Journal of Genetic Syndromes & Gene Therapy

McIntosh et al., J Genet Syndr Gene Ther 2017, 8:2
DOI: 10.4172/2157-7412.1000319

Review Article Open Access

Polyglutamine ataxias: From Clinical and Molecular Features to Current Therapeutic Strategies

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Abstract

Spinocerebellar ataxias are a large group of heterogeneous diseases that all involve selective neuronal degeneration and accompanied cerebellar ataxia. These diseases can be further broken down into discrete groups according to their underlying molecular genetic cause. The most common are the polyglutamine ataxias, of which there are six; Spinocerebellar ataxia type 1, 2, 3, 6, 7 and 17. These diseases are characterised by a pathological expanded cytosine–adenine–guanine (CAG) repeat sequence, in the protein coding region of a given gene. Common clinical features include lack of coordination and gait ataxia, speech and swallowing difficulties, as well as impaired hand and motor functions. The polyglutamine spinocerebellar ataxias are typically late onset diseases that are progressive in nature and often lead to premature death, for which there is currently no known cure or effective treatment strategy. Although caused by the same molecular mechanism, the causative gene and associated protein differ for each disease. The exact mechanism by which disease pathogenesis is caused remains elusive. However, the variable $(CAG)_n$ repeats are codons that may be translated to an expanded glutamine tract, leading to conformational changes in the protein, giving it a toxic gain of function. Several pathogenic pathways have been implicated in polyglutamine spinocerebellar ataxia diseases, such as the hallmark feature of neuronal nuclear inclusions, protein misfolding and aggregation, as well as transcriptional dysregulation. These pathways are attractive avenues for potential therapeutic interventions, as the potential to treat more than one disease exists. Research is ongoing, and several promising therapies are currently underway in an attempt to provide relief for this devastating class of diseases..

Keywords: Spinocerebellar ataxia; Triplet repeat expansion; Polyglutamine diseases; Genetic therapies

Introduction

The spinocerebellar ataxias (SCAs) are a large group of typically late onset, progressive disorders characterised by neurodegeneration and other pathologically heterogeneous clinical features [1,2]. These diseases are often grouped into categories based on their causative mutation, of which there are three:

1. Non-coding repeat ataxias, where the repeat expansion is located outside the protein coding region for the gene of interest.
2. Ataxias where disease is caused by an 'orthodox' mutation, such as missense or splice site mutation.
3. Polyglutamine (polyQ) ataxias where the disease is characterized by an expanded cytosine–adenine–guanine (CAG) repeat located in the coding region of the respective gene [3].

As the triplet CAG encodes for the amino acid glutamine, this leads to an elongated glutamine tract in the translated protein resulting in conformational changes that are thought to cause several pathogenic mechanisms. Although the presence of an expansion does not necessarily correlate to phenotypic modifications, once a threshold for a specific gene is met, this tends to lead to disease and pathogenesis. Due to the nature of these mutations, the pathogenic severity and penetrance is typically determined by the size of the expansion, where there is a common trend: the larger the expansion, the more severe the pathogenesis and/or the earlier the onset [4]. These diseases tend to follow what is known as 'genetic anticipation', whereby the expansion size increases with each successive generation [5,6]. It should be noted that the genetic anticipation in most SCA diseases is typically more likely to be passed down from the paternal gene rather than the maternal. Rare cases have been reported where individuals possess two mutant alleles, leading to more severe symptoms than individuals with just one mutant allele. However, mutations can arise *de novo*, through errors in DNA replication such that two genetically healthy parents can give rise to an affected child.

There are currently six characterised polyQ SCAs (1, 2, 3, 6, 7 and 17) (Table 1), and together with three other diseases, namely Huntington's disease, spinal and bulbar muscular atrophy and dentatorubropallidoluysian atrophy (DRPLA), form a larger category of polyQ diseases [7-10]. There is little relief for individuals suffering from polyQ SCA disorders, with symptomatic treatments the only available option. Long term pharmacological treatment, although admirable, tends to fall short in an effective management strategy, as unwanted complications and low drug efficacy still exist. In addition, none of these diseases have any treatments that slow the progression of the disease; leaving affected individuals with the daunting reality that time may be a severe limiting factor. Several experimental approaches are currently being assessed to overcome these difficulties. This review will focus on the clinical and molecular features of polyQ SCA diseases (which will be referred to as SCA diseases), as well as highlight several promising and emerging therapeutic strategies. We will begin by describing the molecular events leading to a CAG expansion, followed by the SCA diseases in general, before delving into greater depth of all six diseases. The review will then focus on potential therapeutic strategies that target common molecular and pathological features of these diseases.

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Received July 25, 2017; **Accepted** August 04, 2017; **Published** August 11, 2017

Citation: McIntosh CS, Aung-Htut MT, Fletcher S, Wilton SD (2017) Polyglutamine ataxias: From Clinical and Molecular Features to Current Therapeutic Strategies. J Genet Syndr Gene Ther 8: 319. doi: 10.4172/2157-7412.1000319

Disease Name*	ADCA classification**	Age of onset	Age range (years)	Additional characteristic features, other than cerebellar ataxia
SCA1	Type I	3rd-4th decade	Childhood to 60+	Purkinje cell loss, faster progression of disease
SCA2	Type I	3rd-4th decade	Childhood to 60+	Slow saccadic ocular movements, peripheral neuropathy
SCA3	Type I	4th decade	10 to 70+	Pyramidal and extra pyramidal signs, ocular movement abnormalities
SCA6	Type III	5th-6th decade	18 to 70+	Slower progression of the disease, ocular abnormalities
SCA7	Type II	3rd-4th decade	Infancy to 60	Visual loss with accompanying retinal pathology
SCA17	Type I	4th decade	Infancy to 55	Psychiatric abnormalities, dementia, chorea

*SCA: Spinocerebellar Ataxia
**ADCA: Autosomal Dominant Cerebellar Ataxia

Table 1: Clinical and age related features of the six polyQ spinocerebellar ataxias.

Molecular Genetics behind CAG Expansion

Microsatellites are commonly found repetitive arrangements of DNA, typically consisting of 1-6 nucleotide repeated sequences. These repetitive sequences along with short interspaced nuclear elements and long interspaced nuclear elements constitute approximately 30% of the 2.91 billion base-pair human genome [11,12]. Although the repeated sequence may vary in nucleotide length, trinucleotide repeats (TNRs) are the most common microsatellite found in coding regions [13]. Variations in the copy number of these TNRs will not affect the reading frame, but simply allow for variation in the translated protein. In most species, both plant and mammalian, the variation in these repeating sequences is what creates and drives evolutionary diversity between and in species subsets [14-16]. However, in some instances, these normal variations lead to disease through a larger than normal expansion of the copy number. All SCA diseases are caused by mutations from conception, whether it is from a pre-mutation or mutant parent allele or arises *de novo* from an error in gamete production. Additionally, somatic mosaicisms have been reported for several SCA diseases, where several studies have found cases of considerable genetic homogeneity within regions of the brain [17,18]. SCA1 and SCA3 patients were found to have a varying pathogenic CAG length in all but 1 of the 20 regions of the brain examined [17]. Due to their repetitive nature, the CAG repeats are unstable and often prone to mutations that are typically attributed to either slip-strand mispairing during DNA replication and/or errors in mismatch repair [19-21]. These two processes are often thought to be inter-linked, with slip-strand mispairing occurring concurrently, where mismatch repair does not effectively correct the hairpin instability. SCA expansions are typically relatively small and less than 400 TNRs, while the expansion noted in myotonic dystrophy type 1 can be measured in the thousands [22].

Slip-strand mispairing

TNR and other repetitive sequences are prone to the formation of DNA secondary structures. Slip-strand mispairing is an error that occurs when DNA is incorrectly replicated, in particular where the template strand and the synthesised strand become temporarily mis-aligned and result in the mispairing of the complementary pairs [23,24]. As the DNA polymerase β encounters the CAG repeating sequence (or any repeating sequencing for that matter), it momentarily pauses, however the helicase is left unaffected. In an attempt to avoid uncoupling of the helicase and polymerase, a misalignment is caused on the newly synthesised strand. During this separation, single strand loops of CAG repeats are created and the repetitive characteristics of the sequences allow the strands to displace (or slip) by a variable number of TNR (Figure 1) [25,26]. Insertions/expansion are caused when the loop is formed on the synthesised strand, also termed backward slippage, while deletions/concretion of the repeat sequence is caused when the loop forms on the template strand, also termed forward slippage.

In non-dividing cells such as spermatogonia, slip-strand mispairing is typically responsible for small expansions of a few TNRs that are often too small to fall within disease range. It is theoretically possible for numerous slippage events to occur, leading to a larger expansion. It is therefore more likely that large expansions are the result of repair dependant mechanisms in non-dividing cells, such that loop formation may arise during the base and/or nucleotide excision repair of single strand breaks [11].

Mutagenic mismatch repair

DNA mismatch repair is a conserved system that recognises and repairs erroneous DNA replication such as insertions, deletions and the mis-incorporation of nucleotides (nt) [27]. Ironically this system is solely in place as an anti-mutagenic pathway; however, the mutagenic action of mismatch repair has been implicated in TNR expansions. Mismatch repair and error leading to mutagenesis is a complex mechanism involving several factors. As this mechanism is not the main focus of this review, we will only discuss a summarised version, since a current review describes the following paragraph in greater detail [28].

To be effective, the MMR system needs to not only identify the mismatch, but distinguish the difference between the newly synthesised and template strands. This process has been well studied and is highly conserved, even between prokaryotes and eukaryotes [29]. Studies in *Escherichia coli* discovered a gene, that when silenced lead to highly mutable strains, leading researchers to term the proteins encoded for by these genes 'Mut' proteins. In particular, three proteins are vital in directing and facilitating post-replicative MMR; MutS, MutL and MutH, all with eukaryotic homologs (MSHs) [30]. These proteins generate a single strand break that serves as a gateway for exonuclease and DNA helicase II activity that leads to degradation of the newly synthesised strand, until the mismatch is removed [28]. Error occurs through two proposed models; 1) the MutSβ (MSH2 and MSH3 complex) entrapment/hairpin escape model 2) the dysregulated strand directionality model. Both provide attractive arguments for a role of mismatch repair in TNR expansion, as both models propose impaired or dysregulated function of MutS, with the second also accounting for the MutL function [31-33]. Taken together, it can be seen why the presence of both slip-strand mispairing and mismatch repair creates a 'perfect genetic storm', where an error in transcription is introduced and the exact mechanism supposedly responsible for correcting the mutation unwittingly aids in its formation.

Spinocerebellar Ataxia

Autosomal dominant cerebellar ataxias (ADCAs) account for 34 unique disease types [34]. Although not all the causative genes/proteins for this large category of diseases have been identified, all have been unequivocally localised to different subchromosomal loci. With the genes/proteins of the six SCAs being identified no less than 20 years

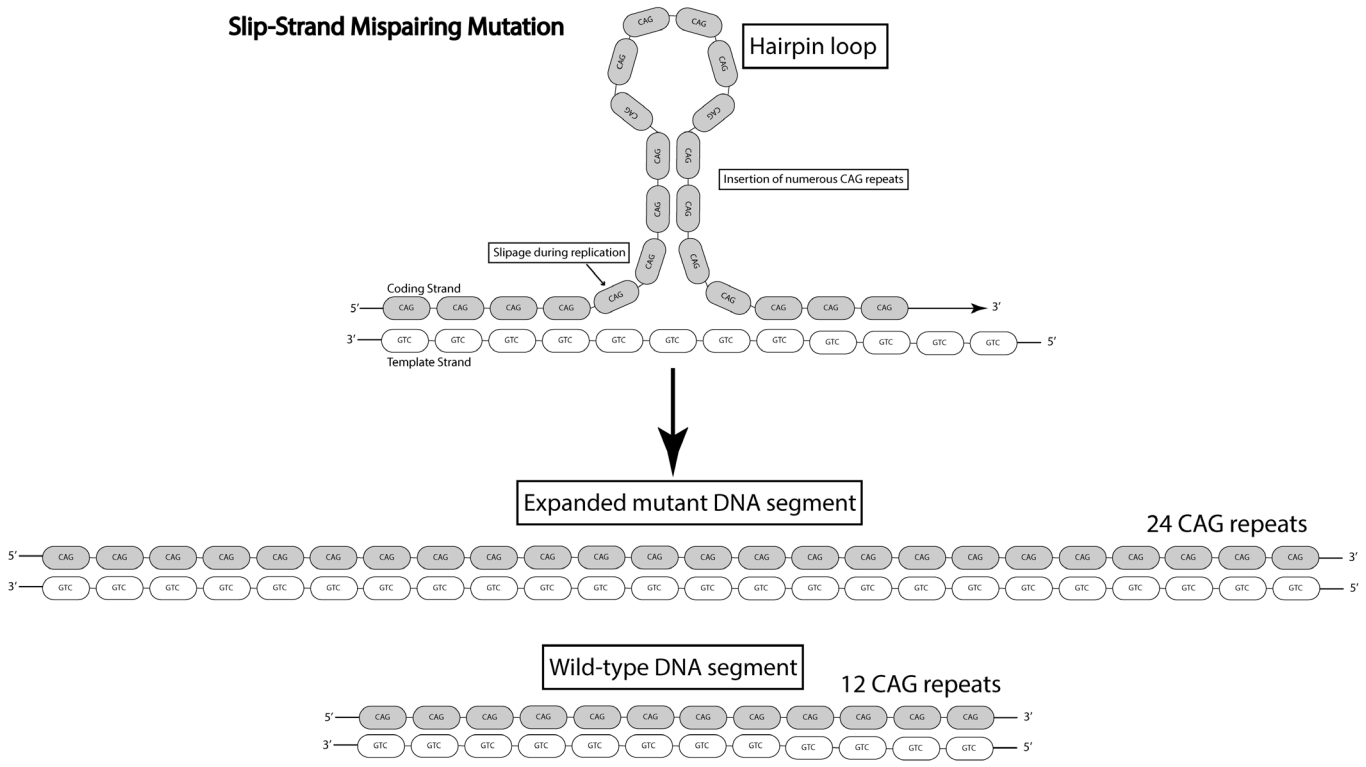

Figure 1: Slip strand mispairing mutation.

ago, with the exception of SCA17, characterised in 1999 (Table 1) [35-41]. Interestingly, the current numbering system of the various SCA diseases has been done in such a manner that follows the chronological order of gene locus discovery [34].

Autosomal dominant cerebellar ataxia (ADCA) type classification

ADCA may refer to any type of spinocerebellar ataxia that is inherited in an autosomal dominant manner. This classification system was introduced by AE Harding in 1982 to account for the varying degree of heterogeneity between the dominantly inherited ataxias [42]. Although, the classification is somewhat out-dated due to the genetically classified SCA system used in the present day, it is of some benefit to mention, as the system broadly classifies all ADCA into three discrete categories [43,44]. These categories were created on the basis of separating dominant ataxias by their complex phenotypes of ataxic and non-ataxic clinical manifestations; termed ADCA type I, II and III (Table 1) [42,45].

ADCA-I: This subtype is the most common form of ADCA and manifests as typical cerebellar ataxia with the addition of extrapyramidal signs, dementia, ophthalmoplegia, optic atrophy and amyotrophy. This classification includes SCA 1, 2, 3 and 17.

