Polyglutamine Diseases: Looking Beyond the Neurodegenerative Universe

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PERSPECTIVES

Multisystem disorders are often manifested by affecting more than one bodily system or tissue. We have recently reported a significantly higher prevalence of co-existing conditions in the cohorts of both pre- and symptomatic Huntington's disease (HD) gene carriers. We reported that even pre-symptomatic HD patients had a significantly higher number of comorbid conditions, while the symptomatic group of HD patients was characterised by a significantly lower percentage of subjects without any comorbidity (7%) in comparison to the control group (50%). This led us to conclude that HD patients have more comorbidities than controls and these increase in number as the disease progresses. For the first time, we identified 8 clusters of comorbid conditions, with musculoskeletal, psychiatric and cardiovascular clusters being significantly more frequent in both pre- and symptomatic HD patients, while neurological and gastrointestinal clusters showed significantly higher occurrences in the HD symptomatic group [1].

Huntington's disease is one of the nine polyglutamine (PolyQ) diseases which are a group of hereditary neurodegenerative disorders caused by expansion of unstable polyQ repeats in their associated disease proteins. PolyQ diseases include types 1, 2, 3, 6, 7 and 17 of spinocerebellar ataxias (SCA), Huntington's disease (HD), Dentatorubral-pallidoluysian atrophy (DRPLA), and spinal and bulbar muscular atrophy X-linked type 1 (SBMA, also known as SMAX1) [2]. Typically, HD and each of the other polyQ diseases are being described by three pathological domains: motor symptoms, cognitive impairment and behavioural disturbances [3]. There is relatively little knowledge about other co-existing conditions in these diseases, despite the fact that all polyQ proteins are ubiquitously expressed among all tissues and cell types. Moreover, some specific tissue-enrichments in polyQ expression levels should be noted. Hence, one may conclude that in addition to neurodegeneration, polyQ-expanded proteins may cause a wide array of abnormalities in peripheral tissues too. These effects can best be recognised at the moment as comorbid conditions, rather than as intrinsic functions of polyQ mutant proteins. So far, only skeletal muscle atrophy has been reported for four polyQ diseases, in both patients and animal models; these diseases are: SBMA, HD, DRPLA and spinocerebellar ataxia type 17 (SCA17).

As mentioned above, skeletal muscle wasting has been widely described as a peripheral pathology in HD. In HD mouse models, we found that muscle dysfunction was characterised by a change in the contractile characteristics of fast twitch muscles, and a decrease in twitch and tetanic force of hindlimb muscles. There was also a significant and progressive decrease in the number of motor units innervating the EDL (Extensor Digitorum

Longus) muscle. These physiological impairments were accompanied by the re-expression of contractile transcripts, and markers of muscle denervation, as well as the apparent deterioration of energy metabolism, decreased oxidation and altered purine metabolism [4]. These metabolic abnormalities were caused by altered transcript levels involved in: purine synthesis, metabolism, degradation and energy metabolism [5]. There is substantial evidence that several of the pathological features underlying HD-related skeletal muscle atrophy have also been reported in both pre- and symptomatic HD patients. Clinical studies revealed that HD patients had reduced muscle strength by 50%; had a reduced phosphocreatine to inorganic phosphate ratio at rest; had a reduced maximum rate of mitochondrial ATP production during recovery from exercise; and had mitochondrial defects like abnormally elongated and swollen mitochondria, with derangement of cristae and vacuoles, as judged by electron microscopy (reviewed in [6]). One might therefore conclude that HD-related skeletal muscle atrophy shares many molecular and physiological mechanisms with muscle cachexia in cancer mouse models [7].

There is still an open question over whether HD-related skeletal muscle wasting is an intrinsic effect of mutant HTT or if the skeletal muscle dysfunction is a consequence of ongoing neurodegenerative processes in the brain. There are a couple of examples in the literature which strongly suggest that skeletal muscle deterioration might be caused by gain or loss of function of mutant HTT itself. Firstly, it has been shown that mutant HTT has a direct impact on the expression of the muscle chloride channel (ClC-1) and *Kcnj2* (Kir2.1 potassium channel) transcripts; these were significantly reduced with apparent defects in mRNA processing [8]. The second example showed that myostatin inhibition resulted in a reduction in loss of muscle mass and grip strength impairment, and consequently delayed end-stage disease by approximately 20% of the HD mouse model lifespan [9]. Since this pharmacological intervention targeted only skeletal muscles locally, these findings show that, in principle, the CNS-skeletal muscle axis can be successfully targeted on the peripheral end. There are two other examples of polyQ diseases, namely SBMA and SCA17, where atrophy of skeletal muscles has been investigated either in affected individuals or in the respective mouse models. Histological studies on SBMA muscle biopsies have shown the presence of fibre-type grouping, atrophic fibres and angulated fibres (which are typical features of chronic denervation), signs of primary myogenic defects like necrotic myofibers, as well as myofibers with centrally located nuclei (reviewed in [10]). SCA17 knock-in mice with polyQ (105 glutamines) in the TATA box-binding protein (TBP) displayed skeletal muscle degeneration, with a reduction in the expression of muscle-specific genes. The morphology of SCA17 muscle tissues showed apparent changes in the intra-fiber Z-band breaks, poorly aligned fibres of the myofibrils, enlarged mitochondria, swollen spaces

