sweating following local pilocarpine application would be important measures to establish a post-ganglionic lesion site.

Another important avenue of research will be the establishment of symptomatic treatment of autonomic changes in CANVAS. There is good cause to believe that symptomatic treatments established in other autonomic neuropathies (e.g. diabetic autonomic neuropathy) will also mitigate autonomic symptoms in CANVAS. However, given the lack of data regarding this approach in CANVAS, treatment decisions will need to be made on an individual basis and extensive monitoring will be required.

In summary, the study by Wu et al. provides evidence that sympathetic and parasympathetic autonomic failure is an integral part of CANVAS. In clinical practice this finding adds to difficulties in the differential diagnosis of adult-onset, progressive ataxias. Additional studies assessing the safety and efficacy of existing drugs for countering autonomic symptoms in CANVAS are urgently required. Finally, from a scientific point of view, it will be interesting to establish the site of the lesion in CANVAS-associated autonomic failure, and we are convinced that future clinical and pathological studies in CANVAS will shed light on the pathomechanisms involved.

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Identification of the SCA21 disease gene: remaining challenges and promising opportunities

This scientific commentary refers to ‘TMEM240 mutations cause spinocerebellar ataxia 21 with mental retardation and severe cognitive impairment’, by Delplanque et al. (doi: 10.1093/brain/awu202).

For nearly one and a half centuries, physicians have observed that disorders of coordination often run in families, and have documented how variable these disorders can be in terms of their natural history and symptomatology (Friedreich, 1863; Menzel, 1891). Transmission of such afflictions from parent to child led to the realisation that they show an autosomal dominant pattern of inheritance. The disorders were named ataxias, from the Greek ‘taxis’ meaning order, and were associated with dysfunction of the cerebellum and its connections. For a century, they were classified mainly based upon their neuropathology, and were collectively known as olivopontocerebellar atrophies (Konigsmark and Weiner, 1970). Anita Harding refined this classification in the 1980s to account for the mode of inheritance and clinical features, and created a very useful classification scheme consisting of three types of autosomal dominant cerebellar ataxia (ADCA). According to the Harding ADCA classification system, patients with type I ADCA present with ataxia in combination with extracerebellar signs, type II ADCA patients display ataxia, extracerebellar signs, and also retinal degeneration, and type III ADCA patients exhibit a pure cerebellar ataxia without any extracerebellar signs (Harding, 1982). With the advent of molecular genetics, the ADCAs were further defined based upon their genetic linkage location and became known as the spinocerebellar ataxias, or ‘SCAs’. Today, there are more than 35 known SCA genetic loci, within which 22 causative genes have been identified (Matilla-Duenas et al., 2012). The SCAs that belong to Harding’s type I ADCA category are a very heterogeneous group of diseases, with highly pleiotropic phenotypes and complex pyramidal and extrapyramidal symptoms, including cognitive decline, motor neuron disease, neuropathy, and depression. In this month’s issue of Brain, Delplanque and colleagues add another piece to the SCA puzzle by refining the clinical description and defining the genetic basis of SCA21 (Delplanque et al., 2014).

References

SCA21 was initially identified by the very same group more than a decade ago (Vuilmaume et al., 2002), and its genetic locus was erroneously mapped to chromosome 7. The original report described a large French family in which patients exhibited gradually progressive gait and limb ataxia in combination with akinines, rigidity, tremor, and hyporeflexia. Several SCA21 patients also presented with mild to severe cognitive impairment. Re-analysis of this same family has now yielded evidence of additional clinical features including delayed acquisition of cognitive and motor skills, mild to severe mental retardation, variable onset of clumsiness, and examples of very slow disease progression (Delplanque et al., 2014). Although cognitive impairment has been observed in a few other SCAs, this phenotype is remarkably consistent in the large SCA21 pedigree described in the current work.