ADCA-II: The most highly specific of the subtypes as it is classified as cerebellar ataxia with associated retinal degeneration. SCA7 is the only SCA disease to be classified into this type.

ACDA-III: The classification that is defined as "pure" cerebellar ataxia, with secondary pathology not as prominent in this classification. SCA6 is the only disease to fall into this category.

Common clinical features

As previously stated, the six diseases are pathogenically heterogeneous; however, there are some features that are common to all types of SCA diseases. These diseases are typically late onset, with the mean age being in the third and fourth decade [46]. The age of onset may vary and is greatly, but not solely, dependent on the size of the expansion in the corresponding gene, with generally no significant difference in the age of onset between the sexes [47]. The average life span is approximately 10-20 years following diagnosis/symptomatic onset [48,49]. These diseases have selective neurodegeneration of areas such as the cerebellum, globus pallidus, pons and substantia nigra [2], along with progressive ataxia (incoordination and imbalance) leading to impairment of gait, speech and movement. In two thirds of SCA 1, 2, 3 and 6 cases, disorders of the individual's gait are the first pathogenic symptoms. Typically, individuals will need walking assistance within the first decade of onset and not long after become wheelchair bound [50]. Other common features include dysarthria, oculomotor disturbances and tremors, leading to impaired motor function of the hand. In addition to the physical aspects, several psychological disorders have been reported, most commonly depression, irritability and impaired sleep [51,52].

Protein aggregation: A hallmark feature

A hallmark molecular feature of SCA diseases, and all polyQ diseases for that matter, is the presence of protein aggregation and neuronal nuclear inclusions. An early example of this was reported by Paulson et al. [53] who provided patient-derived observations that mutant ataxin-3 results in significant nuclear inclusions in the neurons. These inclusions were mainly focused in the ventral pons of the brain,

however, these were also observed less frequently in the dorsal medulla, globus pallidus and the substantia nigra. The same group also showed that intracellular aggregates only occurred when ataxin-3 contained an expanded polyQ repeat, thus providing compelling evidence to the notion that mutant polyQ expansion directly leads to aggregation in the brain [53]. This research taken on its own does not provide definitive backing for the role of the polyQ tract in ataxin-3 aggregation, however, several subsequent studies both in animal and cell models have supported this concept over several different polyQ diseases [54-59]. Along with the presence of an expanded polyQ region, the length of the expansion itself is said to play a major role in aggregation. As studies have shown that the repeat length and the flanking sequences of most proteins implicated in polyQ diseases have an impact on severity of aggregation [60].

The ubiquitin-proteasome system and macroautophagy (autophagy) are two vital biological processes involving self-degradation of proteins and organelles [61]. These pathways are responsible for the clearance and degradation of misfolded and/or superfluous proteins. It stands to reason that these systems are somewhat compromised in SCA patients, and numerous studies have now shown that these systems do in fact exhibit impairment in SCA diseases [61-65]. The dysregulation is believed to play a role in protein aggregation, as the misfolded polyQ proteins are unable to be effectively degraded, which leads to an increased proportion of aggregates. These pathways are currently an obvious and attractive therapeutic avenue for SCA diseases [65,66].

Genetic testing

Each SCA disease has its own defined healthy and disease size ranges (Table 2). The healthy range represents a CAG repeat variation where anticipation and potential transmission does not occur unless a *de novo* mutation arises during embryonic development [67]. Several of the SCAs have a well-documented pre-mutation range, known as incomplete penetrance. These individuals are typically asymptomatic, however due to their dynamic nature, the expansions have the ability to pass on the disease or pass on an increased CAG repeat range [68,69]. Paternally transmitted alleles are typically more unstable and prone to expansion than a maternal allele, so that diseased individuals are those who have met the pathogenic threshold for the given gene, are symptomatic and are likely to pass on a larger expansion [69,70].

The advancement in genetic technology and testing has given rise to readily available commercial tests for all six SCA diseases [1,71,72]. Genetic testing may be conducted for five distinct clinical reasons: predictive testing, diagnostic testing, carrier testing, parental testing and risk factor assessment [67]. Thus, the primary benefit of genetic testing is to provide an accurate and specific diagnosis. These tests are often better done as early as possible, as this gives families precious time to come to terms with the potential disease, while giving affected individuals insight and the ability to make fundamental and necessary lifestyle changes and reproductive decisions. For those individuals where a *de novo* mutation has occurred, genetic testing would most likely provide relief and a psychological benefit in identifying a previously unknown disease and an end to the diagnostic saga.

Prevalence and incidence

Current data shows that SCA3 is the most common of the diseases [46,73-75], with an overall SCA prevalence of approximately 1-2 per 100 000. The epidemiological data available about the prevalence of SCA diseases is limited, and most likely does not represent an accurate occurrence, due to the high degree of variation between difference populations and ethnic backgrounds. Founder effects are seen in Cuba (particularly those of Spanish decent), Portugal and South Africa in SCA2, SCA3 and SCA7, respectively [47,76-78]. Several geographically localised studies have been conducted on the prevalence of ADCAs, with the focus on the six SCA diseases. With regards to SCA3, the prevalence is at its highest in Brazil (69% of SCA cases), Portugal (58%) and China (49%) and relatively low in USA (21%), India (3%) and Italy (1%) [77,79-83]. An interesting case is the founder effect of SCA7 in South Africa, where this disease represents 26.6% of SCA diseases [84], a figure that is significantly higher than the 2% occurrence worldwide [78]. This figure increases again when considering the native African population, in whom SCA7 represents 59% of all SCA diseases, while SCA3 only represents 1% [84]. In fact, distribution of SCAs in South Africa as a whole, drastically differs from that elsewhere [78,84].

SCA1

Clinical features

SCA1 is classified by a "cerebellar plus" syndrome with varying degrees of the commonly described SCA clinical characteristics [34,85]. Disease onset is usually in the third decade and progresses at a faster rate than seen in SCAs 2, 3, 6 and 17. SCA1 is known to have a main pathogenic feature of loss and atrophy of the cerebellar Purkinje cells, the major integrative neuron of the region [86] (Table 1). These cells are some of the largest cells in the brain and are responsible for the output of all motor coordination in the cerebellar cortex [87]. Death of these cells and cerebellar atrophy has a devastating effect on coordination and motor skills, contributing to the rapid decline and increased progression of ataxia [86,88].

Molecular genetics

SCA1 is caused by a CAG expansion in *ATXN1* that is localised to 6p22.3. *ATXN1* was first cloned by Zoghbi and colleagues in 1993, and then identified in 1994 by the same group [36,37]. The gene contains 9 exons with the potentially mutagenic CAG repeat region in exon 8. Although this gene contains 9 exons, only the last two (exons 8 and 9) are protein-coding, with exon 8 containing most of the coding sequence (1,950 nt). Healthy individuals are found to contain 6-39 CAG repeats, while the pathogenic range is from 40-82 [9,37]. Interestingly, repeats greater than 21 may be interrupted by 1-3 repeats of a CAT trinucleotide, which effectively rescues some individuals from disease [37] (Table 2).

Disease Name	Gene	Encoded protein	Locus	Exons in gene	PolyQ location	Healthy repeat range	Pathogenic repeat range
SCA1	*ATXN1*	Ataxin-1	6p22.3	9	Exon 8	6-39	40-82
SCA2	*ATXN2*	Atxain-2	12q24.1	25	Exon 1	17-29	37+
SCA3	*ATXN3*	Ataxin-3	14q24.3	11	Exon 10	7-44	55-86
SCA6	*CACNA1*	α1A subunit of the P/Q calcium-dependent voltage channel	19p13	47	Exon 47	4-18	21-30
SCA7	*AXTN7*	Ataxin-7	3p14.1	13	Exon 3	7-19	36 to >400
SCA17	*TBP*	TATA box binding protein	6q27	8	Exon 3	25-42	47-66

Table 2: Molecular genetics of the six polyQ spinocerebellar ataxias.

Wild-type and mutant protein

ATXN1 is expressed as the ~90 kDa, ataxin-1 protein, although it should be noted that this is an approximation, as the protein length will vary depending on the size of the expansion, a feature holding true across all variable repeat-containing genes and proteins further described. This review will discuss the 815 amino acid (aa) protein with a healthy polyQ region of 29 repeats (ENST00000244769.8). Normal ataxin-1 has the polyglutamine tract located towards the N-terminus, with three known functional regions; a conserved AXH domain, a nuclear localisation signal (NLS) and a Ser^{775} located at the C-terminus of the NLS (Figure 2). The protein is ubiquitously expressed, rather than being localised to the brain [89].

The evolutionary conserved 120 aa AXH domain that spans exon 8 and 9 is highly homologous to the large region of the high mobility group box transcription factor-binding protein 1 (HBP1) [90,91]. The AXH domain is known as a dimerization domain and is the only globular dimer forming region identified in the protein [92]. Additionally, the AXH folds independently into an oligonucleotide-binding fold, able to recognise RNA with a similar nucleotide preference, to that of full length ataxin-1 [91,93]. Second to RNA binding, the AXH domain contains a cluster of charged surface residues that allows for another secondary binding surface. This leads to the several protein-protein interactions through the ataxin-1 AXH domain, namely a paralog of ataxin-1, brother of ataxin-1 (BOAT), which also contains a AXH domain, the SMRT/SMRTER and Capicua proteins [94]. These interactions, along with interactions between several transcriptional factors, give ataxin-1 its transcriptional repression activity [90,91,93].

In terms of mutant ataxin-1, as with all SCA diseases, the addition of glutamines outside the healthy range leads to conformational changes in the affected protein, conferring a toxic gain of function [75,89,95-97]. The increased polyQ stretch not only increases the tendency to aggregate, but also leads to conformational changes of other regions and domains of the SCA proteins. In the case of SCA1, conformational changes to the AXH domain, along with its ability to fold leads to the potential contribution of misfolded protein aggregation [92,98,99]. his was shown by de Chiara et al. [100], where ataxin-1 aggregations were significantly mitigated by the removal of the AXH domain or by replacement of the HBP1 sequence, which is not known to aggregate [100]. Additionally, the self-association of ataxin-1 through its AXH domain leads to multimerisations that have the ability to aggregate polyQ stretches [98,101]. In conclusion, the AXH domain is crucial for both normal and aberrant functions of ataxin-1. Its ability to independently fold and in the presence of an elongated polyQ stretch, allow for rapid aggregation, may be a factor in the increased rate of progression when compared to other SCA diseases.

SCA 2

SCA2 is thought to be among the most prevalent of the SCAs, and demonstrates a founder effect in Cuba, in particular the north-eastern province of Holguin [50]. This region has a 40% prevalence rate of SCA2, well above the national average of approximately 6% [47]. The age of onset can range from as young as 3 years to age 80, with the typical mean age of onset in the third decade [47] (Table 1).

Clinical features

SCA2 demonstrates an atrophy pattern that resembles sporadic multi-system-atrophy, including some associated clinical features. These include early and marked saccadic slowing, initial hyperreflexia that is followed by hyporeflexia, Parkinson rigidity and myoclonus or fasciculation like movements [102,103]. With the slowing of saccadic jumps for horizontal gaze being the noticeable oculomotor abnormality (Table 1) [104-106]. This is attributed to the early, and sometimes rapid degeneration of the pontine brainstem. A study by Velazquez-Perez *et al.* (2004) was conducted to assess the effect on expansion size and maximal saccade velocity of 82 SCA2 patients and 80 healthy controls. It was found that 60-degree maximal saccade velocity was influenced by polyQ size rather than disease duration, suggesting that the saccade velocity of SCA2 patients is sensitive and specific to polyQ length, more so than disease stage or duration [107]. The same group conducted a follow up study in 2016 and found that patients with larger expansions deteriorated at a faster rate when compared to

Protein	Protein Size (amino acids)	Implicated Protein Functions
Ataxin-1 (SCA1)	815 aa	transcriptional repression, RNA metabolism
Ataxin-2 (SCA2)	1313 aa	transcriptional regulation, RNA metabolism, cytoskeletal reorganisation, calcium homeostasis
Ataxin-3 (SCA3)	361 aa	de-ubiquitination, proteasomal protein degradation, regulation of misfolded proteins
CACNA1* (SCA6)	2506 aa	pore formation, neurotransmitter release from presynaptic terminals, gives rise to P/Q type calcium currents
Ataxin-7 (SCA7)	892 aa	transcriptional regulation, mediates interaction of STAGA complex with the CRX, retinal functions
TATA box binding protein (SCA17)	339 aa	general transcription factor, initiation of transcription through TFIID binging to TATA box, interacts with all three RNA polymerases

* α1A subunit of the P/Q calcium-dependent voltage channel

Figure 2: Protein mutation.

those with smaller expansions. It was concluded that rate of decline in maximal saccade velocity is directly influenced by expansion size, thus providing clinicians with a sensitive biomarker and a potential measure of therapeutic success during clinical trials [108].

Other associated secondary characteristics such as insomnia (20% of cases) and sexual dysfunction (10%) are not uncommon for SCA2 patients [109]. Interestingly, individuals with relatively large expansions (>200) are reported to have retinitis pigmentosa/macular degeneration and myoclonus-epilepsy, which are two features commonly found in SCA7 and DRPLA, respectively [110,111]. As with SCA1, the early and significant loss of Purkinje cells is seen in this disease, with analysis indicating that the cerebellar flocculus is also affected to varying degrees [86,112].

Molecular genetics

The causative gene of SCA2 is *ATXN2* that was first localised to 12q24.1 in 1993 by Gispert et al. [113]. It was not until 1996 when three groups independently identified the CAG repeat expansion in *ATXN2* [40,114,115]. *ATXN2* encompasses approximately 130 kb of genomic DNA and consists of 25 exons, with the triplet repeat region located in the first exon (Table 2). The CAG repeat region in healthy individuals was found to have 1-3 interruptions of interspaced CAAs, the most common being two CAA interruptions, with a structure $(CAG)_8$ CAA $(CAG)_4$ CAA $(CAG)_8$ [40,115]. Healthy individuals have 17-29 repeats, while those individuals with incomplete penetrance (pre-mutation) have between 30-36 repeats. Severe pathogenesis is seen in individuals who have an uninterrupted CAG repeating range of 37 and above [40,114,115]. Interestingly, a moderate expansion size has also been implicated in another common motor neuron disease, amyotrophic lateral sclerosis (ALS) [116].

Wild-type and mutant protein

ATXN2 encodes for a highly basic protein (1,313 aa) with a molecular weight of approximately 140 kDa and in this specific isoform of ataxin-2, 23 glutamines are at the polyQ domain (ENST00000377617.7) (Figure 2). Ataxin-2, is the largest of the 'ataxin proteins' and the second largest protein after the CACA1A protein of the SCAs. The polyQ repeat region is located at the N-terminus of the protein. There are four other known functional domains in ataxin-2; two globular domains, spaning exons 2-7 named the Like Sm domain (Lsm; aa 255-346) and the Lsm associated domain (LsmAD; aa 354-476); towards to the C-terminus there is a PAM2 motif, aa 909-926, encoded by exon 16 that facilitates the association of polyA-binding protein (PABP); lastly the C-terminus contains a A2D region, that associates with Mp1 [117,118]. Ataxin-2 is ubiquitously expressed in adult tissues, with common, highly expressed regions such as the brain, gut, liver, heart muscle thyroid and lung. The most prominent expression of the protein is in the brain, with its major localisation being in Purkinje cells [119].

