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Triplet repeats on the rise

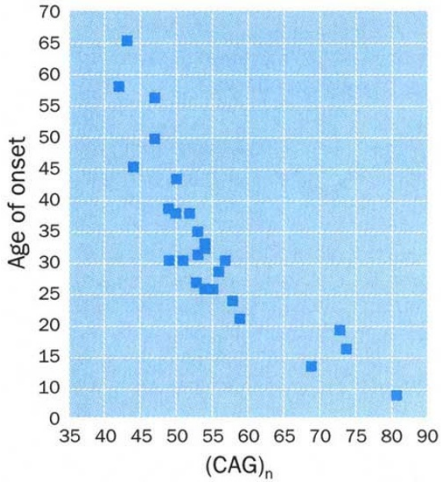
A direct search for expanding trinucleotide repeat sequences has paid off with the identification of the latest disorder caused by this mechanism — spinocerebellar ataxia type 1.

It was only four months ago that the gene responsible for Huntington's disease was cloned[1]. Identification of the gene was itself the cause for high excitement, which grew all the more intense with the news that it harbours yet another example of expanding trinucleotide repeat sequences. Such sequences had already been implicated in three other neurological disorders — fragile X syndrome, myotonic dystrophy and spinobulbar muscular atrophy (SBMA)[2].

In a matter of just a couple of years, triplet repeats have become the most talked about phenomenon in human genetics[3]. Whether they will be confined to neurological disorders remains to be seen — speculation persists that they may underlie diseases such as breast cancer — but the latest example to join the list is another classic neurological disease. Writing in this month's *Nature Genetics*, Harry Orr, Huda Zoghbi and colleagues show that an expanding CAG repeat is associated with spinocerebellar ataxia type 1 (SCA1)[4], and the way in which they made the discovery may expedite the search for similar disease genes.

The hereditary spinocerebellar ataxias are a genetically heterogeneous group of disorders, of which SCA1 is a prevalent form. The disease is characterized by neurodegeneration of the cerebellum, spinal cord and brainstem, and, like Huntington's disease, it becomes evident only in adulthood. Death usually results 10–20 years after onset of the disease. The gene for SCA1 was mapped to the short arm of chromosome 6 almost 20 years ago, and as the region containing the gene had been cloned there was little doubt that a search of the megabase or so of DNA using conventional positional cloning techniques would eventually give up the gene.

But faced with a quest that could take years, Orr *et al.* decided to attempt a short cut. They reasoned that as SCA1 exhibits anticipation — one of the hallmarks of triplet repeat disorders in which the illness becomes progressively more severe in successive generations, as gauged by earlier onset — it could well be another triplet-repeat disorder and be identifiable by that characteristic. Indeed the effectiveness of such a direct approach had already been demonstrated by Caskey and colleagues in cloning the myotonic dystrophy gene[5].



Correlation between expansion of CAG triplet repeat and age-of-onset in spinocerebellar ataxia (SCA1). Note the lack of overlap between repeat lengths of healthy and affected individuals (see ref. 4 for further details).

So Orr *et al.* screened cosmids from the critical region with oligonucleotide probes designed to detect stretches of triplet repeats, and — lo and behold — isolated a small genomic sequence that hybridized to the CAG-repeat probe. When they examined the size of this repeat in families with SCA1, they found the distinctive pattern of repeat expansion in affected members reminiscent of the CAG expansions in Huntington's disease. In looking at more than 30 individuals with SCA1, Orr *et al.* found that affected members possess about twice the number of repeats (43–81) as do healthy unaffected individuals (25–36). Interestingly, in the cases examined so far, there is no overlap between the top end of the normal spectrum and the lower values in the affected range.

Orr *et al.* also examined the correlation between the number of repeats and the severity of the disease, as indicated by the age-of-onset. Patients with the juvenile-onset form of SCA1 tended to inherit larger expansions (typically 59–81) than those with later-onset disease (at least 43 repeats) (see figure). Although the authors have not yet sequenced the entire gene that contains the CAG repeats, they do show that the triplet repeat is detected in a ten-kilobase RNA transcript found chiefly in brain and skeletal muscle. Moreover, the repeat is present in an open-reading frame in the genomic sequence. All in all, it looks as if the repeat is translated as a putative polyglutamine stretch in the corresponding protein, as is the case in SBMA and Huntington's disease.

This new discovery begs any number of questions, of which two stand out. First, how many more triplet-repeat disorders are yet to be identified? There could well be quite a few, and the success of at least two groups in searching directly for such repeats[4,5] means that this is likely to become a popular strategy for tracking down the genes concerned. Two regions that are high on the list for examination in this way are described in a couple of other papers in this month's *Nature Genetics*: two other autosomal dominant ataxias which exhibit variable age-of-onset, SCA type 2 and Machado–Joseph disease, have now been mapped by linkage to the long arm of chromosomes 12 and 14, respectively[6,7]. Another method which may prove useful is called repeat expansion detection (RED)[8]. This technique uses the polymerase chain reaction to amplify large-scale expanded triplet repeats directly from genomic DNA, and has already been used to identify a variable CTG sequence on chromosome 18 (although the clinical consequences of this expansion are unclear).

The other question concerns the biochemical mechanisms by which trinucleotide expansions result in disease. On that we are almost completely in the dark, and it is where progress is now most sorely needed.

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Also in this month's *Nature Genetics*: sequence characterization of 3,400 complementary DNAs expressed in human brain; identification of the *Little* murine mutation; mammalian recombination genes homologous to *recA* and *RAD51*; defects in rhodopsin giving rise to stationary night blindness; a ribosomal RNA mutation causing non-syndromic deafness; and the healing of telomeres.

1. The Huntington's Disease Collaborative Research Group *Cell* **72**, 971–983 (1993).
2. Mandel, J.-L. *Nature Genet.* **4**, 8–9 (1993).
3. Morel, V. *Science* **260**, 1422–1423 (1993).
4. Orr, H.T. *et al. Nature Genet.* **4**, 221–226 (1993).
5. Fu, Y.-H. *et al. Science* **255**, 1253–1255 (1992).
6. Gispert, S. *et al. Nature Genet.* **4**, 295–299 (1993).
7. Takiyama, Y. *et al. Nature Genet.* **4**, 300–304 (1993).
8. Schalling, M., Hudson, T.J., Buetow, K.H. & Housman, D.E. *Nature Genet.* **4**, 135–139 (1993).