 pathogenesis.

In a biochemical approach using the SCA-1 model, it was found that the 14-3-3 protein could bind to Atx-1 in a phosphorylation-dependent manner, leading to the stabilization and accumulation of the toxic protein [122]. The PI3K/AKT pathway was found to regulate the association between Atx-1 and 14-3-3 and affect Atx-1 toxicity. AKT phosphorylates Atx-1 at a conserved Ser776 residue and this phosphorylation event provides a binding site for 14-3-3 [122]. Other endogenous proteins that bind to Atx-1 also plays an important role in SCA-1 pathogenesis. Interestingly, a recent study revealed opposing effects of polyglutamine expansion on native protein complexes that both contribute to SCA1 pathogenesis. It was found that polyglutamine expansion in Atx-1 favors the formation of a protein complex containing RBM17, contributing to SCA1 neuropathology through a gain-of-function mechanism. Concomitantly, polyglutamine expansion attenuates the formation and function of another protein complex containing Atx-1 and capicua, contributing to SCA1 via a loss-of-function mechanism [123]. Another interesting finding is that different polyQ genes interact to modulate neurodegeneration in *Drosophila*. Atx-2 was found to mediate the pathogenic effects of SCA-1 and SCA-3. Loss of endogenous Atx-2 function suppressed the toxicity of both SCA-1 and SCA-3 [124,125]. These findings reveal a previously unknown functional link between these disorders with common clinical features but different etiology, and raise the possibility that therapeutic approaches effective for one disease may benefit others.

PolyQ diseases are caused by the expansion of trinucleotide repeat sequences. Disease occurs only when the normally polymorphic number of repeats expands beyond a critical threshold level. Expanded repeats have a strong tendency to further expand, causing the disease to occur earlier and with greater severity in successive generations, a phenomenon called anticipation. Expression of the SCA-3 polyQ transgene in the *Drosophila* germline recapitulated several central features of human CAG repeat instability, including the broad range of repeat changes and strong bias for expansion. Genetic studies revealed that germline transcription along with nucleotide excision repair and/or transcription-coupled DNA repair facilitated repeat instability. Further, inhibition of cAMP response element–binding protein (CREB)–binding protein (CBP), a consequence of polyQ cellular toxicity, enhances repeat instability, revealing the presence of a vicious cycle in polyQ pathogenesis [126].

Earlier *Drosophila* models of HD were built using N-terminal fragment of Htt protein containing polyQ sequence. Although these models have provided important insights into HD, without comparison to a full-length Htt model, it is not clear if these models reliably recapitulated the disease situation. Recent analysis of a full-length *Drosophila* HD model revealed that expression of expanded full-length Htt leads to increased neurotransmitter release. Resting intracellular $Ca^{2+}$ levels are increased in these flies compared to controls, suggesting a defect in $Ca^{2+}$ homeostasis. Intriguingly, these abnormalities occur before the cleavage and nuclear translocation of Htt protein was evident [127]. This study reveals an unexpected synaptic role for full-length expanded Htt protein in early stages of the disease. Interestingly, in another study, endogenous non-pathogenic Htt was found to protect against the aggregation of polyglutaminated Htt fragments [128]. These studies establish full-length Htt as a relevant and perhaps more physiological system for studying HD pathogenesis.

Another important finding from recent studies is that distinct pathogenic mechanisms underlie different polyQ diseases. For example, despite the isolation of cellular detoxification enzymes as modifiers of SCA-1 pathology, overexpression of SOD has no effect on the HD model [129]. As another example, specific histone deacetylase (HDAC)

and Sirtuins promote disease pathogenesis in a *Drosophila* HD model, with the inhibition of Rpd3 or Sir2 providing neuroprotective effects [130]. In contrast, HDAC6 induces autophagy and protects against neurode-generation in a *Drosophila* SBMA model [131]. These results suggest that different disease mechanism may underlie the different polyQ diseases, or that different HDACs exert distinct functions. Consistent with these findings, comparative analysis of a collection of genetic modifiers of expanded Ataxin-1-induced neurotoxicity in SCA-1 and HD models revealed that while some modifier genes function similarly in these models, others have model-specific effects. Surprisingly, certain modifier genes modify SCA1 and HD models in opposite directions [132]. Continued comparative analysis of other polyQ models will help uncover genes generally involved in polyQ-induced neurodegeneration as well as genes specific for each disease, which will provide excellent drug targets for the treatment of polyQ diseases. In this respect, the recently developed conditional *Drosophila* model of SCA7 will be particularly useful [133].

## Models for other neurodegenerative diseases

### Noncoding trinucleotide repeat diseases

In addition to the polyQ diseases where trinucleotide repeat expansion leads to toxic protein production, there are other diseases caused by trinucleotide repeat expansion within non-coding regions of mRNAs. Examples include SCA-8, −10, −12, Myotonic Dystrophy type 1 and 2, and Fragile × mental retardation (FMR). To understand how the rCGG repeats in the 5′-UTR of the FMR1 gene cause neurodegeneration in premutation carriers, 90 rCGG repeats, which is in-between the number of repeats found in patients (>200) and normal individuals (<60 repeats), was expressed in the fly eye. This 90 non-coding rCGG repeat was sufficient to cause degeneration of photoreceptors [134]. Although no mutant protein was produced from this repeat sequence, the degenerating neurons form neuronal inclusion bodies that are HSP70- and ubiquitin- positive. Further, overexpression of HSP70 was able to suppress rCGG repeats-induced neuronal death. It is possible that abnormal RNAs can cause neurodegeneration by sequestering rCGG repeat binding proteins from their normal function. In a biochemical purification, Pur $\alpha$ and hnRNP A2/B1 were identified as repeat binding proteins. Overex-pression of Pur $\alpha$ in *Drosophila* could suppress rCGG-mediated neurodegeneration in a dose-dependent manner. These findings support rCGG repeat sequestration of specific binding proteins as the pathogenic mechanism [135]. In another fly model of RNA-induced neurodegeneration, human SCA8 gene, which encodes a non-coding RNA with CUG expansion at the 3′ end, induced a late-onset progressive neurodegeneration phenotype when expressed in the fly eye [136]. A genetic modifier screen recovered four neuronally expressed RNA-binding proteins as modifiers of SCA8-induced neurodegeneration. These studies emphasized the importance of RNA binding proteins in noncoding trinucleotide repeat diseases. It remains to be tested whether the RNA-binding proteins identified from the SCA8 study will modify the toxicity induced by the non-coding rCGG expansion described above, and vice versa. This will reveal whether similar mechanisms are involved in noncoding trinucleotide repeat diseases.