Normal ataxin-2 has been shown to be involved in numerous cellular processes, including transcriptional regulation, RNA metabolism, cytoskeletal reorganisation and calcium homeostasis [120-123]. Ataxin-2 is considered a cytoplasmic protein that is localised at the rough endoplasmic reticulum (rER) [124]. The globular domains (Lsm and LsmAD) are two regions that are highly conserved in proteins thought to be involved the process of RNA metabolism, such as splicing and modification [125,126]. The Lastres-Becker et al. study, found that when starved, mouse and human ataxin-2, was able to modulate translational control at the pre-initiation complex via PI3K/mTOR pathways [125]. This study highlights another possible major function of ataxin-2, its ability to regulate some metabolic activity with a study in *C. elegans* showing that down regulation of the ataxin-2 homolog leads to fat storage and an increase in growth. However, even when a dietary intervention was implemented, the *C. elegans* phenotype was unable to be reverted [127]. Several studies in the past several years have implicated ataxin-2 in body weight regulation and fat distribution, with a recent review Carmo-Silva et al. [123] describing the processes in greater detail [127-129].

Interestingly, neuronal nuclear inclusions are not as prominent in SCA2 patients when compared to other SCAs [130,131]. However, Purkinje cell death and selective neuronal degeneration is still apparent [130,131]. The conformational changes in ataxin-2, can be seen in the changes to the Lsm, LsmAD and PAM2 domains. A 2016 study used I-TASSER for 3D structure for protein structure prediction, and found several changes when the pathogenic polyQ repeat number was assessed. These changes are thought to contribute to the pathogenesis of SCA2 [132].

SCA3

SCA3/ Machado-Joseph disease was first described in 1972, in Portuguese immigrants living in Massachusetts, who were known to be decedents of William Machado [133]. SCA3 is the most common of the SCA disorders, and is at its highest prevalence in the Azores Islands, Portugal (~1:3472) [134]. SCA3 is widely known to be the most common of the SCA diseases, however, this does fluctuate between populations [67,69,80,84,85,135].

Clinical features

SCA3 typically onsets in the fourth decade and the average life span after onset is 10 years, with a range of 1-20 years. Along with the common clinical characteristics, SCA3 is often distinguished by the evident pyramidal and extrapyramidal signs [136]. The selective neuronal loss seen in SCA3 is found in the cerebellar dentate neurons, basal ganglia and brainstem [9,34,137]. SCA3 is also known for its higher frequency of lid retraction and infrequent blinking, commonly known as "staring eyes" (Table 1) [67]. The degree of peripheral involvement in pathogenesis is the greatest variable feature of SCA3, some patients have minor disease features, while others develop marked distal amyotrophic characteristics with sensory disturbances and areflexia [67].

Molecular genetics

SCA3 is caused by an expanded tract of CAG repeats in the coding region of the penultimate exon (exon 10) of the *ATXN3* gene (14q32.1), encoding the ataxin-3 protein [35,75]. The *ATXN3* gene, spanning a genomic region of approximately 48 kb (ENST00000558190.5) consists of 11 exons, with the start codon in exon 1 and the CAG repeat sequence located at the 5' end of exon 10 (Table 2) [138]. Healthy individuals have up to 44 CAG repeats, whereas affected individuals possess 55-86 repeats, and the intermediate range of 45-54 repeats is generally associated with incomplete penetrance [75]. However, there is much conjecture as to exactly number of repeats that may be associated with disease, since as few as 51 repeats have been reported to be pathogenic in some cases.

Wild-type and mutant protein

To date, 56 splice variants of *ATXN3* have been identified in blood, and 20 of these have the potential to be translated into a functional ataxin-3 protein [139]. Ataxin-3 has a molecular weight of approximately

42kDa and is 361aa in length with 8 glutamines [53] (Figure 2). Ataxin-3 is believed to have several roles within the cell, acting as an isopeptidase and is also thought to be involved in de-ubiquitination, proteasomal protein degradation, the chaperone system and regulation of misfolded proteins [98]. Although there are several isoforms of the Ataxin-3 protein, this review will refer to the isoform most commonly expressed in brain tissue, a 361aa protein with 8 glutamines at the polyQ region [75]. Located at the N-terminal of ataxin-3 is a Josephin domain, containing the amino acids (cysteine 14 (C) histidine 119 (H) and asparagine 134 (N)) that are crucial for the isopeptidase activity and two nuclear export signals (NES). The C-terminal contains three ubiquitin interacting motifs, a nuclear localisation signal and the polyQ tract [140-142].

Ataxin-3 is not only found in all cells, but is also dispersed throughout the cell. The protein has the ability to translocate from the cytoplasm to the nucleus and vice versa [143,144]. Todi et al. [145] conducted an *in vitro* study, where they catalytically inactivated ataxin-3 by a mutation to the active site cytosine at position 14 (C14A) and found that enzymatically active ataxin-3 preferentially localises to the nucleus, compared to the inactive form [145]. Additionally, the inactive form was degraded at a slower rate by the proteasome, an event found to be independent of ubiquitination. These two findings suggest that the catalytic activity of ataxin-3 regulates a number of its cellular properties and may in turn play a role in the disease pathogenesis of SCA3 [145]. Other studies suggest that there are several regions and factors that affect the localisation of ataxin-3, most of which are believed to be due to the NLS and NES [117,146].

The Josephine domain along with the three ubiquitin interacting motifs can either save proteins from degradation or stimulate the catalytic deubiquitinisation of proteins, thus resulting in protein degradation and clearance [140,147]. Poly-ubiquitin chains of four units or more are known to be recognised by ataxin-3 and the ubiquitin interacting motifs mediate selective binding to these ubiquitin chains and therefore restrict the types of chains that are cleaved [148]. Atxain-3 is also implicated in the regulation of misfolded endoplasmic reticulum protein degradation as it binds to the valosin-containing protein (VCP/p97) involved in ER-associated degradation [149,150].

Two important impaired functions of mutant ataxin-3 have been recognised, impaired protein degradation and transcriptional deregulation [65,151]. As ataxin-3 is thought to play a role in transcriptional regulation due to its interaction with transcriptional regulators, deregulation of this process is thought to be a factor when considering SCA3 pathogenesis. Due to the sequestration of ataxin-3 and other factors into nuclear aggregates, a secondary effect of transcriptional deregulation is thought to occur [152,153]. Additionally, Evert et al. showed that mutant ataxin-3 had impaired transcriptional repression through its ability to inhibit histone acetylase activity [154].

Interestingly, ataxin-3 has been found to bind more efficiently to VCP/p97 compared to normal ataxin-3, but by mechanisms unknown, and it interferes with protein degradation and misfolded protein clearance [155]. Although the exact mechanism by which interference occurs is poorly understood, several theories have been proposed. One such theory postulates that due to the increased binding affinity of mutant ataxin-3, it is unable to release itself after the misfolded proteins are extracted from the endoplasmic reticulum, thus affecting the subsequent round of protein extraction and degradation [156].

It has previously been unclear whether the protein aggregates observed in patients with SCA3 are full-length protein, shorter cleaved fragments or a combination of the two. However, more recently it appears that fragmented mutant ataxin-3 initially forms the intracellular aggregates and then acts a seed or a catalyst for recruitment of the full-length protein [53,157]. A study showed that L-glutamine-induced excitation of neurons lead to Ca^{2+} dependent proteolysis of ataxin-3, creating SDS-insoluble aggregates [57]: suggesting that initial microaggregates catalyse and precede large inclusion bodies that ultimately lead to neural death and rapid disease progression. It was also found that aggregation of the ataxin-3 fragments was calpain dependent and that the phenotypic changes could be abolished by calpain inhibition. The same study also showed that aggregation was neuron specific, which is in keeping with disease pathogenesis as aggregation is almost always observed in neural tissue of SCA3 patients.

Another line of thinking is that although mutant ataxin-3 is able to undergo ubiquitination at similar rates to wild type ataxin-3, mutant ataxin-3 was found to exist longer in the cell compared to wild type ataxin-3, thus facilitating aggregation through the lower rate of protein degradation [158]. There is ever growing evidence that both the truncated and full-length proteins are implicated in the aggregation of ataxin-3 in neurons. It is clear that neural aggregation of ataxin-3 directly leads to disease pathogenesis, however, the exact mechanism by which this is caused remains elusive.

SCA6

Clinical features

Unlike other SCA diseases, SCA6 typically presents as a milder form of disease, and is associated with "pure" cerebellar ataxia. However, ocular-movement abnormalities are often reported, with nystagmus being the most common form [112]. The disease onset tends to be later when compared to other SCA diseases (fifth or sixth decade) and is often less aggressive with average life duration being >25 years (Table 1) [136]. A comprehensive study on the frequency of nonataxic symptoms was conducted in 2008 by Klockgether and colleagues who found that SCA6 had a lower frequency in all but one (rigidity) of the 16 nonataxic symptoms when compared to SCA1, 2 and 3. These features included a number of very common symptoms such as; brainstem oculomotor signs, hyperreflexia, extensor planter and muscular atrophy [159]. It was also found that disease severity is largely age-dependent rather than linked to expansion size. This is not surprising due to the relatively small expansion size range.

Molecular genetics

SCA6 is one of only two SCA diseases to be caused by expansion in a protein other than an ataxin, the other being SCA17. The CAG expansion is seen in the α_{1A} voltage-dependant calcium channel gene (*CACNA1*), localised to 19p13 [39]. The expansion in *CACNA1* is located in the last exon of the gene (exon 47) and is relatively small compared to those in other SCA diseases, with a healthy range of 4-18 repeats and a disease range of 21-30 repeats, with most cases being reported as 22-23 repeats (Table 2) [159,160].

Interestingly, several dominant mutations in *CACNA1* are associated with three different, independent diseases; SCA6, episodic ataxia type 2 and familial hemiplegic migraine-1. Although these mutations arise in the same gene, the disease phenotypes are highly variable, with episodic ataxia type 2 being the most common form of episodic ataxia that typically presents during childhood or early adolescence. Familial hemiplegic migraine-1 is a debilitating variant of migraines, accompanied by aura that is characterised by severe attacks of motor deficits [161]. Traditionally, SCA6 is the only disease

to be associated with the polyQ expansion, as episodic ataxia type 2 is associated with a loss of function mutation, while familial hemiplegic migraine-1 is a gain of function mutation. However, familial studies in recent times have suggested that SCA6 phenotypes may also arise from missense mutations rather than expansion of the CAG repeating unit (more detail on these mutations can be found [162,163]).

Wild-type and mutant protein

CACNA1A encodes for the α1A subunit of the P/Q calcium-dependent voltage channel ($Ca_v2.1$). With the P/Q denoting the cell types in which the currents were initially isolated; P refers to Purkinje cells, while Q refers to granule cells in the cerebellum [164]. Unlike the ataxin proteins, Cav2.1 is brain specific, with no expression found in the heart, liver or muscle. $Ca_v2.1$ is the largest (by a significant manner) of the SCA proteins and its main isoform, of which there are seven, is a 2506 aa, 275 kDa protein with a polyQ region of 13 repeats (ENST00000360228.10) (Figure 2). $Ca_v2.1$ is a subunit responsible for pore-formation and plays a crucial role in neurotransmitter release from presynaptic terminals, and to a lesser extent, intrinsic firing patterns in Purkinje cells [164].

SCA6 has attracted attention due to its apparent 'deviation' from other SCA diseases. The expansions in the other five diseases are between 35-300 repeats, whereas SCA6 is typically 22 [165]. The apparent mechanism of toxic gain of function might not be as applicable to SCA6 as it is to the other SCAs. The belief is that the polyQ expansion in SCA6 somehow affects the normal wild-type function of the protein, altering functions of the P/Q channel, leading to Purkinje cell degeneration. However, this view is contentious and conflicting reports are presented, with conflicting reports on the alterations of the P/Q calcium-dependent voltage channel properties as a direct result of the expanded polyQ region [165-167]. Most recently, a 2015 study on SCA6 knock-in mice was conducted [165] and showed that polyQ length does not directly affect function of the $Ca_v2.1$, as mice with varying CAG lengths (84Q; 30Q and 14Q) had similar changes in current density [165]. This study concluded that like other polyQ diseases, SCA6 is most likely the result of toxic gain of function. These conflicting reports highlight the complexity of SCA diseases and present prime examples of why additional research is required to identify the exact pathogenic mechanisms behind these diseases.

SCA7

Clinical features

SCA7 is almost always distinguished from other SCA disease by virtue of severe cone-rod dystrophy retinal degeneration, and is sometimes referred to as olivopontocerebellar atrophy III [168]. SCA7 is typically the only SCA where affected individuals go completely blind (Table 1) [111]. SCA7 retinal pathology initially affects the cones, with progression to the rod photoreceptors, although cones are found to be more severely affected [78]. The initial sign of retinal degeneration is dyschromatopia in the blue-yellow axis [169]. Due to the large range of expansions in SCA7, infantile onset has devastatingly widespread pathogenesis that extends outside the typical CNS pathology. Magnetic resonance imaging of SCA7 patients show that the cerebellum and the pons are the most affected regions of the brain, however other regions of SCA brains are also effected [170]. SCA7 patients may also present with an increased amount of pontine atrophy when compared to other SCA patients. Hernandez-Castillo et al. conducted a study using 24 confirmed SCA7 patients and assessed the effect of selective degradation and its relationship to ataxic symptoms. It was found that ataxic severity was associated with a decrease in grey matter volume of distinct regions of the cerebellum, as well as degeneration of specific cortical areas [171].

Disease onset is typically in the third or fourth decade, however, SCA7 may present as early as the first year, the earliest onset of all SCA diseases [136]. Interestingly, in early onset patients, visual impairment can be apparent up to almost a decade before ataxic symptoms [73]. Conversely, late onset patients can experience ataxic symptoms decades before first visual pathology is observed [73].

Molecular genetics

The causative gene for SCA7, *ATXN7* was mapped to 3p14.1 and first cloned in 1997 [38]. The gene has 13 exons with the polyQ repeat region in exon 3, the first coding exon of the gene. The healthy repeat range in SCA7 is 7-19, with an incomplete penetrance of 19-35, while a wide disease range of 36 to >400 repeats has been reported (Table 2) [172]. Although all SCA diseases have a tendency for genetic anticipation, *ATXN7* is particularly known to be highly unstable, resulting in extreme anticipation and expansion during paternal transmission [38]. Expansions as large as 60 or more are associated with infantile SCA7 and disease progression is rapid and premature death is almost a certainty [173]. Such is the nature of the disease that pathogenesis may be apparent well before the first year of life in extreme cases [136,174].

