Journal of
NEUROLOGY
NEUROSURGERY
& PSYCHIATRY

Editorial

The DNA laboratory and neurological practice

Molecular genetic techniques have already made an impact on neurological practice, leading to improved diagnosis and genetic counselling in inherited neurological disorders. The purpose of this review is to outline what the DNA laboratory can currently provide the neurologist, in terms of services to patients with neurological disease and their families.

The first neurological disease to be assigned to a specific chromosomal region by means of random linkage analysis using variable DNA markers (polymorphisms) was Huntington's disease (HD).[1] Since then the neurological disease map has become much more dense[2] (table 1). Also, several gene defects have been identified, either after gene mapping and subsequent gene isolation (reverse genetics, now known as positional cloning), such as, hereditary motor and sensory neuropathy (HMSN type Ia)[3,4] or by direct analysis of candidate genes, for example, in some cases of familial Alzheimer's disease[5] and inherited prion diseases[6] (table 2). The small genome found in mitochondria has also been established as a mutation hot spot for some neurological diseases, a possibility suggested by their biochemical features and predominantly maternal mode of transmission.[7]

Gene tracking

There are two general ways of using DNA analysis in clinical practice, and it is important to distinguish between them. The most common involves gene tracking using linked markers. This is required when a gene has been localised to a specific chromosomal region, but the defective gene itself has not been isolated. This approach currently applies most commonly in HD. It is possible to predict which subjects at risk of HD are likely to carry the gene, but only if there are sufficient family members available to determine which allele of a linked polymorphism is segregating with the disease. This usually requires investigating members of three generations of a family if DNA from only one affected member is available, or two generations if more than one patient is alive.[8] As there is a small chance of recombination (crossing over during meiosis) occurring between the linked marker and the HD gene, predictive testing in these circumstances is, at best, about 98% accurate. In smaller pedigrees it is possible to do prenatal exclusion testing for those at risk who are either unable (because of limited pedigree structure), or unwilling, to have predictive testing, but wish to avoid passing on the HD gene to their children.[9] Predictive and prenatal testing is performed by most regional genetic units and their associated DNA laboratories in the UK, most of which participate in a HD predictive testing consortium.[10] To date, over 200 predictive tests have been performed by members of this consortium, so far with few adverse sequelae. This probably relates to the provision of detailed pre-test genetic counselling and post-test support. The test is available only with the informed consent of the individual at risk, and only to those aged 18 years or over. It should be stressed that there is as yet no direct genetic test for HD, so DNA analysis is not useful to confirm or exclude the diagnosis. However, DNA from all patients with definite or probable HD should be stored in regional DNA laboratories for future direct analysis or use in gene tracking in their families. When the HD mutation is identified, studies of seemingly sporadic cases of HD will be particularly useful for their relatives.

The same limitations of gene tracking apply to other autosomal dominant disorders in which the disease gene has been mapped but not isolated, such as, von Hippel-Lindau disease[11] and neurofibromatosis type 2 (bilateral acoustic neurofibromatosis).[12] Prenatal diagnosis in neurofibromatosis type 1 (von Recklinghausen's disease) is also often based on linked markers as it is extremely time consuming and labour intensive to screen the gene for the very large number of mutations reported.[13] In autosomal recessive disorders which have been mapped reasonably precisely, for example, acute infantile and chronic childhood spinal muscular atrophy,[14] Friedreich's ataxia,[15] and Wilson's disease,[16] it is possible to perform prenatal diagnosis for couples who have already had one affected child. This means that it is important to make an accurate diagnosis in the index patient, and store DNA, as early as possible. In Friedreich's ataxia, many families will be complete by the time the first affected child is diagnosed. Couples who may benefit from prenatal diagnosis should be referred for genetic counselling and molecular analysis of the family before embarking on subsequent pregnancies. Carrier testing in such families is of limited value as it is currently not possible to determine carrier status for any of these disorders in the general population, and the risk of the healthy brothers and sisters of patients having affected children is in any case extremely low.