### Prion disease and amyotrophic lateral sclerosis (ALS)

Two other prominent neurodegenerative diseases, amyotrophic lateral sclerosis (ALS) and prion diseases, have been successfully modeled in *Drosophila*. Transgenic flies heterologously expressing disease-associated (P101L) mouse PrP molecules in cholinergic neurons exhibited severe locomotor dysfunction and premature death as larvae and adults. Clinical abnormalities were accompanied by age-dependent accumulation of misfolded PrP molecules, intracellular PrP aggregates, and neuronal vacuoles. These transgenic flies thus exhibited several hallmark features of human Gerstmann–Ströussler–Scheinker syndrome [137].

To model amyotrophic lateral sclerosis (ALS), a motor neuron disease, mutant forms of the gene encoding copper, zinc-superoxide dismutase (SOD1) was ectopically expressed in motor neurons in *Drosophila*. This resulted in progressive climbing deficits accompanied by defective neural circuit electrophysiology, focal accumulation of human SOD1 protein in motor neurons, and a stress response in surrounding glia [138]. Unfortunately, toxicity was not associated with SOD1 oligomerization or neuronal loss in this model. It is possible that the absence of certain mammal-specific co-factors may have hindered full recapitulation of the disease symptoms in *Drosophila*.

Dominant mutations in vesicle-associated membrane protein-associated protein B (VAPB) cause ALS8, a slowly progressive motor neuron disease characterized by fasciculation, cramps, and postural tremor. A fly model of ALS8 was established using the corresponding mutation in *Drosophila* VAPB (dVAP33A). Neuronal expression of disease-associated VAP(P58S) causes phenotypes resembling VAP loss of function mutants and are opposite those of VAP overexpression, suggesting that VAPP58S may function in a dominant-negative fashion. VAP(P58S) formed aggregates and recruited wild type VAP into these aggregates. Further, the ALS8 mutation in dVAP33A was found to interferes with the BMP signaling pathway involved in synaptogenesis at the NMJ [139]. In another study, it was shown that the MSP domains of VAP proteins are cleaved and act as secreted ligands for Eph receptors. The P58S mutation led to a failure to secrete the MSP domain; instead, it induced ubiquitination, accumulation of inclusions in the endoplasmic reticulum, and an unfolded protein response [140]. This study offers new insight into pathogenic mechanisms of ALS.

### Spinal muscular atrophy (SMA) and Batten disease

Other rare neurodegenerative disease have also been successfully modeled in *Drosophila*. Mutations in the survival motor neuron (SMN) gene are associated with SMA, a recessive hereditary neurodegenerative disease characterized by early onset lethality coupled with motor neuron loss and skeletal muscle atrophy. The *Drosophila* SMN concentrates in the post-synaptic regions and is required for synaptogenesis at the NMJ. A genetic modifier screen resulted in the recovery of modifiers not previously known to be associated with this disease. Among the identified modifiers are components of the BMP signaling pathway previously shown to act in a retrograde signaling process to control synaptogenesis at the NMJ [141]. This study implicates synaptic dysfunction as a major cause of SMA.

Mutations in the gene *CLN3* are linked to the neurodegenerative disorder Juvenile Neuronal Ceroid Lipofuscinosis (JNCL), also called Batten Disease. *CLN3* encodes a conserved multi-spanning and hydrophobic transmembrane protein whose molecular function is still unclear. A genetic gain-of-function approach was used in *Drosophila* to identify functional pathways that involve *CLN3*. Previously unknown genetic interactions between *CLN3* and the Notch and JNK signalling pathways were revealed [142]. Further studies of this *Drosophila* model of JNCL will help understand the pathogenesis and offer strategies for the treatment of this devastating disease affecting young children.

## Concluding remarks

The development of highly relevant animal models of human diseases has ushered in a new era in neurodegenerative diseases research. *Drosophila* proves to be an excellent model organism for studying the normal biological function of genes linked to neurodegenerative diseases and how mutations in these genes leads to neuronal dysfunction and cell death. The true power of fly models lies in the ability to perform large-scale forward genetic screens to dissect the cellular signaling pathways involved in the disease process, without making assumptions about what kind of molecules or mechanisms are involved. Once novel

mechanisms or molecules are identified in the fly models, their relevance to mammalian system should be verified. Ongoing and future genetic studies should help answer some of the unresolved questions in the field, such as the molecular nature of the neurotoxic species for each disease type, the key neuronal functions being affected by the neurotoxic agents, and the determinants of the cell type-specific vulnerability underlying each disease. The genetic modifiers identified in flies may also help reveal disease susceptibility genes in humans and ultimately offer new therapeutic targets.

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## Abbreviations

| | |
|---|---|
| **4E-BP** | Eukaryotic initiation factor 4E-binding protein |
| **AD** | Alzheimer's disease |
| **ALS** | Amyotrophic lateral sclerosis |
| **APP** | Amyloid precursor protein |
| **BACE** | *β*-site APP-cleaving enzyme |
| **CEBP** | cAMP-responsive element-binding protein |
| **FMR** | Fragile × mental retardation |
| **HD** | Huntington's disease |
| **HDAC** | Histone deacetylase |
| **JNCL** | Juvenile neuronal ceroid lipofuscinosis |
| **LRRK2** | Leucine-rich repeat kinase 2 |
| **NFT** | Neurofibrillary tangles |
| **NMJ** | Neuromuscular junction |
| **Pael-R** | Parkin associated endothelin-like receptor |
| **PAR-1** | Partioning defective-1 |
| **PD** | Parkinson's disease |
| **PINK1** | Pten-induced kinase 1 |
| **RNAi** | RNA interference |
| **SBMA** | Spinobulbar muscular strophy |
| **SCA** | Spinocerebellar ataxia |
| **SMA** | Spinal muscular atrophy |
| **SOD1** | Copper, zinc-superoxide dismutase |
| **VAPB** | Vesicle-ass ociated membrane protein-associated protein B |
| **SMN** | Survival motor neuron |