Wild-type and mutant protein

ATXN7 encodes for ataxin-7, a ubiquitously expressed protein thought to be involved in transcriptional regulation and …other secondary functions. Ataxin-7 is an 892 aa, 98 kDa protein that has a polyQ repeat region of 10 (ENST00000295900.10) (Figure 2). The N-terminus of ataxin-7 contains the polyQ region, while nearer to the middle, a Block I domain is located with a zinc binding domain and a NLS located toward the C-terminus [175], with the nuclear localisation of ataxin-7 being vital for its transcriptional function. Preferential nuclear localisation occurs in many regions of the brain, such as the cerebellum, pons and inferior olive [176]. Along with these areas of the brain, nuclear localisation is also preferentially seen in the photoreceptor cell nuclei of the retina [177]. This is not surprising, considering the visual pathology seen in SCA7 that may be due to the increased presence of ataxin-7 in the retina. Ataxin-7 is also highly homologous to Sgf73, a protein found in yeast that acts as a subunit of the SAGA complex, which has its own mammalian homologs, namely SPT3/TAF9/GCN5 acetyl-transferase complex (STAGA) [178,179]. Ataxin-7 is thought to play an integral role in the interaction between the STAGA complex and rod-cone homeobox (CRX) [180]. The nuclear localisation of ataxin-7 has been long viewed as vital to its primary physiological function. However, a 2011 study by Okazawa and colleagues provided a novel role for cytoplasmic axtaxin-7. The group found that cytosolic ataxin-7 associates with microtubules, and the expression of ataxin-7 aids in the stabilisation of microtubules against nocodazole treatment. This was further supported by the observation of increased microtubule degradation when ataxin-7 was knocked-down. These findings implicate ataxin-7 in the stabilisation of the cytoskeletal network, providing a novel function for cytosolic ataxin-7 [181].

As the role of ataxin-7 is well established in transcriptional regulation, as well as several biological functions in the retina, it stands to reason that mutant ataxin-7 may impair these functions. La Spada et al. found that an expanded polyQ region in ataxin-7 severely impaired its wild-type function and antagonised CRX function that lead to cone-rod dystrophy in mice [176]. This finding is of interest as typically,

polyQ diseases are attributed to toxic gain of function, while this study seems to suggest that one of the main pathogenesis may be due to loss of normal function. However, ataxin-7 is still thought to undergo protein misfolding and proteolytic cleavage that leads to selective retinal and neuronal toxicity as well as the hallmark neuronal nuclear inclusions.

A 2013 study conducted by Strom and colleagues revealed a novel pathogenic mechanism of ataxin-7 in autophagic dysregulation. The group found that the inhibition of autophagy was through the co-aggregation of p53-FIP200 proteins and ataxin-7 aggregates that resulted in destabilisation of ULK1, due to decreased soluble FIP200. It was further shown that inhibition of P53 is able to restore soluble FIP200 and ULK1, which increases autophagic activity and subsequently reduces mutant ataxin-7 toxicity [62]. Therapeutic strategies targeting the pathogenic autophagic dysregulation would benefit from the understanding of the exact mechanisms by which pathogenesis is caused.

SCA17

SCA17 is an interesting SCA disorder for three reasons; 1) like SCA6, the causative gene is not an *ATXN* but rather the complex and vital *TBP* (*TATA-Box binding protein*) [41,182,183]. The biological functions of most ataxin proteins remain somewhat elusive; however, the TBP is a well-described protein. 2) Although *TBP* function has been described since 1991 and numerous protein studies have been conducted, SCA17 was only designated in 2001, a considerable time after all other SCAs [184,185]. 3) The glutamine tract seen in the TBP is due to a polymorphic repeat region of CAGs interrupted by CAA rather than the typical contiguous CAG repeats seen in the other SCA diseases [183].

Clinical features

SCA17 shows typical ataxic symptoms much like other SCA diseases, with degeneration of the cerebellum, particularly the Purkinje cells. The characteristic feature of SCA17 is the occurrence of frequent seizures and the high prevalence of psychiatric abnormalities (Table 1). Interestingly, immunoreactivity of neuronal nuclear inclusions of SCA17 patients appear to be more widely distributed through the grey matter when compared to other SCA diseases [182]. Age of onset in SCA17 is typically in the fourth decade, however symptoms can be seen in juvenile patients in extreme cases [85]. The disease progression of SCA17 is fairly rapid, with an average life span after diagnosis being 10-20 years [136].

Other common clinical features of SCA17 are progressive dementia and subsequent and concurrent chorea. These clinical features are highly similar to another polyQ disease, Huntington's disease [186], and SCA17 is often initially mis-diagnosed as Huntington's disease prior to genetic testing. Due to this, SCA17 is also known as Huntington's disease-like 4, and seems to be most prevalent in Asia, particularly Japan, China and Korea. It has also been suggested that the polyQ expansion may affect the function of TBP and embryonic development to such an extent that many do not survive the initial stages of development [187].

Molecular genetics

TBP is a well-studied gene localised to 6q27 that has 8 exons. The gene is known to contain a polymorphic CAA/CAG tract in exon 3 that when expanded causes SCA17 [41]. The *TBP* repeat region has been divided into five domains that include two variable domains (II and IV) and three fixed domains (I, III and V) [186]. The variability of domains II and IV gives rise to SCA17 with repeat ranges of 7-11 and 9-21, respectively. Healthy individuals have a total repeat range of 25-42, incomplete penetrance is caused by 43-46 repeats and the disease range varies between 47-66 (Table 2) [186,187].

Mutant and wild-type protein

The TBP is a vital protein that plays a crucial role in the initiation of transcription. The protein consists of 339 aa, is 37 kDa in size, with a polymorphic glutamine tract of 38 repeats and is the smallest of the SCA disease causing proteins (ENST00000230354.10) (Figure 2). TBP is one of many general transcription factors and interacts with all three eukaryotic RNA polymerases [185]. Of these factors, transcription factor IID (TFIID) is the first transcription factor that binds to the TATA box, a highly conserved sequence approximately 25-30 nt upstream of the transcription start site. TBP, along with numerous (approximately 13) TBP-associated factors make up the TFIID, which in turn is a component of the RNA polymerase II pre-initiation complex [185]. The polyQ tract is at the N-terminal of the protein and is thought regulate the DNA binding capability of the C-terminal. The variation of TBP binding to DNA affects the rate of transcription complex formation, and in turn the initiation of transcription [188]. The N-terminus varies in length due to the polyQ region, while the C-terminal is highly conserved [184]. The C-terminal contains two repeats of 88 aa that produce an inverted "U" or "saddle-shape" structure that effectively mounts the DNA and binds to the TATA box [184]. Such is the importance of TBP to cell survival that a 2002 study demonstrated that embryos with homozygous knockout for TBP did not survive past the blastocyst stage [189]. Although there are many other intricate functions and biological aspects of TBP, these are outside the scope of this review. More detail of the functions of TBP and transcription in general can be found at [185,190,191].

Transcriptional dysregulation has been widely implicated in a number of the SCA and polyQ diseases [153,154,192,193]. It stands to reason that a SCA disease in which the causative protein is a well-known transcription factor would have associated transcriptional dysregulation. This theory however is weakened by conflicting reports. One study demonstrated, via mobility shift electrophoresis, that mutant TBP binds less efficiently to DNA contain a TATA box when compared to normal TBP, and the polyQ expansion caused aberrant interactions between TBP and TFIIB [194]. When a similar study was conducted, however using a luciferase assay instead of mobility shift electrophoresis, the expanded polyQ TBP actually stimulated TATA box transcriptional activity [195].

These results suggest that rather than a global model for transcription dysregulation in SCA17 patients, a more specific and localised impairment of transcription occurs. This has been confirmed by numerous studies that implicate mutant TBP in both gain and loss of function pathogenesis [187]. Transcription factors such as Su(H), Sp1, TFIIB and NFY are all associated with toxic gain of function, while XBP1 and MyoD are associated with loss of function [187]. Many cellular processes are thought to be involved with these transcription factors, such as the chaperone system, TrkA and notch signalling as well as ER stress response [196-200], illustrating the complexity of SCA diseases and the need for a clear understanding of pathogenic mechanisms.

Potential Therapeutic Strategies

Although SCA diseases and their mechanisms of pathogenesis have been extensively studied for decades, there is still no effective treatment or prevention strategy. Some therapeutic strategies are in place that

attempt to alleviate symptomatic pathogenesis; however, these are often ineffective and rarely prolong life. For example, in SCA3, some respite is found in pharmaceutical strategies that aim to reduce the severity and progression of the Parkinsonian-like phenotype and other associated symptoms, such as depression [201-203]. Takei and colleagues have shown that Tandospirone, an antidepressant used in Asia, may provide some benefit to patients with SCA3, as significant improvement was observed in several self-rated physical and emotional scales [201]. While these results may support some improvement, the treatment is nonspecific to SCA3 and only symptomatic relief was observed.

It is clear that strategies aimed at alleviating progression and/or delay in onset in SCA diseases are of great interest to the patients and health researchers. In order to achieve such strategies, the events leading to neural aggregation and selective degeneration need to be identified. Although pathogenically heterogeneous, there is strong evidence to suggest a role for a number of potential universal pathogenic pathways. These include; aggregation-autophagy, protein misfolding, the ubiquitin-proteasome system and chaperone system, as well as transcriptional dysregulation, For the benefit of this review, only therapeutic strategies that have the potential to be applied to multiple SCA diseases will be addressed.

Suppression/modification of mutant gene expression and protein assembly

The current consensus is that the expanded CAG region in SCA proteins leads to toxic gain of function. Therefore, a strategy that aims to suppress or modify the mutant genes and their translated proteins is both logical and highly attractive. The strategies currently being trialled include antisense oligonucleotides (AOs) and RNA interference (RNAi).

Antisense oligonucleotides are single stranded oligomers typically 20-25 bp long and are able to anneal to RNA or DNA via Watson-Crick base pairing to a target (sense) strand. This allows the antisense compound to modify gene expression in a very specific manner and is dependent on the type of base and backbone chemistry [204]. For a long period of time, these oligomers offered significant promise but failed to deliver, as first generation oligonucleotides were plagued by issues of inconsistent synthesis, delivery and sensitivity to nuclease-induced degradation. The synthetic single stranded molecules where often able to modulate gene expression *in vitro*, but were unable to survive degradation and effectively enter the cell. However, in recent times, chemical modifications were developed and introduced that not only allowed the AOs to survive nuclease degradation, but also provided additional mechanisms to manipulate gene expression.

Currently, AOs are being used to treat various muscular and neurodegenerative diseases at varying stages of clinical trials, as well as a variety of other disease such as cancer [205]. A recent review showed that over 25 genes have been targeted using splice-switching AOs. These AOs are used as therapeutics to redirect pre-mRNA splicing, in which sequences vital to pre-mRNA processing are targeted, effectively blocking binding of the splicing factors to the region [205,206]. These types of AOs can be used to restore gene function, correct aberrant splicing, produce a novel transcript or even down regulate gene expression [205]. Most notably are *Exondys 51* and *Nusinersen*, which are treatments for Duchenne muscular dystrophy and spinal muscular atrophy, respectively [207,208]. *Exondys 51* gained FDA accelerated approval in September 2016, and is designed to skip *DMD* exon 51 during pre-mRNA processing in order to restore the open reading frame. *Exondys 51* will treat a subset of Duchenne muscular dystrophy patients who have deletions flanking exon 51 [208,209]. *Nusinersen* gained full FDA approval in December 2016 and provides the only approved treatment for patients with spinal muscular atrophy [207].

In terms of SCA diseases, van Roon-Mom et al. [210] have conducted several studies using AOs as a therapeutic strategy [95,211]. In 2011, the group used a single 2'-O-methyl (2'-O-Me) AO on a phosphorothioate backbone that effectively reduced several polyQ disease protein levels, which included ataxin-1 and ataxin-3 [211]. The group used the AOs to reduce transcript levels via selective repeat-length dependent reduction of the various CAG encoding transcripts. The *in vitro* study assessed the efficacy of various 2'-O-Me $(CUG)_n$ triplet repeat AOs and found that $(CUG)_7$ not only reduced the main target protein, Huntington, but also reduced the mutant mRNA levels of ataxin-1 and ataxin-3. Another benefit observed was that reduction in mRNA levels was mutant specific, with the wild-type allele remaining constant or only reduced by a nominal amount. This has major benefits for some of the SCA disease proteins, such as TBP that are essential for normal biological and cellular function. Therefore, a treatment that spares wild-type protein is viewed as the most beneficial strategy.

A subsequent 2013 study on SCA3 and its associated protein, ataxin-3 used AO mediated exon skipping *in vitro* to remove the CAG repeat region in exon 10 and still maintain normal biological function of the ataxin-3 protein [95]. Although results in patient cells were not as convincing as in healthy cells, the study still showed that exon skipping is an attractive potential therapy for SCA diseases. Furthermore, the FDA approval of *Exondys 51* shows that if an effective means of exon skipping *in vivo* is achieved, and modifications to the SCA proteins do not have any negative downstream effects, additional therapies using AO technology are not unrealistic. Most recently, a 2017 study showed that it was possible to suppress mutant ataxin-3 protein levels in a YAC MJD-84.2Q mouse (84 polyQ) transgenic mouse model. Methoxyethyl (MOE) modified gapmers, AOs with modified bases at the ends on a phosphorothioate backbone with a DNA core were injected intracerebroventricularly into the transgenic mice and resulted in widespread delivery of the AOs into several key regions of the brain. AOs were designed to reduce mutant and wildtype ataxin-3 levels by inducing RNase-H degradation of target mRNA at the AO:RNA duplex. Three of the 5 AOs tests showed a >50% reduction in the disease protein levels in the cerebellum, diencephalon and cervical spinal cord [212]. Although both mutant and wild-type mRNAs are suppressed in this study, it has been found that ataxin-3 knockout mice appear to be normal, which suggests that ataxin-3 may not be essential for viability [213]. This avenue therefore provides another strategy for targeting reduction or suppression of mutant ataxin-3.

Synthetic RNA analogues have the potential to bind to RNA target sequences and in turn regulate gene expression. Regulation of gene expression may be achieved in several ways, such as suppression of translation, promotion of specific transcript degradation, redirection of pre-mRNA processing or influencing mRNA stability. These molecules have long been thought of as attractive candidates for therapeutics [214]. Lentiviral encoded short-hairpin RNAs (shRNA) induced allele-specific RNA silencing in a rat model for mutant ataxin-3. This specificity was achieved through lentiviral vectors encoding siRNAs that selectively target a SNP that occurs in over 70% of SCA3 patients. This allele specific mechanism decreased the severity of neurological pathogenesis associated with SCA3 [215]. Furthermore, independent studies used a similar mechanism, adeno-associated viral (AAV) vector based delivery of shRNA and small interfering RNA (siRNA) to rescue phenotypes in SCA1 [216] and SCA7 in affected mouse models,

respectively. In the SCA7 mouse, treatment had a positive effect on Purkinje neuron molecular layer thickness and appeared to reduce the neuronal nuclear inclusions [217].

In 2012 Lima and colleagues noted that 2'-O-methyl (2'-O-Me) and 2'-fluoro (2'-F) modified bases on a phosphorothioate backbone yielded RNA-like single strands with the ability to overcome potential issues observed with vector based unmodified shRNA and siRNA technologies, such as susceptibility to degradation. These modifications, termed single-stranded siRNAs (ss-siRNA), allowed the molecules to effectively silence gene expression, however, the exact mechanisms by which this silencing occurred was not identified. It was hypothesised that the ss-siRNAs were able to enter the protein machinery and induce an RNAi like effect [218]. The same group went on to use these ss-siRNAs to silence expression in the two most common polyQ diseases, HD and SCA3 [219,220]. The ss-siRNA molecules are complementary to the CAG repeat regions found in these genes, thus, inhibiting expression of both ataxin-3 and Huntington via the same mechanisms for both diseases. A more recent 2017 study, using similar a chemistry supported these findings, with selective and efficient downregulation of mutant ataxin-3 observed in cultured fibroblasts [221]. Holistically, the positive results described show that both non-selective and selective modification of *ATXN3* and *HTT* is achievable, and further testing into the safety and efficacy of these treatments is needed. These tests will allow the treatment strategies to progress from preclinical to phase I and II trials, hopefully providing the first viable therapeutic strategies for the treatment of SCA diseases.

