Am. J. Hum. Genet. 59:1–6, 1996

INVITED EDITORIAL
Huntington Disease—Another Chapter Rewritten

Martha A. Nance

Department of Neurology, Hennepin County Medical Center and Minneapolis Veterans Administration Medical Center, Minneapolis

Summary

To those of us who began life when humans had 48 chromosomes and who began working in genetics when the (by then 46) chromosomes had no bands and chromosome 4 could not reliably be distinguished from chromosome 5, the mere ability to diagnose and correlate the clinical phenotypes of genetic disorders with their molecular genotypes is a source of continuing astonishment and pleasure. Indeed, molecular genetic analysis of neurogenetic disorders such as Huntington disease (HD) has provided a steady stream of challenges and surprises to all who believe the genetic principles that they were taught about these disorders. The paper by Rubinsztein et al. in this issue of the *Journal* highlights yet another surprise, which was adumbrated even in the initial paper announcing the discovery of the HD gene: incomplete penetrance of HD gene mutations.

In 1993, the Huntington's Disease Collaborative Research Group reported the discovery of the IT-15 gene, which encodes a protein named "huntingtin." Near the 5′ end of the gene a polymorphic CAG repeat sequence was identified; in 173 normal alleles, there were 11–34 CAG repeats, whereas 74 HD alleles showed expansions of ⩾42 CAG repeats (Huntington's Disease Collaborative Research Group 1993). After this illuminating discovery, a number of old beliefs about HD were either reaffirmed quickly or redefined dramatically.

Anticipation, the earlier onset of disease symptoms among affected individuals in succeeding generations, is a phenomenon noted often by family members that previously had been regarded as a "statistical artifact" by many geneticists (Vogel and Motulsky 1986, p. 11). Anticipation in HD can now be explained by two findings: (1) a negative correlation between age at symptom onset and repeat number and (2) meiotic instability of abnormally expanded CAG repeats, with a tendency to further expansion. Linear correlation coefficients (r) ranging from −.5 to −.89 have been reported by a number of investigators studying the relationship between age at onset and repeat number (Andrew et al. 1993; Duyao et al. 1993; Kremer et al. 1993; Nørremølle et al. 1993; Rubinsztein et al. 1993; Simpson et al. 1993; Snell et al. 1993; Stine et al. 1993; Iliaroshkin et al. 1994; Kieburtz et al. 1994; Legius et al. 1994; Novelletto et al. 1994; Trottier et al. 1994 Lucotte et al. 1995; Soong and Wang 1995). The correlation is stronger for high repeat numbers (and low ages at onset) and is much weaker for low repeat numbers (and older ages at onset), implying that, although CAG repeat length is a major determinant of onset age in juvenile-onset patients, factors other than CAG repeat length contribute significantly to the onset of HD in the elderly (Duyao et al. 1993; Kremer et al. 1993; Stine et al. 1993; Telenius et al. 1993). The wide confidence intervals for age at onset for allele sizes in the range of 40–50 CAG repeats, which constitute >⅔ of HD alleles (fig. 1), has led to the appropriate recommendation that repeat number should not be used to predict age at onset in patients undergoing predictive testing for HD (Barron et al. 1993; Craufurd and Dodge 1993; Duyao et al. 1993; Nørremølle et al. 1993; Illarioshkin et al. 1994; Legius et al. 1994; Novelletto et al. 1994; Soong and Wang 1995).

Unlike normal alleles, most HD alleles manifest meiotic instability. Of 600 parent-child transmissions reported worldwide, 69% have shown CAG repeat expansion or contraction (de Rooij et al. 1993*b;* Duyao et al. 1993; MacMillan et al. 1993*a;* Simpson et al. 1993; Zühlke et al. 1993*a;* Beilby et al. 1994; Illarioshkin et al. 1994; Legius et al. 1994; Novelletto et al. 1994; Trottier et al. 1994; Kremer et al. 1995; Soong and Wang 1995; author's unpublished data). In contrast, <1% of normal alleles show meiotic instability (Zühlke et al. 1993*a;* Kremer et al. 1995). In most (but not all) studies, a sex-of-parent effect is evident. Overall, 69% of father-child pairs but only 32% of mother-child pairs have shown CAG repeat expansion, and, whereas <2% of maternal transmissions reported worldwide show a change of >5 repeats, up to 21% of all paternal transmissions in one study are expansions of >7 repeats (Kremer et al. 1995). The greatest maternal expansion reported is 16 repeats, whereas the greatest paternal

Received April 29, 1996; accepted for publication May 3, 1996.
Address for correspondence and reprints: Dr. Martha A. Nance, Department of Neurology, Minneapolis VA Medical Center, 1 Veterans Drive, Minneapolis, MN 55417. E-mail: nance@minneapolis.va.gov
This editorial represents the opinions of the author and has not been peer reviewed.
© 1996 by The American Society of Human Genetics. All rights reserved.
0002-9297/96/5901-0001$02.00