Direct mutation detection

Amyloidoses

Direct mutation detection is possible in three disorders

Table 1 A gene map for childhood or adult onset neurological disease

Disease	*Chromosome/location*	*Disease*	*Chromosome/location*
HMSN type 1 (some families)	1q2	Hyperkalaemic periodic paralysis	17q22-q24
Nemaline myopathy	1q21-q23	Amyloid neuropathies, TTR-related	18q11·2-q12·1
Juvenile ALS	2q33–35	Myotonic dystrophy	19p13·3
Cerebrotendinous xanthomatosis	2q33-ter	Central core disease	19q12-q13
von Hippel-Lindau disease	3p26-p25	Malignant hyperthermia	19q12-q13·2
Huntington's disease	4p16·3	FAD (late)	19
Facioscapulohumeral dystrophy	4q35	Benign neonatal convulsions	20q
Acute and chronic childhood SMA	5q11·2-q13·3	Inherited prion diseases	20pter-p12
LGD, dominant	5q22·3–31·3	Icelandic amyloid angiopathy	20p11·22–p11·21
Juvenile myoclonic epilepsy	6p	FAD (early, some families)	21pter-q21
Dominant ataxia (some families)	6p24-p23	Familial ALS	21q22·1-q22·2
Friedreich's ataxia	9q13	Unverricht-Lundborg disease	21q22·3
Amyloidosis, Finnish	9q33	Dutch cerebral haemorrhage	21q21
Tuberous sclerosis (some families)	9q34·1-q34·2	Neurofibromatosis	22q11-q13·1
Torsion dystonia (some families)	9q34	Aicardi syndrome	Xp22
Tuberous sclerosis (some families)	11q14-q23	Duchenne/Becker dystrophies	Xp21·2
Ataxia telangiectasia	11q23	X-linked HMSN	Xq11-q13
Amyloid neuropathy, Iowa type	11q23-q24	X-linked spastic paraplegia	Xq12-q22
Acute intermittent porphyria	11q23·2-qter	Dystonia/parkinsonism	Xq21
Wilson's disease	13q14·2-q21	Bulbospinal neuronopathy	Xq21·3-q22
FAD (early, some families)	14q	Fabry's disease	Xq22
LGD, recessive	15q15-q22	Pelizaeus-Merzbacher disease	Xq21·3-q22
Batten's disease	16p	Fragile X syndrome	Xq27·3
Tuberous sclerosis (some families)	16p13	Emery-Dreifuss muscular dystrophy	Xq27·3-q28
HMSN type 1 (most families)	17p11·2	Centronuclear myopathy	Xq27-q28
Neurofibromatosis 1	17q11·2	Adrenoleukodystrophy	Xq28
Paramyotonia congenita	17q22-q24		

p, q represent short and long arms respectively, numbers chromosomal bands; SMA spinal muscular atrophy; LGD

characterised by deposition of the gene product in the form of amyloid. The first of these is a heterogeneous group of diseases causing familial amyloid neuropathy (FAP). The most common genetic abnormality is a point mutation in the transthyretin (TTR, previously known as pre-albumin) gene. At least 20 pathogenic mutations of this gene have been described, and most give rise to a progressive small fibre peripheral neuropathy causing distal sensory loss, usually accompanied by autonomic dysfunction.[17] The most common mutation causes rise to a substitution of methionine for valine at position 30 of the TTR molecule, and is associated with the type of FAP seen most frequently in individuals of Portuguese descent. This disorder, however, is not confined to this racial group, or areas of the world where Portuguese genes are common, and the disorder has been observed in individuals of French and British descent.[18] The so called German (tyrosine 77) mutation has also been observed in Britian and France, and a cluster of cases with late onset amyloid neuropathy and the alanine 60 mutation appears to have originated in northwest Ireland.[18 19] In these types of TTR-related amyloid neuropathies, and also others, it is clear that penetrance is incomplete, and TTR gene analysis is a useful diagnostic procedure in patients with the appropriate phenotype, regardless of ethnic background or family history. Apart from the diagnostic value of this investigation, and the implications of a positive result for genetic counselling of the patient's family, there are preliminary data suggesting that liver transplantation is an effective form of therapy for TTR-related amyloidosis,[20] and this is likely to be of most benefit in early cases.