Prevention of misfolding and protein aggregation

There is much debate as to the exact role neuronal nuclear inclusions plays in pathogenesis, as researchers are still unable to determine whether the aggregates directly induce neurodegeneration or if the presence of misfolded proteins and oligomeric intermediates are to blame [7,8,222-224]. However, the prevention or reversal of neuronal nuclear inclusions still remains one of the most attractive avenues for therapeutic treatment [225].

Of note, the prevention of mutant SCA proteins or the removal of the CAG repeat should have a positive downstream effect on protein aggregation. Prevention of mutant SCA proteins or removal of the toxic CAG expansion will inhibit protein aggregation before it has a chance to be produced. These strategies therefore, have a twofold benefit and are believed to be the most promising of therapeutic possibilities in the opinion of many clinical researchers. In terms of direct inhibition, two main approaches exist; the inhibition of protein misfolding and aggregation; and the upregulation of protein/neuronal nuclear inclusions clearance.

Molecular chaperones are a large group of proteins that aid in covalent folding/unfolding of macromolecular structures, with a primary focus on protein folding. Heat shock proteins (HSP) are one such class of chaperones that are believed to play a key protective role in the aggregation of many SCA disease misfolded proteins [226-228]. Several studies over the past few decades have found therapeutic promise in upregulating the expression of various HSPs due to their potential to refold proteins and increase degradation [225]. Specifically, HSP40 and HSP70 have been shown to repress polyQ-induced neurodegeneration, as well as protect from cell death [227,229].

Of the HSP, the HSPB subfamily that comprises 10 individual protein members has anti-aggregation properties *in vitro* [228]. Vos et al. [228] conducted an *in vitro* study and studies in a Drosophila model, to assess the individual polyQ anti-aggregation properties of the HSPB. Although a significant proportion of the work reported was conducted in *Htt* expressing cells, many inferences can be drawn due to the similarities of pathogenic mechanism. HSPB7 and HSPB9 were the only two proteins shown to reduce aggregates in cells expressing a repeat region of 74, while HSPB7 was the only effective protein in cells expressing a 119 polyQ region. This work was replicated *in vivo* and showed that HSBP7 requires a functional autophagosomal machine in order to function [228]. These results suggest that increasing the expression of HSPB7 could be a potential therapeutic strategy. This may be easily achieved with a simple supplement of calcium pantothenate (a supplemental form of vitamin B5), as it has been reported to modulate gene expression of HSPB7 [230]. However, this is by no means a robust therapy and further studies would need to be conducted on the effects of upregulation and the effectiveness of calcium pantothenate to modulate HSPB expression to levels required for therapeutic benefit. Other drugs such as epigallocatechin and tetracycline have been linked to altered aggregation in SCA diseases, most notably SCA3 [231,232]. The two drugs have been shown to positively alter aggregation and toxicity of mutant ataxin-3 via two distinct mechanisms. Epigallocatechin is believed to inhibit the formation of the amyloidgenic β-sheet-rich structures that are known components in protein aggregation, and steer formation towards non-toxic forms. While tetracycline did not appear to create any structural alterations, it significantly increased the solubility of the previously insoluble aggregates [231].

The ubiquitin proteasome system and autophagy are believed to be the main avenues to improve protein clearance in SCA diseases and would be most beneficial when targeting the removal of short misfolded proteins, while autophagy is preferentially tailored to clear larger toxic fragments that potentially form oligomeric complexes. Autophagy is well known to be negatively regulated by mTOR, therefore therapies targeted at increasing autophagy may increase the biologically clearance of aggregated protein. Rapamycin is one of the most widely known mTOR inhibitors and has been found to induce a higher rate of autophagy in several polyQ diseases [63,233]. Temsirolimus (a rapamycin analogue) was administered to mice via intraperitoneal injection and found to decrease SCA3 aggregates as well as ameliorate motor function and coordination [234]. More recently, a derivate of triazole, OC-13 showed promise in increasing autophagic clearance of insoluble aggregates and demonstrates another plausible therapy in increasing autophagy [66].

Transcriptional dysregulation

Transcriptional dysregulation and repression have been implicated in several of the SCA diseases and none more so than SCA17, with SCA1, 3 and 7 also reported to involve some form of transcriptional dysregulation. Altered histone deacetylase activity has been reported in numerous SCA diseases, with a SCA3 mouse model providing evidence that HDAC overactivity may be apparent [153,235]. Histone deacetylases are a group of evolutionary conserved enzymes that are responsible for the removal of acetyl groups from lysine residues on proteins [236]. The group consists of four classes (I, II, III, IV), that are separated on the basis of biological mechanism [237].

Evidence suggests that SCA3 mouse models show transcriptional repression through the hypoacetylation of H3 and H4 histones [153,235]. A 2011 study by Wang and colleagues found that sodium butyrate, a known HDAC inhibitor, enhanced histone acetylation, which led to increased gene expression in genes that are typically supressed in the cerebellum of SCA3 mice [235]. Additionally, sodium butyrate treatment delayed onset and improved ataxic symptoms in the mouse model, as well as improved survival and the neurological

phenotype. A subsequent study using a different HDAC inhibitor, valproic acid, supported these results. The study used Drosophila and SCA3 cell models in two independent studies. Studies in the fly model demonstrated that prolonged administration of valproic acid slowed neurodegeneration, reduced climbing disability and increased the average lifespan of the flies. In cultured cells, valproic acid supressed apoptosis while increasing the H3 and H4 acetylation levels [238]. These studies show that it is possible to alleviate, to some degree the phenotypes associated with SCA3, and indicates potentially viable options for the treatment of SCA3.

Ataxin-7 has also been shown to interact with HDAC, with an increase in cerebellar (neurons and glia) HDAC3 protein levels observed in transgenic SCA7 mice. In fact, *in vitro* work conducted in transfected HEK293T cells, showed that ataxin-7 directly interacts with HDAC3, and that this interaction may be a possible route of pathogenesis through altered deacetylation [239]. In keeping with this concept, a study demonstrated altered decacetylase activity and lysine acetylation levels, in the brain of transgenic mice. Although the Wang et al.'s and the Yi et al. [238] study showed that HDAC inhibition is a potential therapeutic strategy, a more recent 2014 study reported a contradictory finding. Venkatraman et al. aimed to examine the role of HDAC3 in SCA1 and HDAC3s potential for therapeutic treatment, by testing the effects of a Purkinje-neuron specific, genetically depleted and null model of HDAC on transgenic SCA1 mice. Interestingly, knockout HDAC3 models showed a greater degree of motor impairment and neurodegeneration when compared to unmodified models. These results suggest that HDAC3 inhibition as a therapeutic strategy may pose potential risks, as it is likely HDAC3 function may play a crucial role in normal Purkinje neuron function [240]. It is clear that further research is required to accurately describe the role of HDAC in SCA pathogenesis, for it seems that targeting HDAC3 may not be a viable option.

Final Remarks

Advances in diagnostics in the past few decades have revealed a similar pathogenic mechanism in six of the autosomal dominant spinocerebellar ataxias. These six diseases (SCA1, 2, 3, 6, 7, 17) are caused by an expansion of a CAG repeating sequencing in the coding region of six independent genes. All six diseases present with common ataxic symptoms, however, there is still a large degree of phenotypic variation, from the rapid and early progression of SCA7, to the more slowly progressive, milder form of SCA6. The high degree of phenotypic variation is attributed to the varying number of the CAG repeats as well as the toxic function of the mutant protein. Although significant resources have been utilised in an endeavour to discover the exact mechanisms of toxicity caused by polyQ SCA diseases, a definitive answer still remains elusive, however, there is consensus that disease is caused by a toxic gain of function rather than wild-type loss of function. Other secondary causes such as protein misfolding, aggregation, transcriptional dysregulation and RNA toxicity have also been proposed. These common modes of pathogenesis are logical and suggest potential avenues for therapeutic intervention, with several pre-clinical studies being conducted in an attempt to provide relief for patients, in areas such as protein clearance and modification of gene expression. It is clear that the current symptomatic alleviation treatments are severely limited in their benefit. Ongoing research into the pathogenic cause of these diseases is needed, since once the primary causative mechanism of disease is found, a more focused and directed therapeutic target for this class of devastating diseases may be possible.

References

1. Dueñas AM, Goold R, Giunti P (2006) Molecular pathogenesis of spinocerebellar ataxias. Brain 129: 1357-1370.
2. Underwood BR, Rubinsztein DC (2008) Spinocerebellar ataxias caused by polyglutamine expansions: A review of therapeutic strategies. Cerebellum 7: 215-221.
3. Bettencourt C, Lima M (2011) Machado-Joseph Disease: From first descriptions to new perspectives. Orphanet J Rare Dis 6: 35.
4. Ashley CT Jr, Warren ST (1995) Trinucleotide repeat expansion and human disease. Annu Rev Genet 29: 703-728.
5. Warren ST (1996) The expanding world of trinucleotide repeats. Science 271: 1374-1375.
6. Tsilfidis C, MacKenzie AE, Mettler G, Barceló J, Korneluk RG (1992) Correlation between CTG trinucleotide repeat length and frequency of severe congenital myotonic dystrophy. Nat Genet 1: 192-195.
7. Fan HC, Ho LI, Chi CS, Chen SJ, Peng GS, et al. (2014) Polyglutamine (PolyQ) diseases: Genetics to treatments. Cell Transplant 23: 441-458.
8. Zoghbi HY, Orr HT (1999) Polyglutamine diseases: Protein cleavage and aggregation. Curr Opin Neurobiol 9: 566-570.
9. Matos CA, de Macedo-Ribeiro S, Carvalho AL (2011) Polyglutamine diseases: The special case of ataxin-3 and Machado-Joseph disease. Prog Neurobiol 95: 26-48.
10. Fiszer A, Krzyzosiak WJ (2013) RNA toxicity in polyglutamine disorders: Concepts, models and progress of research. J Mol Med (Berl) 91: 683-691.
11. McMurray CT (2010) Mechanisms of trinucleotide repeat instability during human development. Nat Rev Genet 11: 786-799.
12. Venter JC, Adams MD, Myers EW, Li PW, Mural RJ, et al. (2001) The sequence of the human genome. Science 291: 1304-1351.
13. Subramanian S, Mishra RK, Singh L (2003) Genome-wide analysis of microsatellite repeats in humans: Their abundance and density in specific genomic regions. Genome Biol 4: R13.
14. Slattery JP, Murphy WJ, O'Brien SJ (2000) Patterns of diversity among SINE elements isolated from three Y-chromosome genes in carnivores. Mol Biol Evol 17: 825-829.
15. Kashi Y, King D, Soller M (1997) Simple sequence repeats as a source of quantitative genetic variation. Trends Genet 13: 74-78.
16. Pellerone F, Edwards K, Thomas M (2015) Grapevine microsatellite repeats: isolation, characterisation and use for genotyping of grape germplasm from Southern Italy. VITIS-Journal of Grapevine Research 40: 179.
17. Lopes-Cendes I, Maciel P, Kish S, Gaspar C, Robitaille Y, et al. (1996) Somatic mosaicism in the central nervous system in spinocerebellar ataxia type 1 and Machado-Joseph disease. Ann Neurol 40: 199-206.
18. Benton CS, de Silva R, Rutledge SL, Bohlega S, Ashizawa T, et al. (1998) Molecular and clinical studies in SCA-7 define a broad clinical spectrum and the infantile phenotype. Neurology 51: 1081-1086.
19. Levinson G, Gutman GA (1987) Slipped-strand mispairing: A major mechanism for DNA sequence evolution. Mol Biol Evol 4: 203-21.
20. Pearson CE, Wang YH, Griffith JD, Sinden RR (1998) Structural analysis of slipped-strand DNA (S-DNA) formed in (CTG)n. (CAG)n repeats from the myotonic dystrophy locus. Nucleic Acids Res 26: 816-823.
21. Pearson CE, Edamura KN and Cleary JD (2005) Repeat instability: Mechanisms of dynamic mutations. Nat Rev Genet 6: 729-742.
22. Cho DH, Tapscott SJ (2007) Myotonic dystrophy: Emerging mechanisms for DM1 and DM2. Biochim Biophys Acta 1772: 195-204.
23. Petruska J, Hartenstine MJ, Goodman MF (1998) Analysis of strand slippage in DNA polymerase expansions of CAG/CTG triplet repeats associated with neurodegenerative disease. J Biol Chem 273: 5204-5210.
24. Ho TH, Savkur RS, Poulos MG, Mancini MA, Swanson MS, et al. (2005) Co-localization of muscle blind with RNA foci is separable from mis-regulation of alternative splicing in myotonic dystrophy. J Cell Sci 118: 2923-2933.
25. Wells RD (1996) Molecular basis of genetic instability of triplet repeats. J Biol Chem 271: 2875-2878.

26. Sinden RR, Wells RD (1992) DNA structure, mutations and human genetic disease. Curr Opin Biotechnol 3: 612-622.

27. Iyer RR, Pluciennik A, Burdett V, Modrich PL (2006) DNA mismatch repair: Functions and mechanisms. Chem Rev 106: 302-323.

28. Iyer RR, Pluciennik A, Napierala M, Wells RD (2015) DNA triplet repeat expansion and mismatch repair. Annu Rev Biochem 84: 199-226.

29. Groothuizen FS, Sixma TK (2016) The conserved molecular machinery in DNA mismatch repair enzyme structures. DNA Repair (Amst) 38: 14-23.

30. Acharya S, Foster PL, Brooks P, Fishel R (2003) The coordinated functions of the *E. coli* MutS and MutL proteins in mismatch repair. Mol Cell 12: 233-246.

31. Lang WH, Coats JE, Majka J, Hura GL, Lin Y, et al (2011) Conformational trapping of mismatch recognition complex MSH2/MSH3 on repair-resistant DNA loops. Proc Natl Acad Sci 108: E837-E44.

32. Owen BA, Yang Z, Lai M, Gajek M, Badger JD, et al (2005) (CAG) n-hairpin DNA binds to Msh2–Msh3 and changes properties of mismatch recognition. Nat Struct Mol Biol 12: 663-670.

33. Jakupciak JP, Wells RD (2000) Gene conversion (recombination) mediates expansions of CTG[middle dot]CAG repeats. J Biol Chem 275: 40003-40013.

34. Rüb U, Schöls L, Paulson H, Auburger G, Kermer P, et al. (2013) Clinical features, neurogenetics and neuropathology of the polyglutamine spinocerebellar ataxias type 1, 2, 3, 6 and 7. Prog Neurobiol 104: 38-66.

35. Kawaguchi Y, Okamoto T, Taniwaki M, Aizawa M, Inoue M, et al. (1994) CAG expansions in a novel gene for Machado-Joseph disease at chromosome 14q32.1. Nat Genet 8: 221-228.

36. Banfi S, Servadio A, Chung MY, Kwiatkowski TJ Jr, McCall AE, et al. (1994) Identification and characterization of the gene causing type 1 spinocerebellar ataxia. Nat Genet 7: 513-520.

