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The cause of Canavan's disease

Characterization of genetic defects involving myelin continues with the identification of the gene for Canavan's disease, a condition that results in the destruction of the brain's white matter.

ONCE an esoteric field confined to the neurological textbooks, the leukodystrophies continue to make the news. Not long after the release earlier this year of *Lorenzo's Oil*, a powerful film about a dietary treatment for the fatal demyelinating condition known as X-linked adrenoleukodystrophy (ALD), the gene for that disease was identified by positional cloning[1]. In this month's *Nature Genetics*[2], Reuben Matalon, Rajinder Kaul and colleagues describe the cloning of the gene for a related autosomal recessive disorder called Canavan's disease, or spongy degeneration of the brain.

Canavan's disease, which was first described more than 60 years ago, primarily affects infants of Jewish origin. Many Jewish families with the disease have been traced back to founders in Lithuania and northern Poland. This region overlaps closely with the area in which Tay–Sachs disease, which is also common among Ashkenazi Jews, is thought to have arisen. Canavan's disease also seems to be unusually frequent in Saudi Arabia. Although its severity varies considerably — some patients die before the age of one, others live into their twenties — most patients die within a few years of birth. Mental retardation, muscle hypotonia (especially in the neck), macrocephaly and blindness are all common symptoms. At the histological level, the white matter of the brain shows severe demyelination and vacuolation, giving a customary spongy appearance.

For the past five years, there has been little doubt as to the identity of the gene for Canavan's disease. Matalon *et al.*[3] showed that levels of *N*-acetylaspartic acid (NAA) in patients' urine were up to 200 times higher than normal and, by way of explanation, that the activity of the enzyme aspartoacylase, which processes NAA, was deficient in cultured fibroblasts from patients. Subsequent studies showed that levels of NAA, which is a normal component of brain neurons, are greatly increased in the brain as well[4]. However, as seen in ALD, circumstantial biochemical clues can sometimes be misleading (the primary defect in ALD turned out not to be in a key enzyme, as had been widely suspected, but in a putative peroxisomal transporter[1]). Indeed, formal proof of a genetic defect in aspartoacylase required Kaul *et al.*[2] to adopt a traditional approach: the laborious purification of bovine aspartoacylase, which then allowed them to isolate bovine complementary DNA clones, and eventually the human cDNA, which predicts a protein of 313 amino acids.

With the cDNA clones in hand, obtaining proof that the aspartoacylase gene is mutated in Canavan's disease came quickly. The first few patients examined were found to be homozygous for the same missense mutation — a glutamine to alanine substitution at residue 285. Taking advantage of a new restriction enzyme site created by the single nucleotide change, Kaul *et al.* screened a further 17 patients and found that 29 out of 34 (85 per cent) aspartoacylase alleles had the same missense mutation. Glutamine 285 is a conserved residue which the authors speculate forms part of an esterase-like catalytic site, and when mutated neatly explains the abolition of aspartoacylase hydrolytic activity.

The frequency of the common mutation in Canavan's disease points to a founder effect in the Jewish population, in agreement with earlier theories of the origin of the disease. Indeed, the new molecular data will help researchers to confirm whether recent estimates of the carrier frequency of Canavan's disease, perhaps as high as 1 in 30 among Ashkenazi Jews, are correct. Kaul *et al.* have recently identified two further aspartoacylase mutations, which will shed light on the possible relationship between specific mutations and the variable phenotypes that result. And just as Jewish couples contemplating marriage are routinely tested for Tay–Sachs disease, it will soon be possible — and advisable — for them to be screened for Canavan's disease once 95 per cent of the mutant alleles have been identified. On an equally practical level, a standard polymerase chain reaction assay will rapidly replace the former means of prenatal diagnosis, which involved attempts at measuring the very low aspartoacylase levels from cultured amniocytes.

Why should the failure to maintain normal levels of NAA in the brain produce the devastating effects on myelin, and why are they confined to the white matter? There are no good answers at present. Decreases in NAA are correlated with a number of disorders, including multiple sclerosis, neurodegenerative disorders and stroke, as monitored using nuclear magnetic resonance[5], but these may be secondary manifestations. Interestingly, NAA levels are also increased in the grey matter in patients with Canavan's disease[6], but this is spared any pathological damage.

Although much about the onset of the disease remains an enigma, the identification of the gene defect is yet another valuable piece in the myelin puzzle[7]. Several mutations in another form of X-linked leukodystrophy, Pelizaeus–Merzbacher disease, occur in a major myelin component called the proteolipid protein, and the enzyme defects in other leukodystrophies have been identified. In the peripheral nervous system, mutations in the peripheral myelin protein 22 gene and myelin protein zero give rise to the common hereditary neuropathies, Charcot–Marie–Tooth disease types 1A[8,9] and 1B[10,11], respectively. Moreover, both genes have also undergone mutation in cases of a more severe condition, DeJerine–Sottas disease[12,13].

With the efficacy of Lorenzo's oil proving to be disappointing in clinical trials for a mild form of ALD[14], hope for genuine therapies surely rests with the insights into myelin synthesis, structure and turnover that molecular genetics and the ensuing animal models are likely to provide.

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Also in this month's *Nature Genetics*: imprinting relaxation in Beckwith–Wiedemann syndrome; *de novo* mutations in Huntington's disease; screening for p53 mutations in yeast; mapping the locus for familial spastic paraplegia; and gene therapy advances in models for cystic fibrosis and muscular dystrophy.

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