between individual muscle cells in cross-sections and sarcomere disruption. The SCA17 mice were also characterised by significantly reduced grip strength and rotarod performance [11].

Perhaps the most extensively studied peripheral pathology was performed in pre-clinical and clinical settings for HD. We and others reported a number of molecular events underlying HD-related cardiomyopathy. Those include connexin-43 relocation at gap junctions, a significant deregulation of hypertrophic markers, a contractile dysfunction, re-expression of foetal genes, apoptotic cardiomyocyte loss and interstitial fibrosis in HD mouse models [12]. Further studies confirmed that contractile dysfunction might be caused by energy imbalances, changes in catabolism of purine nucleotides, steady-state internal redox derangements and an activation of AMPK, leading consequently to a shift in the cardiac substrate preference [13]. A comprehensive analysis of cardiovascular events related to HD in pre- and clinical settings has been recently published [14]. Hence, our findings indicating a cardiovascular cluster of diseases, showing a significantly higher occurrence even in the pre-symptomatic HD gene carriers, might mirror previously-described pathological events in both HD mouse models and in HD patients. However, whether those pathological events are driven by an intrinsic mutant HTT function – or are just comorbid conditions – remains to be answered. Possibly the best described example of an intrinsic polyQ effect in the heart was a proof-of-concept study, which studied an artificial transgenic mouse model expressing either a mutant polyQ peptide of 83 glutamines (PQ83), or a control peptide of 19 glutamines (PQ19), under the control of the α-myosin heavy chain promoter (MyHC) to drive cardiomyocyte-specific expression. The PQ83 transgenic mice developed cardiac dysfunction and dilation leading to a shortening of their lifespan – up to around eight months of age. The PQ83-induced heart failure was manifested by cardiomyocyte loss and autophagic and lysosomal content, indicative of increased autophagy [15].

A greater recognition of co-existing conditions in polyQ diseases might help to better understand health outcomes and improve clinical management. So far, we have a very limited knowledge of co-existing conditions in practically all nine polyQ diseases, both in clinical and pre-clinical settings, since previous research concentrated exclusively on neuronal degeneration. Only very recently has it been acknowledged that polyQ diseases might have a wide spectrum of peripheral pathologies that may contribute to disease progression, although there has been only a very limited effort to better understand the molecular events leading to those peripheral pathologies. Despite there being growing evidence about intrinsic effects of mutant polyQ proteins in the pathology of polyQ diseases, especially in the case of Huntington's disease, these illnesses are often still associated solely to pathological changes in the brain. Unfortunately, the majority of scientific bodies,

including research councils, are still driving polyQ research in line with XIXth century dogma, stemming from the time when HD was described. This might have negative consequences for upcoming therapeutic approaches, which are still focusing exclusively on the central nervous system. It is becoming apparent that current delivery routes, which target only specific regions of the brain or spine, might have a very limited efficacy for polyQ diseases. It seems very likely that any future therapeutic intervention for polyQ diseases will be based on a gene therapy approach where, by using genetic tools like ASO, siRNA or ZFP strategies, we will be silencing mutated gene alleles or their transcriptional products systemically. Hence, by treating not only the central nervous system but also the most affected non-neuronal cells/tissues, we might be in position to cure these disease Since we have already found a number of comorbid conditions in pre-symptomatic HD patients, the current strategies to enrol mainly symptomatic patients into clinical trials should be changed. Amended strategies involving pre- or even non-symptomatic patients should be used as a standard. Looking beyond the universe of neuronal degeneration will definitely give researchers and clinicians a better understanding of the complexity of polyQ diseases and will consequently make impending therapies more efficient, ultimately benefiting polyQ patients and their families.

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CONFLICTS OF INTEREST STATEMENT

None declared.

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