Since the 2002 report, dramatic advances have occurred in molecular and genomics technology, permitting refinement of the genetic linkage analysis through the use of genome-wide single nucleotide polymorphism (SNP) microarrays. These new results restricted the chromosomal region of interest for SCA21 to a region on the short arm of chromosome 1. Extensive genomic sequencing of several family members further narrowed this region down to an interval containing the TMEM240 gene, and yielded a non-synonymous SNP in the coding region of the TMEM240 gene that co-segregated perfectly with the SCA21 disease phenotype. Importantly, this variant was absent from almost 1000 control subjects, leading the authors to screen 368 other French families with unidentified SCA phenotypes. Directed sequencing of the TMEM240 gene in these SCA families revealed seven unrelated families with linked mutations in the very same gene. Two of these families possessed the P170L mutation detected in the original large kindred, while the other five families displayed distinct putative mutations in the TMEM240 gene. Interestingly, five of the six mutations reside in the carboxyl terminal of the TMEM240 amino acid coding region. One of these mutations, T80M, is located between two transmembrane alpha helices, and occurs in a family whose phenotype does not include cognitive impairment.

Despite the evidence for TMEM240 as the causal gene for SCA21, the mechanism by which the various mutations cause disease remains unclear. Of the six mutations identified, five are missense and one introduces a premature stop codon. The presence of a nonsense mutation among the six would seem to suggest that haploinsufficiency accounts for SCA21 disease pathogenesis; however, this nonsense mutation is located in the last exon and thus is unlikely to be targeted for nonsense-mediated decay. It is possible that a truncated protein product is generated; hence, it would be premature to conclude that the mutations cause a loss of function in the TMEM240 protein. On the other hand, too little is known about the TMEM240 protein and its normal function to support a gain-of-function mechanism. Indeed, the extent of our collective knowledge of this gene derives from whole brain transcriptome profiling indicating that it is widely expressed throughout the brain (Hawrylycz et al., 2012).

Importantly, Delplanque et al. analysed the degree of conservation of the five mutated amino acid residues and found that all of these amino acids are fully conserved down to zebrafish. Consequently, future studies should focus on the normal function of the TMEM240 protein and how the different disease-causing mutations alter function. Indeed, when one considers the SCAs that are not caused by repeat expansion mutations, one finds very little commonality between the different disease gene products. For example, one group of SCA disease genes consists of regulatory factors and enzymes, while another group encodes proteins that localize to the plasma membrane. Among the plasma membrane proteins implicated in SCAs, mutations in the KCNC3 potassium channel and in the inositol triphosphate receptor ITPR1 cause SCA13 and SCA15, respectively. SCA5 is caused by mutations in β-III spectrin, which normally plays a role in stabilizing EAAT4, a glutamate transporter in Purkinje cells (Perkins et al., 2010). As the TMEM240 protein is a membrane-spanning protein, it too could be involved in modulating ion channel function at the neuronal cell membrane, akin to the role of β-III spectrin.

Certainly, future research efforts should consider how the distinct plasma membrane-spanning proteins implicated in SCAs could be functionally linked: this underscores why the discovery of TMEM240 as the cause of SCA21 is so exciting and potentially important in advancing our understanding of ataxia disease biology. Of course, an understanding of TMEM240 protein function will be crucial not only for generating models of SCA21 disease pathogenesis, but also for developing strategies for treating SCA21 and related disorders.

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**References**


‘Don’t delay, start today’: delaying levodopa does not delay motor complications

This scientific commentary refers to ‘The modern pre-levodopa era of Parkinson’s disease: insights into motor complications from sub-Saharan Africa’, by Cilia et al. (doi:10.1093/brain/awu195).