A small proportion of families with familial Alzheimer's disease has one of several point mutations in the gene encoding the amyloid precursor protein which maps to chromosome 21.[5] It is clear, however, that this is an unusual cause of familial Alzheimer's disease and such mutations have not so far been reported in sporadic cases of Alzheimer's disease.[21] A different mutation in the same gene gives rise to Dutch hereditary cerebral haemorrhage.[22] Prion diseases are also characterised by the deposition of amyloid, derived from prion protein, in the brain; several point mutations and two insertions in this gene have been described in association with a wide variety of phenotypes.[6] These range from the Gerstmann-Sträussler syndrome of slowly progressive dementia and ataxia, usually of autosomal dominant inheritance, to families exhibiting the typical features of Creutzfeldt-Jakob disease (CJD). The last applies to two clusters of cases of CJD, among Libyan Jews and in Slovakia, both of which are associated with a point mutation of the prion protein gene in codon 200. A recent report has described this mutation in familial CJD in a British family.[23] One of the diagnostic problems in this group of disorders is that the phenotype may be relatively nonspecific, comprising slowly progressive dementia without myoclonus or other characteristic features of spongiform encephalopathy, and the characteristic pathology may be lacking.[6 24] Thus prion protein gene analysis is a useful diagnostic procedure in such patients, particularly if there is a positive family history; it can also be used for presymptomatic and prenatal diagnosis.[25] The incidence of mutations in sporadic cases of CJD is not clear as yet, but seems to be about 5% in the UK (J Collinge, personal communication).

Table 2 Inherited neurological diseases in which gene defects have been identified

Disease	*Genetic defect*
Nuclear gene defects:	
Amyloid neuropathies:	
TTR (~20 types)	Point mutations
Apolipoprotein AI-related	Point mutation
Gelsolin-related	Point mutation
Neurofibromatosis type 1	Insertion, deletions, point mutations
HMSN type Ia	Duplication
Facioscapulohumeral dystrophy	? contraction
Hyperkalaemic periodic paralysis	Point mutations
Hexosaminidase deficiencies	Deletion, insertions, point mutations
Dutch cerebral haemorrhage	Point mutation
Familial Alzheimer's disease	Point mutations
Prion diseases	Insertions, point mutations
Myotonic dystrophy	Expansion
Malignant hyperthermia	Point mutation
Duchenne/Becker dystrophies	Deletions (70%), duplications
Bulbospinal neuronopathy	Expansion
Fragile X syndrome	Expansion
Mitochondrial DNA defects:	
Leber's optic atrophy	Point mutations
MERRF, MELAS, myopathy etc.	Point mutations
Ataxia, RP, neuropathy	Point mutation
PEO, KSS	Deletions, duplications

RP retinitis pigmentosa; PEO progressive external ophthalmoplegia; KSS

Diseases associated with unstable repeat sequences

In myotonic dystrophy and in two disorders which map to the X chromosome, the gene defect is characterised by an excessively large and unstable trinucleotide repeat within a gene which exists in normal individuals but is greatly expanded in patients. Detection of the myotonic dystrophy gene expansion appears to be a reliable means of determining which family members are affected.[26] This has enhanced genetic counselling in this disorder enormously as exclusion of carrier status with certainty in this disease was previously very difficult, even using EMG and slit lamp examinations. Prenatal diagnosis is also possible using DNA analysis. The variable expansion seen within the myotonic dystrophy gene has provided an explanation for anticipation observed in myotonic dystrophy pedigrees, as there is a tendency for the expansion to increase in size in successive generations, in parallel with increasing disease severity.

The two X-linked disorders associated with expanded trinucleotide repeats are the fragile X mental retardation syndrome[27] and X-linked bulbospinal neuronopathy (Kennedy's syndrome).[28 29] The latter may be confused with amyotrophic lateral sclerosis (ALS), particularly if there is no history of affected relatives as is the case in about half of patients. This syndrome produces generalised neurogenic muscle weakness, beginning typically between the ages of 30 and 50; there is associated bulbar dysfunction with prominent fasciculation of the tongue and often the face. In contrast to ALS, the tendon reflexes are usually depressed or absent and there is an associated axonal sensory neuropathy in most patients, but usually without clinical sensory loss.[28] The expanded trinucleotide repeat is in the androgen receptor gene,[29] a candidate gene for this disorder because of the fairly frequent occurrence of gynaecomastia and infertility observed in this syndrome, but these features are not universal. Accurate diagnosis is important as this disorder has a relatively good prognosis; all the daughters of a patient are carriers and therefore at risk of having affected sons, and there may be other females in the family who, unknowingly, are also carriers.