## References


1. Bonini NM, Fortini ME. Human neurodegenerative disease modeling using Drosophila. Annu Rev Neurosci 2003;26:627–656.10.1146/annurev.neuro.26.041002.131425 [PubMed: 12704223]
2. Muqit MM, Feany MB. Modelling neurodegenerative diseases in Drosophila: a fruitful approach? Nat Rev Neurosci 2002;3:237–243.10.1038/nrn751 [PubMed: 11994755]
3. Driscoll M, Gerstbrein B. Dying for a cause: invertebrate genetics takes on human neurodegeneration. Nat Rev Genet 2003;4:181–194.10.1038/nrg1018 [PubMed: 12610523]
4. Lu B, Vogel H. Drosophila models of neurodegenerative diseases. Annu Rev Pathol 2008;4:315–342. [PubMed: 18842101]
5. Rogina B, Benzer S, Helfand SL. Drosophila drop-dead mutations accelerate the time course of age-related markers. Proc Natl Acad Sci USA 1997;94:6303–6306.10.1073/pnas.94. 12.6303 [PubMed: 9177212]
6. Kretzschmar D, Hasan G, Sharma S, Heisenberg M, Benzer S. The swiss cheese mutant causes glial hyperwrapping and brain degeneration in Drosophila. J Neurosci 1997;17:7425–7432. [PubMed: 9295388]
7. Akassoglou K, Malester B, Xu J, Tessarollo L, Rosenbluth J, Chao MV. Brain-specific deletion of neuropathy target esterase/swisscheese results in neurodegeneration. Proc Natl Acad Sci USA 2004;101:5075–5080.10.1073/pnas.0401030101 [PubMed: 15051870]
8. Jackson GR, Salecker I, Dong X, et al. Polyglutamine-expanded human huntingtin transgenes induce degeneration of Drosophila photoreceptor neurons. Neuron 1998;21:633–642.10.1016/S0896-6273(00)80573-5 [PubMed: 9768849]
9. Warrick JM, Paulson HL, Gray-Board GL, et al. Expanded polyglutamine protein forms nuclear inclusions and causes neural degeneration in Drosophila. Cell 1998;93:939–949.10.1016/S0092-8674(00)81200-3 [PubMed: 9635424]
10. Feany MB, Bender WW. A Drosophila model of Parkinson's disease. Nature 2000;404:394–398.10.1038/35006074 [PubMed: 10746727]
11. Fernandez-Funez P, Nino-Rosales ML, de Gouyon B, et al. Identification of genes that modify ataxin-1-induced neurodegeneration. Nature 2000;408:101–106.10.1038/35040584 [PubMed: 11081516]
12. Greene JC, Whitworth AJ, Kuo I, Andrews LA, Feany MB, Pallanck LJ. Mitochondrial pathology and apoptotic muscle degeneration in Drosophila parkin mutants. Proc Natl Acad Sci USA 2003;100:4078–4083.10.1073/pnas.0737556100 [PubMed: 12642658]
13. Pesah Y, Pham T, Burgess H, et al. Drosophila parkin mutants have decreased mass and cell size and increased sensitivity to oxygen radical stress. Development 2004;131:2183–2194.10.1242/dev.01095 [PubMed: 15073152]
14. Yang Y, Nishimura I, Imai Y, Takahashi R, Lu B. Parkin suppresses dopaminergic neuron-selective neurotoxicity induced by Pael-R in Drosophila. Neuron 2003;37:911–924.10.1016/S0896-6273(03)00143-0 [PubMed: 12670421]
15. Rong YS, Golic KG. Gene targeting by homologous recombination in Drosophila. Science 2000;288:2013–2018.10.1126/ science.288.5473.2013 [PubMed: 10856208]
16. Coulom H, Birman S. Chronic exposure to rotenone models sporadic Parkinson's disease in *Drosophila melanogaster*. J Neurosci 2004;24:10993–10998.10.1523/JNEUROSCI.2993-04.2004 [PubMed: 15574749]
17. Chaudhuri A, Bowling K, Funderburk C, et al. Interaction of genetic and environmental factors in a Drosophila parkinsonism model. J Neurosci 2007;27:2457–2467.10.1523/JNEUR OSCI.4239-06.2007 [PubMed: 17344383]
18. Dexter DT, Sian J, Rose S, et al. Indices of oxidative stress and mitochondrial function in individuals with incidental Lewy body disease. Ann Neurol 1994;35:38–44.10.1002/ana.410350107 [PubMed: 8285590]
19. Dunnett SB, Bjorklund A. Prospects for new restorative and neuroprotective treatments in Parkinson's disease. Nature 1999;399:A32–A39.10.1038/19899 [PubMed: 10392578]
20. Bertoli-Avella AM, Oostra BA, Heutink P. Chasing genes in Alzheimer's and Parkinson's disease. Hum Genet 2004;114(5):413–438. [PubMed: 14999561]