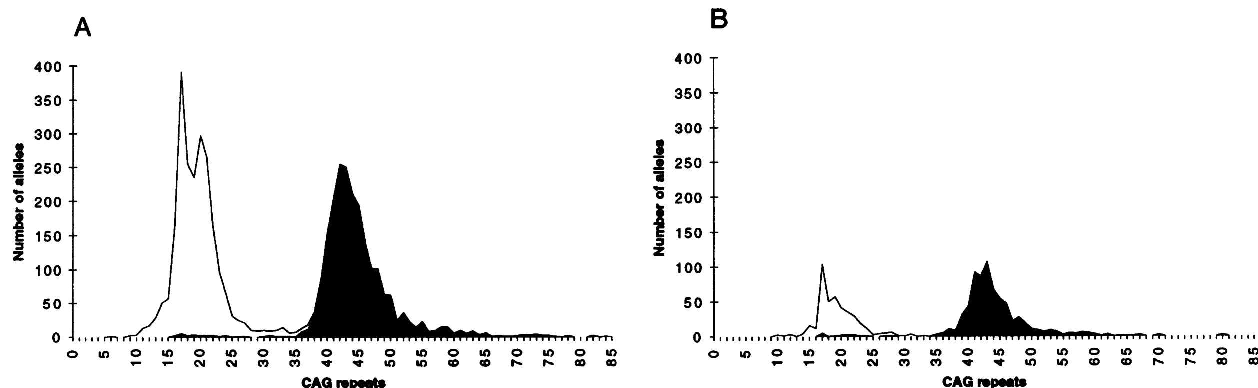

Figure 1 Reported CAG repeat lengths for normal (unshaded) and HD (shaded) alleles, using (*A*) primers amplifying both the CAG and CCG repeats (mean: 19.5 repeats for normal alleles and 44.7 repeats for HD alleles) and (*B*) primers amplifying only the CAG repeat (mean: 19.3 repeats for normal alleles and 43.9 repeats for HD alleles). Six alleles >85 repeats are not included.

expansion is 74 repeats (Kremer et al. 1995). The finding that very large CAG expansions occur almost exclusively among male transmissions provides a molecular corollary to the previously unexplained clinical observation that patients with juvenile-onset HD most often acquire the HD gene from an affected father (Merritt et al. 1969).

These case reports regarding transmission of the HD gene have been biased toward ascertainment of parent-offspring pairs with either late onset in the parent, early onset in the child, or both. In addition, the inclusion in these reports of multiple offspring from the same parent limits a formal comparison of male and female transmissions. Nonetheless, it is evident that CAG instability within the HD gene is greater for male meiosis than for female meiosis. A high degree of mosaicism has been observed among sperm carrying expanded HD alleles, whereas >99% of sperm carrying normal alleles have the same repeat lengths as do somatic cells (MacDonald et al. 1993; Leeflang et al. 1995). Both somatic repeat number and allele history (i.e., whether the allele was derived from an HD patient, an unaffected member of a new mutation family, or the general population) may contribute to interindividual differences in meiotic variability (MacDonald et al. 1993; Goldberg et al. 1995; Leeflang et al. 1995). Family-specific or ethnic factors may modify the degree of intergenerational repeat instability within families or populations, as suggested by several reports of families with relatively invariant repeat numbers (Stine et al. 1993; Rubinsztein et al. 1994; Tzagournissakis et al. 1995) and of certain populations that appear to manifest less instability of expanded alleles than do others (Beilby et al. 1994; Trottier et al. 1994).

Meiotic instability for CAG repeat number provides an explanation for the existence of new mutations for HD. The oft-echoed statement that "the genetic defect that causes HD originated from a common source and . . . new mutations are rare and possibly nonexistent" (Martin and Gusella 1986, p. 1267; similar versions can be found in the works of Hayden [1981], Vogel and Motulsky [1986, p. 419], and Harper [1991, p. 287] and elsewhere) has proved to be wrong. For a small number of sporadic HD cases with available parents, it has been possible to demonstrate a parental (almost always paternal) allele with a CAG repeat number in the low to mid 30s (Goldberg et al. 1993*a*, 1993*b*; Myers et al. 1993; Sánchez et al. 1995). Although in large series ~9%–11% of HD-affected individuals have a missing or negative family history of HD (Goldberg et al. 1993*b*; Nance and Westphal 1996), it has been difficult in the past to demonstrate that such cases meet stringent criteria for the diagnosis of a new mutation (including diagnosis of HD in the proband, exclusion of nonpaternity, parents free of HD at age >60 years, and transmission of HD from the proband to offspring [Stevens and Parsonage 1969]). With the advent of HD gene analysis, reasonable criteria would now include genotyping and negative clinical evaluation of both parents with exclusion of nonpaternity, as well as clinical and molecular confirmation of HD in the proband. Under these criteria, 1.2% (8/650) of unrelated HD cases in a recent large study represent new mutations (Goldberg et al. 1993*b*). Under a less stringent definition, up to 3% of cases may represent new mutations (Goldberg et al. 1993*b*).