Hereditary motor and sensory neuropathy (HMSN) type I

HMSN type I is genetically heterogeneous, but this autosomal dominant demyelinating neuropathy maps to chromosome 17 in the majority of families (type Ia). In 1991, two groups showed independently that the disease is associated with a large (at least 1 megabase) duplication of the proximal short arm of chromosome 17.[3 4] This duplication is not visible on cytogenetic analysis, but is detectable using molecular genetic techniques. It has been shown that a high proportion of seemingly sporadic cases of HMSN type I (that is, with normal parents) have the duplication, indicating that it occurs as a fresh mutational event fairly frequently.[30] Thus detection of the duplication is not only useful for confirming the diagnosis of HMSN type Ia in families, and providing the option of prenatal diagnosis, but also in aiding the differential diagnosis of chronic demyelinating neuropathy in the absence of a family history. This has important clinical implications, for example, in distinguishing HMSN type Ia from chronic inflammatory demyelinating polyneuropathy.

Muscular dystrophies

A further large gene rearrangement has recently been reported in facioscapulohumeral (FSH) muscular dystrophy, following localisation of the disease locus to the distal long arm of chromosome 4.[31 32] This rearrangement appears to be a contraction of a tandem repeat, and also occurs *de novo* in cases of FSH dystrophy without similarly affected relatives.[32] Detection of this abnormality has immediate clinical applications in diagnosis and genetic counselling.

The use of dystrophin gene analysis in the diagnosis of Duchenne and Becker muscular dystrophies (BMD) has been reviewed elsewhere,[33 34] but it should be stressed that defects of the dystrophin gene may be seen in a wide variety of phenotypes, including the relatively benign syndrome of muscle cramps and calf hypertrophy[35] and in patients previously thought to have limb girdle dystrophy (LGD), with or without characteristic features such as hypertrophy of the calf muscles.[36] The diagnosis of dystrophinopathy is of particular importance in patients, particularly males, without a history of affected relatives. The distinction between BMD and LGD is of great importance in genetic counselling. About half of patients with BMD have a deletion of a particular part of the dystrophin gene. The majority of the other half presumably have more subtle gene defects such as point mutations which are not detectable during routine DNA analyses. The investigation of choice in such individuals is dystrophin analysis of muscle biopsies using immunohistochemistry or Western blot analysis.[33 34]

Mitochondrial diseases

Defects of mitochondrial DNA (deletions and point mutations) have been described in two groups of disorders, mitochondrial encephalomyopathies and Leber's hereditary optic neuropathy.[7] Deletions of muscle mitochondrial DNA occur in approximately half of patients with progressive external ophthalmoplegia and ragged-red fibres on muscle biopsy, and most patients with the Kearns-Sayre syndrome.[37] These are not readily detected in blood samples and thus require investigation of muscle DNA. The demonstration of a deletion confers a very low recurrence risk to family members and their offspring. The two most common mitochondrial DNA point mutations observed in mitochondrial encephalomyopathies are detectable in blood samples in the vast majority of cases,[39] and are associated with the syndromes of myoclonic epilepsy with ragged red fibres (MERRF) and mitochondrial encephalopathy with lactic acidosis and stroke-like episodes (MELAS). Index patients from families with MERRF, which is commonly maternally inherited, usually have the classic triad of myoclonus, ataxia and seizures, but their relatives may be oligosymptomatic, with solitary features such as occasional seizures. Not all patients with the MERRF syndrome have ragged red fibres on muscle biopsy and in many, muscle weakness is slight or absent. Blood mitochondrial DNA analysis is a useful screening procedure for mitochondrial disease in patients with progressive myoclonic syndromes and arguably should be performed before muscle biopsy. The contribution of this mitochondrial DNA mutation to epilepsy as a whole is as yet unclear, but it is likely to be small.

The mitochondrial DNA point mutation at position 3243 is seen in the majority of patients with MELAS, but it has also been described in association with other mitochondrial encephalomyopathies, such as dementia, ataxia and deafness, and even myopathy alone.[38] Again, ragged red fibres are not seen in all patients with this mutation and neurological disease. Screening of blood mitochondrial DNA for the 3243 mutation is a useful diagnostic procedure in patients with suspected mitochondrial encephalomyopathy, particularly the MELAS syndrome; it should be considered in patients who have more than one stroke-like event without obvious cause under the age of about 45 years, particularly if there are other features such as deafness or short stature. The stroke-like

episodes of mitochondrial disease often have characteristic features with migrainous elements including headache, nausea and vomiting, malaise, visual disturbances, and frequently a focal seizure preceding the neurological deficit.