Preventing the development of motor complications is one of the principal concerns when treating patients with Parkinson’s disease, and ‘levodopa-sparing’ approaches have been commonly touted as the best method of achieving this. Early use of dopamine agonists, rather than levodopa, was the preferred management strategy in the 1990–2000s, and several large randomized controlled trials reported delayed development of motor complications with this approach. However, follow-up studies revealed that once patients were started on levodopa, they developed motor complications of the same severity and at the same rate irrespective of whether levodopa had been initiated earlier or later. Indeed, after 10–14 years of treatment, patient profiles were essentially identical regardless of how they began dopaminergic therapy (Katzschlager et al., 2008). These results, combined with increasing awareness of the major side-effects of dopamine agonists—particularly sleepiness and impulse-control disorders—have led many physicians to switch to earlier use of levodopa as monotherapy, particularly in patients over 60 years of age in whom the risk of dyskinesia is lower.

In this issue of Brain, Cilia and colleagues present data that help endorse this strategy (Cilia et al., 2014). This cross-sectional and 4-year longitudinal study recruited patients with Parkinson’s disease from Ghana (n = 91) and Italy (n = 2282). The authors took advantage of the opportunity to study patients from sub-Saharan Africa who typically experience longer delays in receiving levodopa therapy than patients in first world countries. Demographics and disease-related factors influencing the development of motor fluctuations and levodopa-induced dyskinesia (LID) were evaluated for both groups. Although accurate recording of LID can be difficult, the authors attempted to determine dates of onset as precisely as possible.

A subgroup of subjects from Italy (n=50) who were levodopa-naive were matched to 59 subjects from Ghana who, for economic and medical reasons, had longer disease duration before starting levodopa (mean 4.2 years) than the therapy-, age- and sex-matched patients in Italy (mean 1.8 years). The subjects had comparable disease severity [as per Unified Parkinson’s Disease Rating Scale (UPDRS) part III motor scores OFF drug]. Disease duration at the time of developing motor fluctuations (mean 5–6 years) and LID (mean 6–7 years) was also similar in the two groups; thus the group from Ghana developed motor complications more quickly after starting levodopa. Logistic regression analysis for predictors of motor complications showed that higher levodopa dose, expressed as mg/kg, and longer disease duration at the time of starting levodopa, were the main risk factors. There was no association with duration of levodopa use.

The pathophysiology of levodopa-induced motor complications is likely due to a combination of disease-related factors, as well as the pharmacokinetics of levodopa itself. The relative contributions of these are hard to tease apart, as individuals with Parkinson’s disease are generally not left untreated for long periods, thus the effects of disease progression cannot be separated from those of chronic levodopa use. However, the fact that individuals with Parkinson’s disease develop motor complications over time suggests the involvement of progressive pathological changes, particularly degeneration of nigro-striatal dopamine terminals. Thus, early and more severe dyskinesia occurs in patients with more severe dopamine depletion, dyskinesia occurs earlier and more prominently on the side of the body most affected by parkinsonism, and subjects with severe secondary parkinsonism, e.g. MPTP-parkinsonism or multiple system atrophy type parkinsonism, have early onset of dyskinesia.

In the MPTP non-human primate model of Parkinson’s disease, at least 50% loss of dopamine is needed for normal doses of levodopa to induce dyskinesia. In the typical model where there is >90% dopamine depletion, dyskinesia can often be seen on first exposure to levodopa (Fox et al., 2010). However, Cilia and colleagues report that disease duration and clinical severity (as opposed to severity of striatal dopamine deficiency) do not necessarily equate as risk factors for the development of LID. This ‘disconnect’ is possibly due to the roles of various pre- and postsynaptic mechanisms in compensating for striatal dopamine deficiency. As the authors point out, historical data from the very early levodopa era (as described by Cotzias et al., 1969) are consistent with the conclusions of this ‘modern day’ study, i.e. that it is disease duration, not clinical severity or duration of levodopa, that seems to be most important. Further studies into the nature of these compensatory mechanisms may help identify potential therapeutic strategies.

However, the unusual pharmacokinetic properties of levodopa per se also contribute to the pathogenesis of motor complications,