21. Dawson TM, Dawson VL. Molecular pathways of neurodegeneration in Parkinson's disease. Science 2003;302:819–822.10.1126/science.1087753 [PubMed: 14593166]
22. Polymeropoulos MH, Lavedan C, Leroy E, et al. Mutation in the alpha-synuclein gene identified in families with Parkinson's disease. Science 1997;276:2045–2047.10.1126/science. 276.5321.2045 [PubMed: 9197268]
23. Kitada T, Asakawa S, Hattori N, et al. Mutations in the parkin gene cause autosomal recessive juvenile parkinsonism. Nature 1998;392:605–608.10.1038/33416 [PubMed: 9560156]
24. Leroy E, Boyer R, Auburger G, et al. The ubiquitin pathway in Parkinson's disease. Nature 1998;395:451–452.10.1038/ 26652 [PubMed: 9774100]
25. Bonifati V, Rizzu P, Squitieri F, et al. DJ-1 (PARK7), a novel gene for autosomal recessive, early onset parkinsonism. Neurol Sci 2003;24:159–160.10.1007/s10072-003-0108-0 [PubMed: 14598065]
26. Valente EM, Abou-Sleiman PM, Caputo V, et al. Hereditary early-onset Parkinson's disease caused by mutations in PINK1. Science 2004;304:1158–1160.10.1126/science.1096284 [PubMed: 15087508]
27. Zimprich A, Biskup S, Leitner P, et al. Mutations in LRRK2 cause autosomal-dominant parkinsonism with pleomorphic pathology. Neuron 2004;44:601–607.10.1016/j.neuron. 2004.11.005 [PubMed: 15541309]
28. Paisan-Ruiz C, Jain S, Evans EW, et al. Cloning of the gene containing mutations that cause PARK8-linked Parkinson's disease. Neuron 2004;44:595–600.10.1016/j.neuron.2004. 10.023 [PubMed: 15541308]
29. Auluck PK, Chan HY, Trojanowski JQ, Lee VM, Bonini NM. Chaperone suppression of alpha-synuclein toxicity in a Drosophila model for Parkinson's disease. Science 2002;295:865–868.10.1126/science.1067389 [PubMed: 11823645]
30. Singleton AB, Farrer M, Johnson J, et al. Alpha-synuclein locus triplication causes Parkinson's disease. Science 2003;302:841.10.1126/science.1090278 [PubMed: 14593171]
31. Wu YR, Wang CK, Chen CM, et al. Analysis of heat-shock protein 70 gene polymorphisms and the risk of Parkinson's disease. Hum Genet 2004;114:236–241.10.1007/s00439-003-1050-1 [PubMed: 14605873]
32. Dong Z, Wolfer DP, Lipp HP, Bueler H. Hsp70 gene transfer by adeno-associated virus inhibits MPTP-induced nigrostriatal degeneration in the mouse model of Parkinson disease. Mol Ther 2005;11:80–88.10.1016/j.ymthe.2004.09.007 [PubMed: 15585408]
33. Fujiwara H, Hasegawa M, Dohmae N, et al. Alpha-synuclein is phosphorylated in synucleinopathy lesions. Nat Cell Biol 2002;4:160–164.10.1038/ncb841 [PubMed: 11813001]
34. Chen L, Feany MB. Alpha-synuclein phosphorylation controls neurotoxicity and inclusion formation in a Drosophila model of Parkinson disease. Nat Neurosci 2005;8:657–663.10.1038/nn1443 [PubMed: 15834418]
35. Periquet M, Fulga T, Myllykangas L, Schlossmacher MG, Feany MB. Aggregated alpha-synuclein mediates dopaminergic neurotoxicity in vivo. J Neurosci 2007;27:3338–3346.10.1523/JNEUROSCI.0285-07.2007 [PubMed: 17376994]
36. Outeiro TF, Kontopoulos E, Altmann SM, et al. Sirtuin 2 inhibitors rescue alpha-synuclein-mediated toxicity in models of Parkinson's disease. Science 2007;317:516–519.10.1126/science. 1143780 [PubMed: 17588900]
37. Botella JA, Bayersdorfer F, Schneuwly S. Superoxide dismutase overexpression protects dopaminergic neurons in a Drosophila model of Parkinson's disease. Neurobiol Dis 2008;30:65–73.10.1016/j.nbd.2007.11.013 [PubMed: 18243716]
38. Cooper AA, Gitler AD, Cashikar A, et al. Alpha-synuclein blocks ER-Golgi traffic and Rab1 rescues neuron loss in Parkinson's models. Science 2006;313:324–328.10.1126/science. 1129462 [PubMed: 16794039]
39. Scherzer CR, Jensen RV, Gullans SR, Feany MB. Gene expression changes presage neurodegeneration in a Drosophila model of Parkinson's disease. Hum Mol Genet 2003;12:2457–2466.10.1093/hmg/ddg265 [PubMed: 12915459]

40. Xun Z, Sowell RA, Kaufman TC, Clemmer DE. Quantitative proteomics of a presymptomatic A53T alpha-synuclein Drosophila model of Parkinson disease. Mol Cell Proteomics 2008;7:1191–1203.10.1074/mcp.M700467-MCP200 [PubMed: 18353766]
41. Cha GH, Kim S, Park J, et al. Parkin negatively regulates JNK pathway in the dopaminergic neurons of Drosophila. Proc Natl Acad Sci USA 2005;102:10345–10350.10.1073/pnas.0500346102 [PubMed: 16002472]
42. Whitworth AJ, Theodore DA, Greene JC, Benes H, Wes PD, Pallanck LJ. Increased glutathione S-transferase activity rescues dopaminergic neuron loss in a Drosophila model of Parkinson's disease. Proc Natl Acad Sci USA 2005;102:8024–8029.10.1073/pnas.0501078102 [PubMed: 15911761]
43. Imai Y, Soda M, Inoue H, Hattori N, Mizuno Y, Takahashi R. An unfolded putative transmembrane polypeptide, which can lead to endoplasmic reticulum stress, is a substrate of Parkin. Cell 2001;105:891–902.10.1016/S0092-8674(01)00407-X [PubMed: 11439185]
44. Kitao Y, Imai Y, Ozawa K, et al. Pael receptor induces death of dopaminergic neurons in the substantia nigra via endoplasmic reticulum stress and dopamine toxicity, which is enhanced under condition of parkin inactivation. Hum Mol Genet 2007;16:50–60.10.1093/hmg/ddl439 [PubMed: 17116640]
45. Sang TK, Chang HY, Lawless GM, et al. A Drosophila model of mutant human parkin-induced toxicity demonstrates selective loss of dopaminergic neurons and dependence on cellular dopamine. J Neurosci 2007;27:981–992.10.1523/JNE UROSCI.4810-06.2007 [PubMed: 17267552]
46. Wang C, Lu R, Ouyang X, et al. Drosophila overexpressing parkin R275 W mutant exhibits dopaminergic neuron degeneration and mitochondrial abnormalities. J Neurosci 2007;27:8563–8570.10.1523/JNEUROSCI.0218-07.2007 [PubMed: 17687034]
47. Clark IE, Dodson MW, Jiang C, et al. Drosophila pink1 is required for mitochondrial function and interacts genetically with parkin. Nature 2006;441:1162–1166.10.1038/nature04779 [PubMed: 16672981]
48. Park J, Lee SB, Lee S, et al. Mitochondrial dysfunction in Drosophila PINK1 mutants is complemented by parkin. Nature 2006;441:1157–1161.10.1038/nature04788 [PubMed: 16672980]
49. Yang Y, Gehrke S, Imai Y, et al. Mitochondrial pathology and muscle and dopaminergic neuron degeneration caused by inactivation of Drosophila Pink1 is rescued by Parkin. Proc Natl Acad Sci USA 2006;103:10793–10798.10.1073/pnas.0602493103 [PubMed: 16818890]
50. Wang D, Qian L, Xiong H, et al. Antioxidants protect PINK1-dependent dopaminergic neurons in Drosophila. Proc Natl Acad Sci USA 2006;103:13520–13525.10.1073/pnas.0604661103 [PubMed: 16938835]
51. Kim Y, Park J, Kim S, et al. PINK1 controls mitochondrial localization of Parkin through direct phosphorylation. Biochem Biophys Res Commun 2008;377:975–980.10.1016/j.bbrc.2008.10.104 [PubMed: 18957282]
52. Yang Y, Ouyang Y, Yang L, et al. Pink1 regulates mitochondrial dynamics through interaction with the fission/fusion machinery. Proc Natl Acad Sci USA 2008;105:7070–7075.10.1073/pnas.0711845105 [PubMed: 18443288]
53. Poole AC, Thomas RE, Andrews LA, McBride HM, Whitworth AJ, Pallanck LJ. The PINK1/Parkin pathway regulates mitochondrial morphology. Proc Natl Acad Sci USA 2008;105:1638–1643.10.1073/pnas.0709336105 [PubMed: 18230723]
54. Deng H, Dodson MW, Huang H, Guo M. The Parkinson's disease genes pink1 and parkin promote mitochondrial fission and/or inhibit fusion in Drosophila. Proc Natl Acad Sci USA 2008;105:14503–14508.10.1073/pnas.0803998105 [PubMed: 18799731]
55. Park J, Lee G, Chung J. The PINK1-Parkin pathway is involved in the regulation of mitochondrial remodeling process. Biochem Biophys Res Commun 2009;378:518–523.10.1016/j.bbrc.2008.11.086 [PubMed: 19056353]
56. Chan DC. Mitochondria: dynamic organelles in disease, aging, and development. Cell 2006;125:1241–1252.10.1016/j.cell.2006.06.010 [PubMed: 16814712]