37. Orr HT, Chung MY, Banfi S, Kwiatkowski TJ Jr, Servadio A, et al. (1993) Expansion of an unstable trinucleotide CAG repeat in spinocerebellar ataxia type 1. Nat Genet 4: 221-226.

38. David G, Abbas N, Stevanin G, Dürr A, Yvert G, et al. (1997) Cloning of the SCA7 gene reveals a highly unstable CAG repeat expansion. Nat Genet 17: 65-70.

39. Zhuchenko O, Bailey J, Bonnen P, Ashizawa T, Stockton DW, et al. (1997) Autosomal dominant cerebellar ataxia (SCA6) associated with small polyglutamine expansions in the alpha 1A-voltage-dependent calcium channel. Nat Genet 15: 62-69.

40. Imbert G, Saudou F, Yvert G, Devys D, Trottier Y, et al. (1996) Cloning of the gene for spinocerebellar ataxia 2 reveals a locus with high sensitivity to expanded CAG/glutamine repeats. Nat Genet 14: 285-291.

41. Nakamura K, Jeong SY, Uchihara T, Anno M, Nagashima K, et al. (2001) SCA17, a novel autosomal dominant cerebellar ataxia caused by an expanded polyglutamine in TATA-binding protein. Hum Mol Genet 10: 1441-1448.

42. Harding A (1982) The clinical features and classification of the late onset autosomal dominant cerebellar ataxias. A study of 11 families, including descendants of the Drew family of Walworth. Brain: 105: 1-28.

43. Sun YM, Lu C, Wu ZY (2016) Spinocerebellar ataxia: Relationship between phenotype and genotype - A review. Clin Genet 90: 305-314.

44. Whaley NR, Fujioka S, Wszolek ZK (2011) Autosomal dominant cerebellar ataxia type I: A review of the phenotypic and genotypic characteristics. Orphanet J Rare Dis 6: 33.

45. Harding AE (1984) The hereditary ataxias and related disorders. Churchill Livingstone.

46. Durr A1 (2010) Autosomal dominant cerebellar ataxias: Polyglutamine expansions and beyond. Lancet Neurol 9: 885-894.

47. Pérez LV, Cruz GS, Falcón NS, Mederos LEA, Batallan KE, et al (2009) Molecular epidemiology of spinocerebellar ataxias in Cuba: insights into SCA2 founder effect in Holguin. Neurosci Lett 454: 157-160.

48. Gusella JF, MacDonald ME (2000) Molecular genetics: Unmasking polyglutamine triggers in neurodegenerative disease. Nat Rev Neurosci 1: 109-115.

49. Seidel K, Siswanto S, Fredrich M, Bouzrou M, Brunt ER, et al. (2016) Polyglutamine aggregation in Huntington's disease and spinocerebellar ataxia type 3: Similar mechanisms in aggregate formation. Neuropathol Appl Neurobiol 42: 153-166.

50. Lastres-Becker I, Rüb U, Auburger G (2008) Spinocerebellar ataxia 2 (SCA2). Cerebellum 7: 115-124.

51. D'Abreu A, França MC, Paulson HL, Lopes-Cendes I (2010) Caring for Machado-Joseph disease: current understanding and how to help patients. Parkinsonism Relat Disord 16: 2-7.

52. Schmahmann JD (2004) Disorders of the cerebellum: Ataxia, dysmetria of thought and the cerebellar cognitive affective syndrome. J Neuropsychiatry Clin Neurosci 16: 367-378.

53. Paulson H, Perez M, Trottier Y, Trojanowski J, Subramony S, et al (1997) Intranuclear inclusions of expanded polyglutamine protein in spinocerebellar ataxia type 3. Neuron 19: 333-344.

54. Evert BO, Vogt IR, Kindermann C, Ozimek L, de Vos RA, et al. (2001) Inflammatory genes are upregulated in expanded ataxin-3-expressing cell lines and spinocerebellar ataxia type 3 brains. J Neurosci 21: 5389-5396.

55. Schmidt T, Lindenberg KS, Krebs A, Schöls L, Laccone F, et al. (2002) Protein surveillance machinery in brains with spinocerebellar ataxia type 3: Redistribution and differential recruitment of 26S proteasome subunits and chaperones to neuronal intranuclear inclusions. Ann Neurol 51: 302-310.

56. Evert BO, Wüllner U, Schulz JB, Weller M, Groscurth P, et al. (1999) High level expression of expanded full-length ataxin-3 *in vitro* causes cell death and formation of intranuclear inclusions in neuronal cells. Hum Mol Genet 8: 1169-1176.

57. Koch P, Breuer P, Peitz M, Jungverdorben J, Kesavan J, et al. (2011) Excitation-induced ataxin-3 aggregation in neurons from patients with Machado-Joseph disease. Nature 480: 543-546.

58. Petrakis S, Schaefer MH, Wanker EE, Andrade-Navarro MA (2013) Aggregation of polyQ-extended proteins is promoted by interaction with their natural coiled-coil partners. Bioessays 35: 503-507.

59. Ross CA, Poirier MA (2004) Protein aggregation and neurodegenerative disease. Nat Med 10: S10-17.

60. Thakur AK, Jayaraman M, Mishra R, Thakur M, Chellgren VM, et al (2009) Polyglutamine disruption of the huntingtin exon 1 N terminus triggers a complex aggregation mechanism. Nat Struct Mol Biol 16: 380-389.

61. Jimenez-Sanchez M, Thomson F, Zavodszky E, Rubinsztein DC (2012) Autophagy and polyglutamine diseases. Prog Neurobiol 97: 67-82.

62. Yu X, Muñoz-Alarcón A, Ajayi A, Webling KE, Steinhof A, et al (2013) Inhibition of autophagy via p53-mediated disruption of ULK1 in a SCA7 polyglutamine disease model. J Mol Neurosci 50: 586-99.

63. Sarkar S, Ravikumar B, Floto R, Rubinsztein D (2009) Rapamycin and mTOR-independent autophagy inducers ameliorate toxicity of polyglutamine-expanded huntingtin and related proteinopathies. Cell Death Differ 16: 46-56.

64. Alves S, Cormier-Dequaire F, Marinello M, Marais T, Muriel MP, et al. (2014) The autophagy/lysosome pathway is impaired in SCA7 patients and SCA7 knock-in mice. Acta Neuropathol 128: 705-722.

65. Nath SR, Lieberman AP (2017) The ubiquitination, disaggregation and proteasomal degradation machineries in polyglutamine disease. Front Mol Neurosci 10: 78.

66. Hsieh CH, Lee LC, Leong WY, Yang TC, Yao CF, et al. (2016) A triazole derivative elicits autophagic clearance of polyglutamine aggregation in neuronal cells. Drug Des Devel Ther 10: 2947-2957.

67. Paulson HL (2009) The spinocerebellar ataxias. J Neuroophthalmol 29: 227-237.

68. Michalik A, Martin JJ, Van Broeckhoven C (2004) Spinocerebellar ataxia type 7 associated with pigmentary retinal dystrophy. Eur J Hum Genet 12: 2-15.

69. Martindale JE (2017) Diagnosis of spinocerebellar ataxias caused by trinucleotide repeat expansions. Curr Protoc Hum Genet.

70. Wagner JL, O'Connor DM, Donsante A, Boulis NM (2016) Gene, stem cell and alternative therapies for SCA 1. Front Mol Neurosci 9: 67.

71. Klockgether T (2008) The clinical diagnosis of autosomal dominant spinocerebellar ataxias. Cerebellum 7: 101-105.

72. Klockgether T, Paulson H (2011) Milestones in ataxia. Mov Disord 26: 1134-1141.

73. Schöls L, Bauer P, Schmidt T, Schulte T, Riess O (2004) Autosomal dominant cerebellar ataxias: Clinical features, genetics and pathogenesis. Lancet Neurol 3: 291-304.

74. van de Warrenburg BP, Sinke RJ, Verschuuren-Bemelmans CC, Scheffer H, Brunt ER, et al. (2002) Spinocerebellar ataxias in the Netherlands: Prevalence and age at onset variance analysis. Neurology 58: 702-708.

75. Evers MM, Toonen LJ, van Roon-Mom WM (2014) Ataxin-3 protein and RNA toxicity in spinocerebellar ataxia type 3: Current insights and emerging therapeutic strategies. Mol Neurobiol 49: 1513-1531.

76. Orozco G, Estrada R, Perry TL, Araña J, Fernandez R, et al. (1989) Dominantly inherited olivopontocerebellar atrophy from eastern Cuba. Clinical, neuropathological and biochemical findings. J Neurol Sci 93: 37-50.

77. Sequeiros J, Coutinho P (1993) Epidemiology and clinical aspects of Machado-Joseph disease. Adv Neurol 61: 139-153.

78. Watson L, Wood M, Smith D, Scholefield J, Ballo R, et al (2016) Spinocerebellar ataxia type 7 in South Africa: Epidemiology, pathogenesis and therapy. S Afr Med J 106: 107-109.

79. Teive HA, Munhoz RP, Raskin S, Werneck LC (2008) Spinocerebellar ataxia type 6 in Brazil. Arq Neuropsiquiatr 66: 691-694.

80. Tang B, Liu C, Shen L, Dai H, Pan Q, et al. (2000) Frequency of SCA1, SCA2, SCA3/MJD, SCA6, SCA7, and DRPLA CAG trinucleotide repeat expansion in patients with hereditary spinocerebellar ataxia from Chinese kindreds. Arch Neurol 57: 540-544.

81. Moseley ML, Benzow KA, Schut LJ, Bird TD, Gomez CM, et al. (1998) Incidence of dominant spinocerebellar and Friedreich triplet repeats among 361 ataxia families. Neurology 51: 1666-1671.

82. Faruq M, Scaria V, Singh I, Tyagi S, Srivastava AK, et al. (2009) SCA-LSVD: A repeat-oriented locus-specific variation database for genotype to phenotype correlations in spinocerebellar ataxias. Hum Mutat 30: 1037-1042.

83. Brusco A1, Gellera C, Cagnoli C, Saluto A, Castucci A, et al. (2004) Molecular genetics of hereditary spinocerebellar ataxia: Mutation analysis of spinocerebellar ataxia genes and CAG/CTG repeat expansion detection in 225 Italian families. Arch Neurol 61: 727-733.

84. Smith DC, Bryer A, Watson LM, Greenberg LJ (2012) Inherited polyglutamine spinocerebellar ataxias in South Africa. S Afr Med J 102: 683-686.

85. Manto MU (2005) The wide spectrum of spinocerebellar ataxias (SCAs). Cerebellum 4: 2-6.

86. Clark HB, Burright EN, Yunis WS, Larson S, Wilcox C, et al. (1997) Purkinje cell expression of a mutant allele of SCA1 in transgenic mice leads to disparate effects on motor behaviors, followed by a progressive cerebellar dysfunction and histological alterations. J Neurosci 17: 7385-7395.

87. Ito M (2002) Historical review of the significance of the cerebellum and the role of Purkinje cells in motor learning. Ann N Y Acad Sci 978: 273-288.

88. Mullen RJ, Eicher EM, Sidman RL (1976) Purkinje cell degeneration, a new neurological mutation in the mouse. Proc Natl Acad Sci USA 73: 208-212.

89. Zoghbi HY, Orr HT (2009) Pathogenic mechanisms of a polyglutamine-mediated neurodegenerative disease, spinocerebellar ataxia type 1. J Biol Chem 284: 7425-7429.

90. de Chiara C, Giannini C, Adinolfi S, de Boer J, Guida S, et al. (2003) The AXH module: An independently folded domain common to ataxin-1 and HBP1. FEBS Lett 551: 107-112.

91. Deriu MA, Grasso G, Tuszynski JA, Massai D, Gallo D, et al (2016) Characterization of the AXH domain of ataxin-1 using enhanced sampling and functional mode analysis. Proteins 84: 666-673.

92. de Chiara C, Rees M, Menon RP, Pauwels K, Lawrence C, et al. (2013) Self-assembly and conformational heterogeneity of the AXH domain of ataxin-1: An unusual example of a chameleon fold. Biophys J 104: 1304-1313.

93. Grasso G, Deriu MA, Tuszynski JA, Gallo D, Morbiducci U, et al. (2016) Conformational fluctuations of the AXH monomer of Ataxin-1. Proteins 84: 52-59.

94. Mizutani A, Wang L, Rajan H, Vig PJ, Alaynick WA, et al. (2005) Boat, an AXH domain protein, suppresses the cytotoxicity of mutant ataxin-1. EMBO J 24: 3339-3351.

95. Evers MM, Tran HD, Zalachoras I, Pepers BA, Meijer OC, et al. (2013) Ataxin-3 protein modification as a treatment strategy for spinocerebellar ataxia type 3: Removal of the CAG containing exon. Neurobiol Dis 58: 49-56.

96. Taylor JP, Hardy J, Fischbeck KH (2002) Toxic proteins in neurodegenerative disease. Science 296: 1991-1995.

97. Orr HT, Zoghbi HY (2007) Trinucleotide repeat disorders. Annu Rev Neurosci 30: 575-621.

98. Kuiper EF, de Mattos EP, Jardim LB, Kampinga HH, Bergink S (2017) Chaperones in polyglutamine aggregation: Beyond the Q-stretch. Front Neurosci 11: 145.

99. de Chiara C, Menon RP, Kelly G, Pastore A (2013) Protein-protein interactions as a strategy towards protein-specific drug design: the example of ataxin-1. PLoS ONE 8: e76456.

100. de Chiara C, Menon RP, Dal Piaz F, Calder L, Pastore A (2005) Polyglutamine is not all: The functional role of the AXH domain in the ataxin-1 protein. J Mol Biol 354: 883-893.

101. Lasagna-Reeves CA, Rousseaux MW, Guerrero-Muñoz MJ, Park J, Jafar-Nejad P, et al (2015) A native interactor scaffolds and stabilizes toxic ATAXIN-1 oligomers in SCA1. Elife 4: e07558.

102. Furtado S, Payami H, Lockhart PJ, Hanson M, Nutt JG, et al (2004) Profile of families with parkinsonism-predominant spinocerebellar ataxia type 2 (SCA2). Movement disorders 19: 622-629.

103. Sasaki H, Fukazawa T, Wakisaka A, Hamada K, Hamada T, et al. (1996) Central phenotype and related varieties of spinocerebellar ataxia 2 (SCA2): A clinical and genetic study with a pedigree in the Japanese. J Neurol Sci 144: 176-181.

104. Termsarasab P, Thammongkolchai T, Rucker JC, Frucht SJ (2015) The diagnostic value of saccades in movement disorder patients: A practical guide and review. J Clin Mov Disord 2: 14.

105. Federighi P, Cevenini G, Dotti MT, Rosini F, Pretegiani E, et al. (2011) Differences in saccade dynamics between spinocerebellar ataxia 2 and late-onset cerebellar ataxias. Brain 134: 879-891.

106. Bürk K, Fetter M, Skalej M, Laccone F, Stevanin G, et al (1997) Saccade velocity in idiopathic and autosomal dominant cerebellar ataxia. J Neurol Neurosurg Psychiatry 62: 662-624.

107. Velázquez-Pérez L, Seifried C, Santos-Falcón N, Abele M, Ziemann U, et al. (2004) Saccade velocity is controlled by polyglutamine size in spinocerebellar ataxia 2. Ann Neurol 56: 444-447.