Leber's hereditary optic neuropathy is associated with at least three pathogenic point mutations of mitochondrial DNA, the most common of which is at position 11778 base pairs (bp), the second most frequent being at position 3460 bp.[39 40] In Europe, these two mutations account for 90% or more of families with this disease, as defined by the typical clinical picture (subacute optic neuropathy with a positive maternal family history). In the presence of the 11778 mutation the prognosis for useful recovery of visual function is poor.[40] A further mitochondrial DNA mutation, at position 13708 bp, has been observed in a small number of families with LHON, but the significance of this is uncertain as it also occurs in the normal population.[41] Studies of patients with the 11778 mutation have broadened our view of the phenotype of this disorder, and it is clear that some patients have onset of visual symptoms quite late in life, occasionally not until the seventh decade, and in some the characteristic fundal appearances of microangiopathy are not present at onset of symptoms.[40 42] About 50% of patients with a clinical picture of LHON and a pathogenic mitochondrial DNA mutation do not have a history of similarly affected relatives, and it is clear that detection of pathogenic mutations is extremely useful diagnostically in this context.[40] Investigation of maternally related relatives will show that most, if not all, of them will have high amounts of mutant mitochondrial DNA but genetic counselling is complex as it is clear that not all males at risk who have the relevant mutation will develop symptoms, and not all genotypically similar female carriers will transmit the disease to their sons. An interesting association between a multiple sclerosis-like illness with severe bilateral optic neuropathy at onset has been observed in females from LHON families and the 11778 mutation.[43]

Practical issues

Nearly all the analyses referred to above require the study of 10 (preferably 20) mls of blood in EDTA tubes which should be received fresh (not frozen) by the relevant laboratory within a maximum of 4 to 5 days after sampling, preferably after discussion between the clinician and laboratory staff. Investigations requiring gene tracking, for example in Huntington's disease, should be done in conjunction with the appropriate regional clinical genetics unit which can provide counselling for relevant family members, collection of samples and their analysis. Dystrophin gene analyses are routinely available in most regional DNA laboratories. Those for the myotonic dystrophy mutation are currently less widely available, but this situation is likely to change in the next year or so. Direct mutation detection of some of the more rare disorders (for example, amyloidoses, prion diseases, HMSN type Ia, X-linked bulbospinal neuronopathy and mitochondrial DNA diseases) are performed in only a few centres; regional genetic units and the DNA Service Laboratory at the Institute of Neurology can advise about these. In the case of positive results, it is essential to take into account the potential needs of relatives in terms of information and genetic advice, as identification of a genetic disease in a seemingly sporadic disorder has implications for the whole family.

I thank Drs EM and PD Thompson for helpful discussion.

AE HARDING

University Department of Clinical Neurology (Neurogenetics Section), Institute of Neurology, London, UK

Correspondence to: Professor Harding, University Department of Clinical Neurology (Neurogenetics Section), Institute of Neurology, Queen Square, London WC1N 3BG, UK.