57. Li Z, Okamoto K, Hayashi Y, Sheng M. The importance of dendritic mitochondria in the morphogenesis and plasticity of spines and synapses. Cell 2004;119:873–887.10.1016/j.cell.2004.11.003 [PubMed: 15607982]
58. Bossy-Wetzel E, Barsoum MJ, Godzik A, Schwarzenbacher R, Lipton SA. Mitochondrial fission in apoptosis, neurodegeneration and aging. Curr Opin Cell Biol 2003;15:706–716.10.1016/j.ceb.2003.10.015 [PubMed: 14644195]
59. Petrucelli L, O'Farrell C, Lockhart PJ, et al. Parkin protects against the toxicity associated with mutant alpha-synuclein: proteasome dysfunction selectively affects catecholaminergic neurons. Neuron 2002;36:1007–1019.10.1016/S0896-6273(02) 01125-X [PubMed: 12495618]
60. Sacktor B. Biochemical adaptations for flight in the insect. Biochem Soc Symp 1976;41:111–131. [PubMed: 788715]
61. Park SS, Schulz EM, Lee D. Disruption of dopamine homeostasis underlies selective neurodegeneration mediated by alpha-synuclein. Eur J Neurosci 2007;26:3104–3112.10.1111/j.1460-9568.2007.05929.x [PubMed: 18005066]
62. Meulener M, Whitworth AJ, Armstrong-Gold CE, et al. Drosophila DJ-1 mutants are selectively sensitive to environmental toxins associated with Parkinson's disease. Curr Biol 2005;15:1572–1577.10.1016/j.cub.2005.07.064 [PubMed: 16139213]
63. Menzies FM, Yenisetti SC, Min KT. Roles of Drosophila DJ-1 in survival of dopaminergic neurons and oxidative stress. Curr Biol 2005;15:1578–1582.10.1016/j.cub.2005.07.036 [PubMed: 16139214]
64. Yang Y, Gehrke S, Haque ME, et al. Inactivation of Drosophila DJ-1 leads to impairments of oxidative stress response and phosphatidylinositol 3-kinase/Akt signaling. Proc Natl Acad Sci USA 2005;102:13670–13675.10.1073/pnas.0504 610102 [PubMed: 16155123]
65. Kim RH, Peters M, Jang Y, et al. DJ-1, a novel regulator of the tumor suppressor PTEN. Cancer Cell 2005;7:263–273.10.1016/j.ccr.2005.02.010 [PubMed: 15766664]
66. van der Brug MP, Blackinton J, Chandran J, et al. RNA binding activity of the recessive parkinsonism protein DJ-1 supports involvement in multiple cellular pathways. Proc Natl Acad Sci USA 2008;105:10244–10249.10.1073/pnas.070851 8105 [PubMed: 18626009]
67. Lee SB, Kim W, Lee S, Chung J. Loss of LRRK2/PARK8 induces degeneration of dopaminergic neurons in Drosophila. Biochem Biophys Res Commun 2007;358:534–539.10.1016/j. bbrc.2007.04.156 [PubMed: 17498648]
68. Liu Z, Wang X, Yu Y, et al. A Drosophila model for LRRK2-linked parkinsonism. Proc Natl Acad Sci USA 2008;105:2693–2698.10.1073/pnas.0708452105 [PubMed: 18258746]
69. Imai Y, Gehrke S, Wang HQ, et al. Phosphorylation of 4E-BP by LRRK2 affects the maintenance of dopaminergic neurons in Drosophila. EMBO J 2008;27:2432–2443.10.1038/emboj. 2008.163 [PubMed: 18701920]
70. De Strooper B. Aph-1, Pen-2, and nicastrin with presenilin generate an active γ-secretase complex. Neuron 2003;38:9–12.10.1016/S0896-6273(03)00205-8 [PubMed: 12691659]
71. Cleveland DW, Hwo SY, Kirschner MW. Purification of tau, a microtubule-associated protein that induces assembly of microtubules from purified tubulin. J Mol Biol 1977;116:207–225.10.1016/0022-2836(77)90213-3 [PubMed: 599557]
72. Grundke-Iqbal I, Iqbal K, Tung YC, Quinlan M, Wisniewski HM, Binder LI. Abnormal phosphorylation of the microtubule-associated protein tau (tau) in Alzheimer cyto-skeletal pathology. Proc Natl Acad Sci USA 1986;83:4913–4917.10.1073/pnas.83.13.4913 [PubMed: 3088567]
73. Greenberg SG, Davies P. A preparation of Alzheimer paired helical filaments that displays distinct tau proteins by polyacrylamide gel electrophoresis. Proc Natl Acad Sci USA 1990;87:5827–5831.10.1073/pnas.87.15.5827 [PubMed: 2116006]
74. Lee VM, Balin BJ, Otvos L Jr, Trojanowski JQ. A68: a major subunit of paired helical filaments and derivatized forms of normal Tau. Science 1991;251:675–678.10.1126/science. 1899488 [PubMed: 1899488]
75. Foster NL, Wilhelmsen K, Sima AA, Jones MZ, D'Amato CJ, Gilman S. Frontotemporal dementia and parkinsonism linked to chromosome 17: a consensus conference. Conference participants. Ann Neurol 1997;41:706–715.10.1002/ana.41041 0606 [PubMed: 9189031]