108. Rodríguez-Labrada R, Velázquez-Pérez L, Auburger G, Ziemann U, Canales-Ochoa N, et al. (2016) Spinocerebellar ataxia type 2: Measures of saccade changes improve power for clinical trials. Mov Disord 31: 570-578.

109. Reynaldo-Arminan R, Reynaldo-Hernandez R, Paneque-Herrera M, Prieto-Avila L, Perez-Ruiz E (2001) Mental disorders in patients with spinocerebellar ataxia type 2 in Cuba. Revista de Neurologia 35: 818-821.

110. Tan N, Zhou Y, Tan A, Chong S, Lee W (2004) Spinocerebellar ataxia type 2 with focal epilepsy-an unusual association. Ann Acad Med Singapore 33: 103-106.

111. Mao R, Aylsworth AS, Potter N, Wilson WG, Breningstall G, et al. (2002) Childhood-onset ataxia: Testing for large CAG-repeats in SCA2 and SCA7. Am J Med Genet 110: 338-345.

112. Ying SH, Choi SI, Lee M, Perlman SL, Baloh RW, et al. (2005) Relative atrophy of the flocculus and ocular motor dysfunction in SCA2 and SCA6. Ann N Y Acad Sci 1039: 430-435.

113. Gispert S, Twells R, Orozco G, Brice A, Weber J, et al. (1993) Chromosomal assignment of the second locus for autosomal dominant cerebellar ataxia (SCA2) to chromosome 12q23-24.1. Nat Genet 4: 295-299.

114. Sanpei K, Takano H, Igarashi S, Sato T, Oyake M, et al. (1996) Identification of the spinocerebellar ataxia type 2 gene using a direct identification of repeat expansion and cloning technique, DIRECT. Nat Genet 14: 277-284.

115. Pulst SM, Nechiporuk A, Nechiporuk T, Gispert S, Chen XN, et al. (1996) Moderate expansion of a normally biallelic trinucleotide repeat in spinocerebellar ataxia type 2. Nat Genet 14: 269-276.

116. Elden AC, Kim HJ, Hart MP, Chen-Plotkin AS, Johnson BS, et al. (2010) Ataxin-2 intermediate-length polyglutamine expansions are associated with increased risk for ALS. Nature 466: 1069-1075.

117. Albrecht M, Golatta M, Wüllner U, Lengauer T (2004) Structural and functional analysis of ataxin-2 and ataxin-3. Eur J Biochem 271: 3155-3170.

118. Meunier C, Bordereaux D, Porteu F, Gisselbrecht S, Chrétien S, et al. (2002) Cloning and characterization of a family of proteins associated with Mpl. J Biol Chem 277: 9139-9147.

119. Huynh DP, Del Bigio MR, Ho DH, Pulst SM (1999) Expression of ataxin-2 in brains from normal individuals and patients with Alzheimer's disease and spinocerebellar ataxia 2. Ann Neurol 45: 232-241.

120. Stubenvoll MD, Medley JC, Irwin M, Song MH (2016) ATX-2, the *C. elegans* ortholog of human Ataxin-2, regulates centrosome size and microtubule dynamics. PLoS Genet 12: e1006370.

121. Gnazzo MM, Uhlemann EE, Villarreal AR, Shirayama M, Dominguez EG, et al. (2016) The RNA-binding protein ATX-2 regulates cytokinesis through PAR-5 and ZEN-4. Mol Biol Cell 27: 3052-3064.

122. Magaña JJ, Velázquez-Pérez L, Cisneros B (2013) Spinocerebellar ataxia type 2: Clinical presentation, molecular mechanisms and therapeutic perspectives. Mol Neurobiol 47: 90-104.

123. Carmo-Silva S, Nobrega C, Pereira de Almeida L, Cavadas C (2017) Unraveling the role of Ataxin-2 in metabolism. Trends Endocrinol Metab 28: 309-318.

124. van de Loo S1, Eich F, Nonis D, Auburger G, Nowock J (2009) Ataxin-2 associates with rough endoplasmic reticulum. Exp Neurol 215: 110-118.

125. Lastres-Becker I, Nonis D, Eich F, Klinkenberg M, Gorospe M, et al (2016) Mammalian ataxin-2 modulates translation control at the pre-initiation complex via PI3K/mTOR and is induced by starvation. Biochimica et Biophysica Acta (BBA)-Molecular Basis of Disease 1862: 1558-1569.

126. Yokoshi M, Li Q, Yamamoto M, Okada H, Suzuki Y, et al. (2014) Direct binding of Ataxin-2 to distinct elements in 3' UTRs promotes mRNA stability and protein expression. Mol Cell 55: 186-198.

127. Bar DZ, Charar C, Dorfman J, Yadid T, Tafforeau L, et al (2016) Cell size and fat content of dietary-restricted Caenorhabditis elegans are regulated by ATX-2, an mTOR repressor. Proc Natl Acad Sci 113: E4620-E4629.

128. Mercader JM, González JR, Lozano JJ, Bak M, Kauppinen S, et al. (2012) Aberrant brain microRNA target and miRISC gene expression in the anx/anx anorexia mouse model. Gene 497: 181-190.

129. Dong L, Yan H, Huang X, Hu X, Yang Y, et al. (2015) A2BP1 gene polymorphisms association with olanzapine-induced weight gain. Pharmacol Res 99: 155-161.

130. Huynh DP, Yang HT, Vakharia H, Nguyen D, Pulst SM (2003) Expansion of the polyQ repeat in ataxin-2 alters its Golgi localization, disrupts the Golgi complex and causes cell death. Hum Mol Genet 12: 1485-1496.

131. Huynh DP, Figueroa K, Hoang N and Pulst S-M (2000) Nuclear localization or inclusion body formation of ataxin-2 are not necessary for SCA2 pathogenesis in mouse or human. Nat Genet 26: 44-50.

132. Wen J, Scoles DR, Facelli JC (2017) Effects of the enlargement of polyglutamine segments on the structure and folding of ataxin-2 and ataxin-3 proteins. J Biomol Struct Dyn 35: 504-519.

133. Nakano KK, Dawson DM, Spence A (1972) Machado disease. A hereditary ataxia in Portuguese emigrants to Massachusetts. Neurology 22: 49-55.

134. Bettencourt C, Santos C, Kay T, Vasconcelos J, Lima M (2008) Analysis of segregation patterns in Machado-Joseph disease pedigrees. J Hum Genet 53: 920-923.

135. Wang Y, Yang X, Ma W, Li J, Zhang Q, et al. (2016) Clinical features and genetic diagnosis of hereditary spinocerebellar ataxia 3. Mol Med Rep 14: 3731-3734.

136. Jayadev S, Bird TD (2013) Hereditary ataxias: Overview. Genet Med 15: 673-683.

137. Alves S, Régulier E, Nascimento-Ferreira I, Hassig R, Dufour N, et al (2008) Striatal and nigral pathology in a lentiviral rat model of Machado-Joseph disease. Hum Mol Genet 17: 2071-2083.

138. Ichikawa Y, Goto J, Hattori M, Toyoda A, Ishii K, et al. (2001) The genomic structure and expression of MJD, the Machado-Joseph disease gene. J Hum Genet 46: 413-422.

139. Bettencourt C, Santos C, Montiel R, do Carmo Costa M, Cruz-Morales P, et al (2010) Increased transcript diversity: Novel splicing variants of Machado–Joseph Disease gene (ATXN3). Neurogenetics. 11: 193-202.

140. Mao Y, Senic-Matuglia F, Di Fiore PP, Polo S, Hodsdon ME, et al. (2005) Deubiquitinating function of ataxin-3: Insights from the solution structure of the Josephin domain. Proc Natl Acad Sci U S A 102: 12700-12705.

141. Nicastro G, Menon RP, Masino L, Knowles PP, McDonald NQ, et al. (2005) The solution structure of the Josephin domain of ataxin-3: Structural determinants for molecular recognition. Proc Natl Acad Sci U S A 102: 10493-10498.

142. Nicastro G, Todi SV, Karaca E, Bonvin AM, Paulson HL, et al. (2010) Understanding the role of the Josephin domain in the PolyUb binding and cleavage properties of ataxin-3. PLoS ONE 5: e12430.

143. Tait D, Riccio M, Sittler A, Scherzinger E, Santi S, et al. (1998) Ataxin-3 is transported into the nucleus and associates with the nuclear matrix. Hum Mol Genet 7: 991-997.

144. Pozzi C, Valtorta M, Tedeschi G, Galbusera E, Pastori V, et al. (2008) Study of subcellular localization and proteolysis of ataxin-3. Neurobiol Dis 30: 190-200.

145. Todi SV, Laco MN, Winborn BJ, Travis SM, Wen HM, et al. (2007) Cellular turnover of the polyglutamine disease protein ataxin-3 is regulated by its catalytic activity. J Biol Chem 282: 29348-29358.

146. Antony PMA, Mäntele S, Mollenkopf P, Boy J, Kehlenbach RH, et al (2009) Identification and functional dissection of localization signals within ataxin-3. Neurobiology of disease 36: 280-292.

147. Winborn BJ, Travis SM, Todi SV, Scaglione KM, Xu P, et al. (2008) The deubiquitinating enzyme ataxin-3, a polyglutamine disease protein, edits Lys63 linkages in mixed linkage ubiquitin chains. J Biol Chem 283: 26436-26443.

148. Berke SJS, Chai Y, Marrs GL, Wen H, Paulson HL (2005) Defining the role of ubiquitin-interacting motifs in the polyglutamine disease protein, ataxin-3. J Biol Chem 280: 32026-32034.

149. Zhong X, Pittman RN (2006) Ataxin-3 binds VCP/p97 and regulates retrotranslocation of ERAD substrates. Hum Mol Genet 15: 2409-2420.

150. Tsou WL, Ouyang M, Hosking RR, Sutton JR, Blount JR, et al. (2015) The deubiquitinase ataxin-3 requires Rad23 and DnaJ-1 for its neuroprotective role in *Drosophila melanogaster*. Neurobiol Dis 82: 12-21.

151. Kristensen LV, Oppermann FS, Rauen MJ, Hartmann-Petersen R, Thirstrup K (2017) Polyglutamine expansion of ataxin-3 alters its degree of ubiquitination and phosphorylation at specific sites. Neurochem Int 105: 42-50.

152. Perez MK, Paulson HL, Pendse SJ, Saionz SJ, Bonini NM, et al. (1998) Recruitment and the role of nuclear localization in polyglutamine-mediated aggregation. J Cell Biol 143: 1457-1470.

153. Chou AH, Yeh TH, Ouyang P, Chen YL, Chen SY, et al. (2008) Polyglutamine-expanded ataxin-3 causes cerebellar dysfunction of SCA3 transgenic mice by inducing transcriptional dysregulation. Neurobiol Dis 31: 89-101.

154. Evert BO, Araujo J, Vieira-Saecker AM, de Vos RA, Harendza S, et al. (2006) Ataxin-3 represses transcription via chromatin binding, interaction with histone deacetylase 3 and histone deacetylation. J Neurosci 26: 11474-11486.

155. Laço MN, Cortes L, Travis SM, Paulson HL, Rego AC (2012) Valosin-containing protein (VCP/p97) is an activator of wild-type ataxin-3. PLoS ONE 7: e43563.

156. Li X, Liu H, Fischhaber PL, Tang TS (2015) Toward therapeutic targets for SCA3: Insight into the role of Machado-Joseph disease protein ataxin-3 in misfolded proteins clearance. Prog Neurobiol 132: 34-58.

157. Haacke A, Broadley SA, Boteva R, Tzvetkov N, Hartl FU, et al. (2006) Proteolytic cleavage of polyglutamine-expanded ataxin-3 is critical for aggregation and sequestration of non-expanded ataxin-3. Hum Mol Genet 15: 555-568.

158. Matsumoto M, Yada M, Hatakeyama S, Ishimoto H, Tanimura T, et al. (2004) Molecular clearance of ataxin-3 is regulated by a mammalian E4. EMBO J 23: 659-669.

159. Schmitz-Hübsch T, Coudert M, Bauer P, Giunti P, Globas C, et al. (2008) Spinocerebellar ataxia types 1, 2, 3 and 6: Disease severity and nonataxia symptoms. Neurology 71: 982-989.

160. Sinke RJ, Ippel EF, Diepstraten CM, Beemer FA, Wokke JH, et al. (2001) Clinical and molecular correlations in spinocerebellar ataxia type 6: A study of 24 Dutch families. Arch Neurol 58: 1839-1844.

161. Rajakulendran S, Kaski D, Hanna MG (2012) Neuronal P/Q-type calcium channel dysfunction in inherited disorders of the CNS. Nat Rev Neurol 8: 86-96.

162. Bürk K, Kaiser FJ, Tennstedt S, Schöls L, Kreuz FR, et al. (2014) A novel missense mutation in CACNA1A evaluated by *in silico* protein modeling is associated with non-episodic spinocerebellar ataxia with slow progression. Eur J Med Genet 57: 207-211.

163. Pradotto L, Mencarelli M, Bigoni M, Milesi A, Di Blasio A, et al (2016) Episodic ataxia and SCA6 within the same family due to the D302N CACNA1A gene mutation. J Neurol Sci 371: 81-84.

164. Westenbroek RE, Sakurai T, Elliott EM, Hell JW, Starr TV, et al. (1995) Immunochemical identification and subcellular distribution of the alpha 1A subunits of brain calcium channels. J Neurosci 15: 6403-6418.

165. Watase K (2015) Spinocerebellar ataxia type 6: Lessons from faithful knock-in mouse models. Neurol Clin Neurosci 3: 14-17.

166. Matsuyama Z, Wakamori M, Mori Y, Kawakami H, Nakamura S, et al. (1999) Direct alteration of the P/Q-type Ca^{2+} channel property by polyglutamine expansion in spinocerebellar ataxia 6. J Neurosci 19: RC14.

167. Toru S, Murakoshi T, Ishikawa K, Saegusa H, Fujigasaki H, et al. (2000) Spinocerebellar ataxia type 6 mutation alters P-type calcium channel function. J Biol Chem 275: 10893-10898.

168. Garden GA, La Spada AR (2008) Molecular pathogenesis and cellular pathology of spinocerebellar ataxia type 7 neurodegeneration. Cerebellum 7: 138-149.

169. McLaughlin ME, Dryja TP (2002) Ocular findings in spinocerebellar ataxia 7. Arch Ophthalmol 120: 655-659.

170. Bang OY, Huh K, Lee PH, Kim HJ (2003) Clinical and neuroradiological features of patients with spinocerebellar ataxias from Korean kindreds. Arch Neurol 60: 1566-1574.

171. Hernandez-Castillo CR, Galvez V, Diaz R, Fernandez-Ruiz J (2016) Specific cerebellar and cortical degeneration correlates with ataxia severity in spinocerebellar ataxia type 7. Brain Imaging Behav 10: 252-257.

172. Horton LC, Frosch MP, Vangel MG, Weigel-DiFranco C, Berson EL, et al. (2013) Spinocerebellar ataxia type 7: Clinical course, phenotype-genotype correlations and neuropathology. Cerebellum 12: 176-193.