1 Gusella JF, Wexler NS, Conneally PM, *et al.* A polymorphic DNA marker genetically linked to Huntington's disease. *Nature* 1983;**306**:234–8.
2 Human Gene Mapping 11. *Cytogenet Cell Genet* 1991;**58**:1–2197.
3 Raeymakers P, Timmerman V, Nelis E *et al.* Duplication in chromosome 17p11·2 in Charcot-Marie-Tooth neuropathy type 1a (CMT 1a). *Neuromuscular Disorders* 1991;**1**:93–98.
4 Lupski JR, Montes de Oca-Luna R, Slaugenhaupt S, *et al.* DNA duplication associated with Charcot-Marie-Tooth disease type IA. *Cell* 1991; **66**:219-32.
5 Hardy J. Framing beta-amyloid. *Nature Genetics* 1992;**1**:233–4.
6 Collinge J. Inherited prion diseases. In: Harding AE, Deufel T, eds. *Inherited Ataxias*. New York: Raven Press, 1992: (in press).
7 Wallace DC. Diseases of the mitochondrial DNA. *Annu Rev Biochem* 1992;**61**:1175–212.
8 Morris MJ. Huntington's disease: presymptomatic testing. *Curr Opinion Neurol Neurosurg* 1990;**3**:337–41.
9 Tyler A, Quarrell OWJ, Lazarou LP, Meredith AL, Harper PS. Exclusion testing in pregnancy for Huntington's disease. *J Med Genet* 1990;**27**: 488–95.
10 Tyler A, Ball D, Crawfurd D, *et al.* Presymptomatic testing for Huntington's disease in the United Kingdom. *BMJ* 1992;**304**:1593–6.
11 Maher ER, Bentley E, Yates JRW, *et al.* Mapping of the von Hippel-Lindau disease locus to a small region of chromosome 3p by genetic linkage analysis. *Genomics* 1991;**10**:957–60.
12 Narod SA, Parry DM, Parboosingh J, *et al.* Neurofibromatosis type 2 appears to be a genetically homogeneous disease. *Am J Hum Genet* 1992;**51**:486–96.
13 Gutmann DH, Collins FS. Recent progress toward understanding the molecular biology of Von Recklinghausen neurofibromatosis. *Ann Neurol* 1992;**31**:555–61.
14 Daniels RJ, Thomas NH, MacKinnon RN, *et al.* Linkage analysis of spinal muscular atrophy. *Genomics* 1992;**12**:335–9.
15 Wallis J, Shaw J, Wilkes D, *et al.* Prenatal diagnosis for Friedreich's ataxia. *Am J Med Genet* 1989;**34**:458–61.
16 Farrer LA, Bowcock AM, Hebert JM, *et al.* Predictive testing for Wilson's disease using tightly linked and flanking DNA markers. *Neurology* 1991;**41**:992–9.
17 Benson MD. Inherited amyloidosis. *J Med Genet* 1991;**28**:408–26.
18 Bhatia K, Reilly M, Adams D, *et al.* Transthyretin gene mutations in British and French patients with amyloid neuropathy. *J Neurol Neurosurg Psychiatry* 1993(in press).
19 Staunton H, Davis MB, Guiloff RJ, Nakazato M, Miyazato N, Harding AE. Irish (Donegal) amyloidosis is associated with the transthyretin ALA 60 (Appalachian) variant. *Brain* 1991;**114**:2675–9.
20 Holmgren G, Steen L, Ekstedt J, *et al.* Biochemical effect of liver transplantation in two Swedish patients with familial amyloidotic polyneuropathy (FAP-met 30). *Clin Genet* 1991;**40**:242–6.
21 Tanzi RE, Vaula G, Romano DM, *et al.* Assessment of amyloid beta-protein precursor gene mutations in a large set of familial and sporadic Alzheimer disease cases. *Am J Hum Genet* 1992;**51**:273–82.
22 Levy E, Carman MD, Fernandez-Madrid IJ, *et al.* Mutation of the Alzheimer's disease amyloid gene in hereditary cerebral hemorrhage, Dutch type. *Science* 1990;**248**:1124–6.
23 Collinge J, Palmer MS, Campbell T, *et al.* Inherited prion disease (PrP lysine 200) in the UK. *BMJ* (in press).
24 Collinge J, Brown J, Hardy J *et al.* Inherited prion disease with 144 base pair gene insertion. II. Clinical and pathological features. *Brain* 1992;**115**:687–710.
25 Collinge J, Poulter M, Davis MB, *et al.* Presymptomatic detection or exclusion of prion protein gene defects in families with inherited prion diseases. *Am J Hum Genet* 1991;**49**:1351–4.
26 Suthers GK, Huson SM, Davies KE. Instability versus predictability: the molecular diagnosis of myotonic dystrophy. *J Med Genet* 1992;**29**: 761–5.
27 Rousseau F, Heitz D, Biancalana V, *et al.* Efficient and reliable direct diagnosis of the fragile X mental retardation syndrome. *N Engl J Med* 1991;**325**:1673–81.
28 Harding AE, Thomas PK, Baraitser M, Bradbury PG, Morgan-Hughes JA, Ponsford JR. X-linked recessive bulbospinal neuronopathy: a report of ten cases. *J Neurol Neurosurg Psychiatry* 1982;**45**:1012–9.
29 La Spada AR, Wilson EM, Lubahn DB, Harding AE, Fischbeck KH. Androgen receptor gene mutations in X-linked spinal and bulbar muscular atrophy. *Nature* 1991;**352**:77–9.
30 Hoogendijk JE, Hensels GW, Gabreels-Festen AAWM, *et al.* De-novo mutation in hereditary motor and sensory neuropathy type I. *Lancet* 1992;**339**:1081–2.
31 Upadhyaya M, Lunt PW, Sarfarazi M, *et al.* A closely linked DNA marker for facioscapulohumeral disease on chromosome 4q. *J Med Genet* 1991;**28**:665–71.
32 Wijmenga C, Hewitt JE, Sandkuijl LA, *et al.* Chromosome 4q rearrangements associated with facioscapulohumeral muscular dystrophy. *Nature Genetics* 1992;**2**:26–30.
33 Beggs AH, Kunkel LM. Improved diagnosis of Duchenne/Becker muscular dystrophy. *J Clin Invest* 1990;**85**:613–9.
34 Worton RG, Gillard EF. Duchenne muscular dystrophy. In: Connealy PM, ed. *Molecular basis of neurology*. Oxford: Blackwell Scientific, 1993:73–112.
35 Gospe SM, Lazaro RP, Lava NS, Grootscholten PM, Scott MO, Fischbeck KH. Familial X-linked myalgia and cramps: a non-progressive myopathy associated with a deletion in the dystrophin gene. *Neurology* 1989;**39**:1277–80.
36 Arikawa E, Hoffman EP, Kaido M, Nonaka I, Sugita H, Arahata K. The frequency of patients with dystrophin abnormalities in a limb-girdle patient population. *Neurology* 1991;**41**:1491–6.
37 Holt IJ, Harding AE, Cooper JM, *et al.* Mitochondrial myopathies: clinical and biochemical features in 30 cases with major deletions of muscle mitochondrial DNA. *Ann Neurol* 1989;**26**:699–708.