76. Hutton M, Lendon CL, Rizzu P, et al. Association of missense and 5′-splicesite mutations in tau with the inherited dementia FTDP-17. Nature 1998;393:702–705.10.1038/31508 [PubMed: 9641683]
77. Poorkaj P, Bird TD, Wijsman E, et al. Tau is a candidate gene for chromosome 17 frontotemporal dementia. Ann Neurol 1998;43:815–825.10.1002/ana.410430617 [PubMed: 9629852]
78. Spillantini MG, Murrell JR, Goedert M, Farlow MR, Klug A, Ghetti B. Mutation in the tau gene in familial multiple system tauopathy with presenile dementia. Proc Natl Acad Sci USA 1998;95:7737–7741.10.1073/pnas.95.13.7737 [PubMed: 9636220]
79. Luo L, Tully T, White K. Human amyloid precursor protein ameliorates behavioral deficit of flies deleted for Appl gene. Neuron 1992;9:595–605.10.1016/0896-6273(92)90024-8 [PubMed: 1389179]
80. Carmine-Simmen K, Proctor T, Tschape J, et al. Neurotoxic effects induced by the Drosophila amyloid-beta peptide suggest a conserved toxic function. Neurobiol Dis 2008;33(2):274–281. [PubMed: 19049874]
81. Finelli A, Kelkar A, Song HJ, Yang H, Konsolaki M. A model for studying Alzheimer's Abeta42-induced toxicity in *Drosophila melanogaster*. Mol Cell Neurosci 2004;26:365–375.10.1016/j.mcn.2004.03.001 [PubMed: 15234342]
82. Iijima K, Liu HP, Chiang AS, Hearn SA, Konsolaki M, Zhong Y. Dissecting the pathological effects of human Abeta40 and Abeta42 in Drosophila: a potential model for Alzheimer's disease. Proc Natl Acad Sci USA 2004;101:6623–6628.10.1073/pnas.0400895101 [PubMed: 15069204]
83. Stokin GB, Almenar-Queralt A, Gunawardena S, et al. Amyloid precursor protein-induced axonopathies are independent of amyloid-beta peptides. Hum Mol Genet 2008;17:3474–3486.10.1093/hmg/ddn240 [PubMed: 18694898]
84. Tan L, Schedl P, Song HJ, Garza D, Konsolaki M. The Toll–>NFkappaB signaling pathway mediates the neuropathological effects of the human Alzheimer's Abeta42 polypeptide in Drosophila. PLoS One 2008;3:e3966.10.1371/journal.pone. 0003966 [PubMed: 19088848]
85. Iijima-Ando K, Hearn SA, Granger L, et al. Overexpression of neprilysin reduces alzheimer amyloid-beta42 (Abeta42)-induced neuron loss and intraneuronal Abeta42 deposits but causes a reduction in cAMP-responsive element-binding protein-mediated transcription, age-dependent axon pathology, and premature death in Drosophila. J Biol Chem 2008;283:19066–19076.10.1074/jbc.M710509200 [PubMed: 18463098]
86. Williams DW, Tyrer M, Shepherd D. Tau and tau reporters disrupt central projections of sensory neurons in Drosophila. J Comp Neurol 2000;428:630–640.10.1002/1096-9861(20001225)428:4≤630::AID-CNE4≥3.0.CO;2-X [PubMed: 11077417]
87. Wittmann CW, Wszolek MF, Shulman JM, et al. Tauopathy in Drosophila: neurodegeneration without neurofibrillary tangles. Science 2001;293:711–714.10.1126/science.1062382 [PubMed: 11408621]
88. Jackson GR, Wiedau-Pazos M, Sang TK, et al. Human wild-type tau interacts with wingless pathway components and produces neurofibrillary pathology in Drosophila. Neuron 2002;34:509–519.10.1016/S0896-6273(02)00706-7 [PubMed: 12062036]
89. Mershin A, Pavlopoulos E, Fitch O, Braden BC, Nanopoulos DV, Skoulakis EM. Learning and memory deficits upon TAU accumulation in Drosophila mushroom body neurons. Learn Mem 2004;11:277–287.10.1101/lm.70804 [PubMed: 15169857]
90. Lee VM, Goedert M, Trojanowski JQ. Neurodegenerative tauopathies. Annu Rev Neurosci 2001;24:1121–1159.10.1146/annurev.neuro.24.1.1121 [PubMed: 11520930]
91. Nishimura I, Yang Y, Lu B. PAR-1 kinase plays an initiator role in a temporally ordered phosphorylation process that confers tau toxicity in Drosophila. Cell 2004;116:671–682.10.1016/S0092-8674(04)00170-9 [PubMed: 15006350]
92. Guo S, Kemphues KJ. par-1, a gene required for establishing polarity in *C. elegans* embryos, encodes a putative Ser/Thr kinase that is asymmetrically distributed. Cell 1995;81:611–620.10.1016/0092-8674(95)90082-9 [PubMed: 7758115]
93. Drewes G, Ebneth A, Preuss U, Mandelkow EM, Mandelkow E. MARK, a novel family of protein kinases that phosphorylate microtubule-associated proteins and trigger microtubule disruption. Cell 1997;89:297–308.10.1016/S0092-8674(00) 80208-1 [PubMed: 9108484]

94. Bohm H, Brinkmann V, Drab M, Henske A, Kurzchalia TV. Mammalian homologues of *C. elegans* PAR-1 are asymmetrically localized in epithelial cells and may influence their polarity. Curr Biol 1997;7:603–606.10.1016/S0960-9822 (06)00260-0 [PubMed: 9259552]

95. Biernat J, Wu YZ, Timm T, et al. Protein kinase MARK/PAR-1 is required for neurite outgrowth and establishment of neuronal polarity. Mol Biol Cell 2002;13:4013–4028.10.1091/mbc.02-03-0046 [PubMed: 12429843]