173. David G, Dürr A, Stevanin G, Cancel G, Abbas N, et al. (1998) Molecular and clinical correlations in autosomal dominant cerebellar ataxia with progressive macular dystrophy (SCA7). Hum Mol Genet 7: 165-170.

174. Ansorge O, Giunti P, Michalik A, Van Broeckhoven C, Harding B, et al. (2004) Ataxin-7 aggregation and ubiquitination in infantile SCA7 with 180 CAG repeats. Ann Neurol 56: 448-452.

175. Helmlinger D, Hardy S, Sasorith S, Klein F, Robert F, et al. (2004) Ataxin-7 is a subunit of GCN5 histone acetyl transferase-containing complexes. Hum Mol Genet 13: 1257-1265.

176. La Spada AR, Fu YH, Sopher BL, Libby RT, Wang X, et al. (2001) Polyglutamine-expanded ataxin-7 antagonizes CRX function and induces cone-rod dystrophy in a mouse model of SCA7. Neuron 31: 913-927.

177. Lindenberg KS, Yvert G, Müller K, Landwehrmeyer GB (2000) Expression analysis of ataxin-7 mRNA and protein in human brain: Evidence for a widespread distribution and focal protein accumulation. Brain Pathol 10: 385-394.

178. Mohan RD, Dialynas G, Weake VM, Liu J, Martin-Brown S, et al (2014) Loss of Drosophila Ataxin-7, a SAGA subunit, reduces H2B ubiquitination and leads to neural and retinal degeneration. Genes Dev 28: 259-272.

179. McMahon SJ, Pray-Grant MG, Schieltz D, Yates JR 3rd, Grant PA (2005) Polyglutamine-expanded spinocerebellar ataxia-7 protein disrupts normal SAGA and SLIK histone acetyltransferase activity. Proc Natl Acad Sci U S A 102: 8478-8482.

180. Palhan VB, Chen S, Peng GH, Tjernberg A, Gamper AM, et al. (2005) Polyglutamine-expanded ataxin-7 inhibits STAGA histone acetyltransferase activity to produce retinal degeneration. Proc Natl Acad Sci U S A 102: 8472-8477.

181. Nakamura Y, Tagawa K, Oka T, Sasabe T, Ito H, et al. (2012) Ataxin-7 associates with microtubules and stabilizes the cytoskeletal network. Hum Mol Genet 21: 1099-1110.

182. Rolfs A, Koeppen AH, Bauer I, Bauer P, Buhlmann S, et al. (2003) Clinical features and neuropathology of autosomal dominant spinocerebellar ataxia (SCA17). Ann Neurol 54: 367-375.

183. Koide R, Kobayashi S, Shimohata T, Ikeuchi T, Maruyama M, et al. (1999) A neurological disease caused by an expanded CAG trinucleotide repeat in the TATA-binding protein gene: A new polyglutamine disease? Hum Mol Genet 8: 2047-2053.

184. Nikolov DB, Hu SH, Lin J, Gasch A, Hoffmann A, et al. (1992) Crystal structure of TFIID TATA-box binding protein. Nature 360: 40-46.

185. Roeder RG (1991) The complexities of eukaryotic transcription initiation: Regulation of pre-initiation complex assembly. Trends Biochem Sci 16: 402-408.

186. Cui Y, Yang S, Li XJ, Li S (2017) Genetically modified rodent models of SCA17. J Neurosci Res 95: 1540-1547.

187. Yang S, Li XJ, Li S (2016) Molecular mechanisms underlying spinocerebellar ataxia 17 (SCA17) pathogenesis. Rare Diseases. 4: 349-365.

188. Lescure A, Lutz Y, Eberhard D, Jacq X, Krol A, et al (1994) The N-terminal domain of the human TATA-binding protein plays a role in transcription from TATA-containing RNA polymerase II and III promoters. EMBO J 13: 1166.

189. Martianov I, Viville S, Davidson I (2002) RNA polymerase II transcription in murine cells lacking the TATA binding protein. Science 298: 1036-1039.

190. van Roon-Mom WM, Reid SJ, Faull RL, Snell RG (2005) TATA-binding protein in neurodegenerative disease. Neuroscience 133: 863-872.

191. White RJ, Jackson SP (1992) The TATA-binding protein: A central role in transcription by RNA polymerases I, II and III. Trends Genet 8: 284-288.

192. Mohan RD, Abmayr SM, Workman JL (2014) The expanding role for chromatin and transcription in polyglutamine disease. Curr Opin Genet Dev 26: 96-104.

193. Anglada-Huguet M, Vidal-Sancho L, Cabezas-Llobet N, Alberch J, Xifró X (2017) Pathogenesis of Huntington's disease: How to fight excitotoxicity and transcriptional dysregulation. Huntington's Disease-Molecular Pathogenesis and Current Models.

194. Friedman MJ, Shah AG, Fang ZH, Ward EG, Warren ST, et al. (2007) Polyglutamine domain modulates the TBP-TFIIB interaction: implications for its normal function and neurodegeneration. Nat Neurosci 10: 1519-1528.

195. Reid SJ, Rees MI, van Roon-Mom WM, Jones AL, MacDonald ME, et al. (2003) Molecular investigation of TBP allele length: A SCA17 cellular model and population study. Neurobiol Dis 13: 37-45.

196. Friedman MJ, Li S, Li XJ (2009) Activation of gene transcription by heat shock protein 27 may contribute to its neuronal protection. J Biol Chem 284: 27944-27951.

197. Huang S, Ling JJ, Yang S, Li XJ, Li S (2011) Neuronal expression of TATA box-binding protein containing expanded polyglutamine in knock-in mice reduces chaperone protein response by impairing the function of nuclear factor-Y transcription factor. Brain 134: 1943-1958.

198. Lee LC, Chen CM, Wang HC, Hsieh HH, Chiu IS, et al. (2012) Role of the CCAAT-binding protein NFY in SCA17 pathogenesis. PLoS ONE 7: e35302.

199. Ren J, Jegga AG, Zhang M, Deng J, Liu J, et al. (2011) A Drosophila model of the neurodegenerative disease SCA17 reveals a role of RBP-J/Su(H) in modulating the pathological outcome. Hum Mol Genet 20: 3424-3436.

200. Shah AG, Friedman MJ, Huang S, Roberts M, Li XJ, et al. (2009) Transcriptional dysregulation of TrkA associates with neurodegeneration in spinocerebellar ataxia type 17. Hum Mol Genet 18: 4141-4152.

201. Takei A, Hamada S, Homma S, Hamada K, Tashiro K, et al. (2010) Difference in the effects of tandospirone on ataxia in various types of spinocerebellar degeneration: An open-label study. Cerebellum 9: 567-570.

202. Takei A, Hamada T, Yabe I, Sasaki H (2005) Treatment of cerebellar ataxia with 5-HT1A agonist. Cerebellum 4: 211-215.

203. Buhmann C, Bussopulos A, Oechsner M (2003) Dopaminergic response in Parkinsonian phenotype of Machado-Joseph disease. Mov Disord 18: 219-221.

204. Uhlmann E, Peyman A (1990) Antisense oligonucleotides: A new therapeutic principle. Chem Rev 90: 543-584.

205. Havens MA, Hastings ML (2016) Splice-switching antisense oligonucleotides as therapeutic drugs. Nucleic Acids Res 44: 6549-6563.

206. Bauman J, Jearawiriyapaisarn N, Kole R (2009) Therapeutic potential of splice-switching oligonucleotides. Oligonucleotides 19: 1-13.

207. Finkel RS, Chiriboga CA, Vajsar J, Day JW, Montes J, et al (2017) Treatment of infantile-onset spinal muscular atrophy with nusinersen: A phase 2, open-label, dose-escalation study. Lancet 388: 3017-3026.

208. Mendell JR, Rodino-Klapac LR, Sahenk Z, Roush K, Bird L, et al. (2013) Eteplirsen for the treatment of Duchenne muscular dystrophy. Ann Neurol 74: 637-647.

209. Charleston J, Schnell F, Dworzak J, Donoghue C, Lewis S, et al (2016) Long-term treatment with eteplirsen promotes exon 51 skipping and novel dystrophin protein production in Duchenne muscular dystrophy patients. Neuromuscul Disord 26: S153.

210. Toonen LJ, Schmidt I, Luijsterburg MS, van Attikum H, van Roon-Mom WM (2016) Antisense oligonucleotide-mediated exon skipping as a strategy to reduce proteolytic cleavage of ataxin-3. Sci Rep 6: 35200.

211. Evers MM, Pepers BA, van Deutekom JC, Mulders SA, den Dunnen JT, et al. (2011) Targeting several CAG expansion diseases by a single antisense oligonucleotide. PLoS ONE 6: e24308.

212. Moore LR, Rajpal G, Dillingham IT, Qutob M, Blumenstein KG, et al (2017) Evaluation of antisense oligonucleotides targeting ATXN3 in SCA3 mouse models. Mol Ther Nucleic Acids 7: 200-210.

213. Schmitt I, Linden M, Khazneh H, Evert BO, Breuer P, et al. (2007) Inactivation of the mouse Atxn3 (ataxin-3) gene increases protein ubiquitination. Biochem Biophys Res Commun 362: 734-739.

214. Watts JK, Corey DR (2012) Silencing disease genes in the laboratory and the clinic. J Pathol 226: 365-379.

215. Alves S, Nascimento-Ferreira I, Auregan G, Hassig R, Dufour N, et al. (2008) Allele-specific RNA silencing of mutant ataxin-3 mediates neuroprotection in a rat model of Machado-Joseph disease. PloS ONE 3: e3341.

216. Xia H, Mao Q, Eliason SL, Harper SQ, Martins IH, et al (2004) RNAi suppresses polyglutamine-induced neurodegeneration in a model of spinocerebellar ataxia. Nat Med 10: 816-820.

217. Ramachandran PS, Boudreau RL, Schaefer KA, La Spada AR, Davidson BL (2014) Non-allele specific silencing of ataxin-7 improves disease phenotypes in a mouse model of SCA7. Molecular Therapy 22: 1635-1642.

218. Lima WF, Prakash TP, Murray HM, Kinberger GA, Li W, et al. (2012) Single-stranded siRNAs activate RNAi in animals. Cell 150: 883-894.

219. Liu J, Yu D, Aiba Y, Pendergraff H, Swayze EE, et al. (2013) ss-siRNAs allele selectively inhibit ataxin-3 expression: multiple mechanisms for an alternative gene silencing strategy. Nucleic Acids Res 41: 9570-9583.

220. Yu D, Pendergraff H, Liu J, Kordasiewicz HB, Cleveland DW, et al. (2012) Single-stranded RNAs use RNAi to potently and allele-selectively inhibit mutant Huntington expression. Cell 150: 895-908.

221. Fiszer A, Ellison-Klimontowicz ME, Krzyzosiak WJ (2017) Silencing of genes responsible for polyQ diseases using chemically modified single-stranded siRNAs. Acta Biochimica Polonica 63: 759-764.

222. Lipinski MM, Yuan J (2004) Mechanisms of cell death in polyglutamine expansion diseases. Curr Opin Pharmacol 4: 85-90.

223. Shao J, Diamond MI (2007) Polyglutamine diseases: Emerging concepts in pathogenesis and therapy. Hum mol genet 16: R115-R123.

224. Sengupta S, Bhattacharya S (2016) Polyglutamine diseases-understanding the mechanism of pathogenesis. J Neurol Disord 4: 2.

225. Escalona-Rayo O, Fuentes-Vázquez P, Leyva-Gómez G, Cisneros B, Villalobos R, et al (2017) Nanoparticulate strategies for the treatment of polyglutamine diseases by halting the protein aggregation process. Drug Dev Ind Pharm 43: 871-888.

226. Reina CP, Nabet BY, Young PD, Pittman RN (2012) Basal and stress-induced Hsp70 are modulated by ataxin-3. Cell Stress Chaperones 17: 729-742.

227. Muchowski PJ, Schaffar G, Sittler A, Wanker EE, Hayer-Hartl MK, et al. (2000) Hsp70 and hsp40 chaperones can inhibit self-assembly of polyglutamine proteins into amyloid-like fibrils. Proc Natl Acad Sci 97: 7841-7846.

228. Vos MJ, Zijlstra MP, Kanon B, van Waarde-Verhagen MA, Brunt ER, et al. (2010) HSPB7 is the most potent polyQ aggregation suppressor within the HSPB family of molecular chaperones. Hum Mol Genet 19: 4677-4693.

229. Jana NR, Tanaka M, Wang Gh, Nukina N (2000) Polyglutamine length-dependent interaction of Hsp40 and Hsp70 family chaperones with truncated N-terminal Huntington: Their role in suppression of aggregation and cellular toxicity. Hum Mol Genet 9: 2009-2018.

230. Wiederholt T, Heise R, Skazik C, Marquardt Y, Joussen S, et al. (2009) Calcium pantothenate modulates gene expression in proliferating human dermal fibroblasts. Exp Dermatol 18: 969-978.

231. Bonanomi M, Visentin C, Natalello A, et al. (2015) How Epigallocatechin-3-gallate and tetracycline interact with the Josephin domain of Ataxin-3 and alter its aggregation mode. Chemistry 21: 18383-18393.

232. Bonanomi M, Natalello A, Visentin C, Pastori V, Penco A, et al. (2014) Epigallocatechin-3-gallate and tetracycline differently affect ataxin-3 fibrillogenesis and reduce toxicity in spinocerebellar ataxia type 3 model. Hum Mol Genet 23: 6542-6552.

233. Ravikumar B, Vacher C, Berger Z, Davies JE, Luo S, et al. (2004) Inhibition of mTOR induces autophagy and reduces toxicity of polyglutamine expansions in fly and mouse models of Huntington disease. Nat Genet 36: 585-595.

234. Berger Z, Ravikumar B, Menzies FM, Oroz LG, Underwood BR, et al. (2006) Rapamycin alleviates toxicity of different aggregate-prone proteins. Hum Mol Genet 15: 433-442.

235. Wang HL, Chou AH, Chen SY, Yeh TH, Weng YH (2011) HDAC inhibitor sodium butyrate reverses transcriptional downregulation and ameliorates ataxic symptoms in a transgenic mouse model of SCA3. Neurobiol Dis 41: 481-488.

236. Dokmanovic M, Clarke C, Marks PA (2007) Histone deacetylase inhibitors: Overview and perspectives. Mol Cancer Res 5: 981-989.

237. Haberland M, Montgomery RL, Olson EN (2009) The many roles of histone deacetylases in development and physiology: Implications for disease and therapy. Nat Rev Genet 10: 32-42.

238. Yi J, Zhang L, Tang B, Han W, Zhou Y, et al. (2013) Sodium valproate alleviates neurodegeneration in SCA3/MJD via suppressing apoptosis and rescuing the hypoacetylation levels of histone H3 and H4. PLoS ONE 8: e54792.

239. Duncan CE, An MC, Papanikolaou T, Rugani C, Vitelli C, et al. (2013) Histone deacetylase-3 interacts with ataxin-7 and is altered in a spinocerebellar ataxia type 7 mouse model. Mol Neurodegener 8: 42.

240. Venkatraman A, Hu Y-S, Didonna A, Cvetanovic M, Krbanjevic A, et al. (2014) The histone deacetylase HDAC3 is essential for Purkinje cell function, potentially complicating the use of HDAC inhibitors in SCA1. Hum Mol Genet 23: 3733-3745.