38 Hammans SR, Sweeney MG, Brockington M, Morgan-Hughes JA Harding AE. Mitochondrial encephalopathies: molecular genetic diagnosis from blood samples. *Lancet* 1991;**337**:1311–13.
39 Newman NJ, Lott MT, Wallace DC. The clinical characteristics of pedigrees of Leber's hereditary optic neuropathy with the 11778 mutation. *Am J Ophthalmol* 1991;**111**:750–62.
40 Huoponen K, Vilkki J, Aula P, Nikoskelainen EK, Savontaus M-L. A new mtDNA mutation associated with Leber hereditary optic neuroretinopathy. *Am J Hum Genet* 1991;**48**:1147–53.
41 Johns DR, Berman, J. Alternative, simultaneous complex I mitochondrial DNA mutations in Leber's hereditary optic neuropathy. *Biochem Biophys Res Com* 1991;**174**:1324–30.
42 Borruat F-X, Green WT, Graham EM, Sweeney MG, Morgan-Hughes JA, Sanders MD. Late onset Leber's optic neuropathy: a case confused with ischaemic optic neuropathy. *Brit J Ophthalmol* 1991;**76**:571–573.
43 Harding AE, Sweeney MG, Miller DH, *et al.* Occurrence of a multiple sclerosis-like illness in women who have a Leber's hereditary optic neuropathy mitochondrial DNA mutation. *Brain* 1992;**115**:979–89.

Neurological stamp

Carolus Linnaeus (Carl Linné) 1707–78

Linnaeus' lasting contribution was his botanical classification. He began studying medicine at the University of Lund in 1727. In 1738 after returning from Holland, where he wrote his doctoral thesis, he established himself as a physician in Stockholm, where he became royal physician and Admiralty doctor before moving to Uppsala. He became professor of medicine there in 1741 and a year later he was appointed to the Chair of Botany, a position which he retained for the rest of his life.

Linnaeus was probably the first to describe the classic symptoms of aphasia. He was also aware of involuntary movements and he established a vogue for classification in medicine. His book *Systema Pantarum* (1753) contains the final form of his classification of plants. The book *Systems Natura* (1735) classified more than 4000 animals. It was Linnaeus who invented the term *homo sapiens*. He was also the first to use the now commonly accepted signs for male and female.

Linnaeus was knighted by the Swedish Government in 1761 in recognition of his work. Shortly after, he officially changed his name to Carl von Linné. In 1774 he had his first premonition of death—an apoplectic stroke. He died in 1778 and is buried in the beautiful cathedral at Uppsala. Linné was honoured with this stamp in 1939 on the Bicentenary of the Swedish Academy of Sciences which he co-founded and was its first president (Stanley Gibbons 231, 233; Scott 294, 295).

L F HAAS