96. Shulman JM, Feany MB. Genetic modifiers of tauopathy in Drosophila. Genetics 2003;165:1233–1242. [PubMed: 14668378]

97. Hanger DP, Betts JC, Loviny TL, Blackstock WP, Anderton BH. New phosphorylation sites identified in hyperphosphorylated tau (paired helical filament-tau) from Alzheimer's disease brain using nanoelectrospray mass spectrometry. J Neurochem 1998;71:2465–2476. [PubMed: 9832145]

98. Chatterjee S, Sang TK, Lawless GM, Jackson GR. Dissociation of tau toxicity and phosphorylation: role of GSK-3beta, MARK and Cdk5 in a Drosophila model. Hum Mol Genet 2009;18:164–177.10.1093/hmg/ddn326 [PubMed: 18930955]

99. Steinhilb ML, Dias-Santagata D, Fulga TA, Felch DL, Feany MB. Tau phosphorylation sites work in concert to promote neurotoxicity in vivo. Mol Biol Cell 2007;18:5060–5068.10.1091/mbc.E07-04-0327 [PubMed: 17928404]

100. Karsten SL, Sang TK, Gehman LT, et al. A genomic screen for modifiers of tauopathy identifies puromycin-sensitive aminopeptidase as an inhibitor of tau-induced neurodegeneration. Neuron 2006;51:549–560.10.1016/j.neuron.2006.07.019 [PubMed: 16950154]

101. Wang JW, Imai Y, Lu B. Activation of PAR-1 kinase and stimulation of tau phosphorylation by diverse signals require the tumor suppressor protein LKB1. J Neurosci 2007;27:574–581.10.1523/JNEUROSCI.5094-06.2007 [PubMed: 17234589]

102. Selkoe DJ. Alzheimer's disease is a synaptic failure. Science 2002;298:789–791.10.1126/science.1074069 [PubMed: 12399581]

103. Wang P, Yang G, Mosier DR, et al. Defective neuromuscular synapses in mice lacking amyloid precursor protein (APP) and APP-Like protein 2. J Neurosci 2005;25:1219–1225.10.1523/JNEUROSCI.4660-04.2005 [PubMed: 15689559]

104. Ashe KH. Synaptic structure and function in transgenic APP mice. Ann N Y Acad Sci 2000;924:39–41. [PubMed: 11193800]

105. Roberson ED, Scearce-Levie K, Palop JJ, et al. Reducing endogenous tau ameliorates amyloid beta-induced deficits in an Alzheimer's disease mouse model. Science 2007;316:750–754.10.1126/science.1141736 [PubMed: 17478722]

106. Chee FC, Mudher A, Cuttle MF, et al. Overexpression of tau results in defective synaptic transmission in Drosophila neuromuscular junctions. Neurobiol Dis 2005;20:918–928.10.1016/j.nbd.2005.05.029 [PubMed: 16023860]

107. Yoshiyama Y, Higuchi M, Zhang B, et al. Synapse loss and microglial activation precede tangles in a P301S tauopathy mouse model. Neuron 2007;53:337–351.10.1016/j.neuron.2007.01.010 [PubMed: 17270732]

108. Blard O, Feuillette S, Bou J, et al. Cytoskeleton proteins are modulators of mutant tau-induced neurodegeneration in Drosophila. Hum Mol Genet 2007;16:555–566.10.1093/hmg/ddm011 [PubMed: 17309878]

109. Zhang Y, Guo H, Kwan H, Wang JW, Kosek J, Lu B. par-1 kinase phosphorylates dlg and regulates its postsynaptic targeting at the Drosophila neuromuscular junction. Neuron 2007;53:201–215.10.1016/j.neuron.2006.12.016 [PubMed: 17224403]

110. Almeida CG, Tampellini D, Takahashi RH, et al. Beta-amyloid accumulation in APP mutant neurons reduces PSD-95 and GluR1 in synapses. Neurobiol Dis 2005;20:187–198.10.1016/j.nbd.2005.02.008 [PubMed: 16242627]

111. Gylys KH, Fein JA, Yang F, Wiley DJ, Miller CA, Cole GM. Synaptic changes in Alzheimer's disease: increased amyloid-beta and gliosis in surviving terminals is accompanied by decreased PSD-95 fluorescence. Am J Pathol 2004;165:1809–1817. [PubMed: 15509549]

112. Roselli F, Tirard M, Lu J, et al. Soluble beta-amyloid1–40 induces NMDA-dependent degradation of postsynaptic density-95 at glutamatergic synapses. J Neurosci 2005;25:11061–11070.10.1523/JNEUROSCI.3034-05.2005 [PubMed: 16319306]

113. Knight D, Iliadi K, Charlton MP, Atwood HL, Boulianne GL. Presynaptic plasticity and associative learning are impaired in a Drosophila presenilin null mutant. Dev Neurobiol 2007;67:1598–1613.10.1002/dneu.20532 [PubMed: 17562530]

114. Li A, Xie Z, Dong Y, McKay KM, McKee ML, Tanzi RE. Isolation and characterization of the Drosophila ubiquilin ortholog dUbqln: in vivo interaction with early-onset Alzheimer disease genes. Hum Mol Genet 2007;16:2626–2639.10.1093/hmg/ddm219 [PubMed: 17704509]

115. Gusella JF, MacDonald ME. Molecular genetics: unmasking polyglutamine triggers in neurodegenerative disease. Nat Rev Neurosci 2000;1:109–115.10.1038/35039051 [PubMed: 11252773]

116. Kretzschmar D, Tschape J, Bettencourt Da Cruz A, et al. Glial and neuronal expression of polyglutamine proteins induce behavioral changes and aggregate formation in Drosophila. Glia 2005;49:59–72.10.1002/glia.20098 [PubMed: 15390099]

117. Lievens JC, Rival T, Iche M, Chneiweiss H, Birman S. Expanded polyglutamine peptides disrupt EGF receptor signaling and glutamate transporter expression in Drosophila. Hum Mol Genet 2005;14:713–724.10.1093/hmg/ddi067 [PubMed: 15677486]

118. Lievens JC, Iche M, Laval M, Faivre-Sarrailh C, Birman S. AKT-sensitive or insensitive pathways of toxicity in glial cells and neurons in Drosophila models of Huntington's disease. Hum Mol Genet 2008;17:882–894.10.1093/hmg/ddm360 [PubMed: 18065778]

119. Bilen J, Bonini NM. Genome-wide screen for modifiers of ataxin-3 neurodegeneration in Drosophila. PLoS Genet 2007;3:1950–1964.10.1371/journal.pgen.0030177 [PubMed: 17953484]

120. Li LB, Yu Z, Teng X, Bonini NM. RNA toxicity is a component of ataxin-3 degeneration in Drosophila. Nature 2008;453:1107–1111.10.1038/nature06909 [PubMed: 18449188]

121. Bilen J, Liu N, Burnett BG, Pittman RN, Bonini NM. MicroRNA pathways modulate polyglutamine-induced neurodegeneration. Mol Cell 2006;24:157–163.10.1016/j.molcel.2006.07.030 [PubMed: 17018300]

122. Chen HK, Fernandez-Funez P, Acevedo SF, et al. Interaction of Akt-phosphorylated ataxin-1 with 14-3-3 mediates neurodegeneration in spinocerebellar ataxia type 1. Cell 2003;113:457–468.10.1016/S0092-8674(03)00349-0 [PubMed: 12757707]

123. Lim J, Crespo-Barreto J, Jafar-Nejad P, et al. Opposing effects of polyglutamine expansion on native protein complexes contribute to SCA1. Nature 2008;452:713–718.10.1038/nature 06731 [PubMed: 18337722]

124. Lessing D, Bonini NM. Polyglutamine genes interact to modulate the severity and progression of neurodegeneration in Drosophila. PLoS Biol 2008;6:e29.10.1371/journal.pbio.0060029 [PubMed: 18271626]

125. Al-Ramahi I, Perez AM, Lim J, et al. dAtaxin-2 mediates expanded Ataxin-1-induced neurodegeneration in a Drosophila model of SCA1. PLoS Genet 2007;3:e234.10.1371/journal.pgen. 0030234 [PubMed: 18166084]

126. Jung J, Bonini N. CREB-binding protein modulates repeat instability in a Drosophila model for polyQ disease. Science 2007;315:1857–1859.10.1126/science.1139517 [PubMed: 17332375]

127. Romero E, Cha GH, Verstreken P, et al. Suppression of neurodegeneration and increased neurotransmission caused by expanded full-length huntingtin accumulating in the cytoplasm. Neuron 2008;57:27–40.10.1016/j.neuron.2007.11.025 [PubMed: 18184562]

128. Mugat B, Parmentier ML, Bonneaud N, Chan HY, Maschat F. Protective role of engrailed in a Drosophila model of Huntington's disease. Hum Mol Genet 2008;17:3601–3616.10.1093/hmg/ddn255 [PubMed: 18718937]

129. Bahadorani S, Hilliker AJ. Antioxidants cannot suppress the lethal phenotype of a *Drosophila melanogaster* model of Huntington's disease. Genome 2008;51:392–395.10.1139/G08-012 [PubMed: 18438443]

130. Pallos J, Bodai L, Lukacsovich T, et al. Inhibition of specific HDACs and sirtuins suppresses pathogenesis in a Drosophila model of Huntington's disease. Hum Mol Genet 2008;17:3767–3775.10.1093/hmg/ddn273 [PubMed: 18762557]

131. Pandey UB, Nie Z, Batlevi Y, et al. HDAC6 rescues neurodegeneration and provides an essential link between autophagy and the UPS. Nature 2007;447:859–863.10.1038/nature05853 [PubMed: 17568747]

132. Branco J, Al-Ramahi I, Ukani L, et al. Comparative analysis of genetic modifiers in Drosophila points to common and distinct mechanisms of pathogenesis among polyglutamine diseases. Hum Mol Genet 2008;17:376–390.10.1093/hmg/ddm315 [PubMed: 17984172]

133. Latouche M, Lasbleiz C, Martin E, et al. A conditional pan-neuronal Drosophila model of spinocerebellar ataxia 7 with a reversible adult phenotype suitable for identifying modifier genes. J Neurosci 2007;27:2483–2492.10.1523/JNEUROSCI. 5453-06.2007 [PubMed: 17344386]

134. Jin P, Zarnescu DC, Zhang F, et al. RNA-mediated neurodegeneration caused by the fragile × premutation rCGG repeats in Drosophila. Neuron 2003;39:739–747.10.1016/S0896-6273(03)00533-6 [PubMed: 12948442]

135. Jin P, Duan R, Qurashi A, et al. Pur alpha binds to rCGG repeats and modulates repeat-mediated neurodegeneration in a Drosophila model of fragile × tremor/ataxia syndrome. Neuron 2007;55:556–564.10.1016/j.neuron.2007.07.020 [PubMed: 17698009]

136. Mutsuddi M, Marshall CM, Benzow KA, Koob MD, Rebay I. The spinocerebellar ataxia 8 noncoding RNA causes neurodegeneration and associates with staufen in Drosophila. Curr Biol 2004;14:302–308. [PubMed: 14972680]

137. Gavin BA, Dolph MJ, Deleault NR, et al. Accelerated accumulation of misfolded prion protein and spongiform degeneration in a Drosophila model of Gerstmann-Straussler-Scheinker syndrome. J Neurosci 2006;26:12408–12414.10.1523/JNEUROSCI.3372-06.2006 [PubMed: 17135402]

138. Watson MR, Lagow RD, Xu K, Zhang B, Bonini NM. A drosophila model for amyotrophic lateral sclerosis reveals motor neuron damage by human SOD1. J Biol Chem 2008;283:24972–24981.10.1074/jbc.M804817200 [PubMed: 18596033]

139. Ratnaparkhi A, Lawless GM, Schweizer FE, Golshani P, Jackson GR. A Drosophila model of ALS: human ALS-associated mutation in VAP33A suggests a dominant negative mechanism. PLoS One 2008;3:e2334.10.1371/journal.pone. 0002334 [PubMed: 18523548]

140. Tsuda H, Han SM, Yang Y, et al. The amyotrophic lateral sclerosis 8 protein VAPB is cleaved, secreted, and acts as a ligand for Eph receptors. Cell 2008;133:963–977.10.1016/j.cell.2008.04.039 [PubMed: 18555774]

141. Chang HC, Dimlich DN, Yokokura T, et al. Modeling spinal muscular atrophy in Drosophila. PLoS One 2008;3:e3209.10.1371/journal.pone.0003209 [PubMed: 18791638]

142. Tuxworth RI, Vivancos V, O'Hare MB, Tear G. Interactions between the juvenile Batten disease gene, CLN3, and the Notch and JNK signalling pathways. Hum Mol Genet 2008;18(4):667–678. [PubMed: 19028667]