The term "intermediate allele" has been used to refer to alleles that do not cause HD in individuals carrying the allele but that are meiotically unstable and able to cause HD in the next generation (Goldberg et al. 1993*a*, 1993*b*; Myers et al. 1993). The lower limit of the intermediate allele range may be as low as 27 CAG repeats (McGlennan et al. 1995). Analysis of markers outside and within the HD gene has shown that two major chromosomal haplotypes are associated with up to 77% of HD cases (MacDonald et al. 1992; Squitieri et al. 1994). Most (but not all) new HD mutations occur on

chromosomes carrying one of the major haplotypes (de Rooij et al. 1993*b;* Myers et al. 1993; Squitieri et al. 1994). The mean CAG length among normal chromosomes carrying one of the two major HD-associated haplotypes is significantly larger than the mean CAG length among all chromosomes, with a distribution that includes relatively more values >30 repeats (Squitieri et al. 1994). Thus, it appears that certain regional chromosomal sequences predispose to increased CAG repeat instability, leading to higher CAG repeat numbers in normal alleles and to eventual expansion into the HD range.

The final issue clarified by mutation analysis of the HD gene has been differential diagnosis. The HD mutation is not present in patients with other psychiatric or neurological disorders (Kremer et al. 1994; Rubinsztein et al. 1994). Some patients with "benign hereditary chorea," however, do have CAG expansions in the HD gene, whereas others do not (MacMillan et al. 1993*a,* 1993*b;* Kremer et al. 1994; Britton et al. 1995). Two reports of schizophrenic patients with 36 CAG repeats are interesting but probably represent a chance coincidence (Rubinsztein et al. 1994; St. Clair 1994).

Incomplete Penetrance of HD Mutations

Anticipating the findings addressed by Rubinsztein et al. in this issue, the Huntington's Disease Collaborative Research Group (1993) reported that an unaffected sibling of a patient with "sporadic" HD had 36 repeats. By the end of 1993, 10 groups from six different countries had reported a normal range of 6–37 repeats, an "intermediate range" of 30–38 repeats, and an HD range of 35–121 repeats (Andrew et al. 1993; Barron et al. 1993; Craufurd and Dodge 1993; de Rooij et al. 1993*a,* 1993*b;* Kremer et al. 1993; MacMillan et al. 1993*b;* Myers et al. 1993; Nørremølle et al. 1993; Rubinsztein et al. 1993; Snell et al. 1993; Stine et al. 1993; Zühlke et al. 1993*b*). Notably, 48 clinically affected patients (~2% of the total), some included and some excluded from the authors' analyses, had two "normal" alleles.

Complicating matters, six trinucleotides downstream from the CAG repeat is a polymorphic CCG repeat sequence, which can itself vary in length, being 7–12 repeats. The first PCR assays for CAG repeat length used primers that amplified both polymorphic sequences. Subsequently, primer sets that exclude the CCG repeat were designed (Warner et al. 1993). In the European population, the CCG repeat is polymorphic primarily when associated with normal CAG repeat lengths, and the CCG-7 allele cosegregates with the expanded CAG repeat in 93%–99% of cases (Andrew et al. 1994*b;* Barron et al. 1994; Squitieri et al. 1994). In populations with a low incidence of HD, such as the Japanese, the Chinese, and the Finns, the overall frequency of the CCG-7 allele is somewhat lower, mean normal CAG repeat lengths are lower, and haplotype distributions for chromosomes carrying expanded HD alleles are different than those in the European Caucasian population (Squitieri et al. 1994). Although the details of the interactions between the CAG repeat, regional haplotypes, and the adjacent CCG repeat remain to be elucidated, from a practical perspective it is clear that estimation of CAG repeat length by an assay that includes a second polymorphic site is potentially inaccurate.

Since 1993, a number of studies from centers around the world have confirmed the presence of expanded CAG repeats in virtually all patients with HD. Figure 1 shows the combined data, representing 2,848 HD alleles and 2,641 normal alleles, of 24 series (Andrew et al. 1993; Craufurd and Dodge 1993; de Rooij et al. 1993*b;* MacMillan et al. 1993*b;* Myers et al. 1993; Nørremølle et al. 1993; Rubinsztein et al. 1993; Simpson et al. 1993; Snell et al. 1993; Stine et al. 1993; Zühlke et al. 1993*b;* Ashizawa et al. 1994; Barron et al. 1994; Beilby et al. 1994; Davis et al. 1994; Kieburtz et al. 1994; Legius et al. 1994; Novelletto et al. 1994; Trottier et al. 1994; Dürr et al. 1995; Illarioshkin et al. 1995; Lucotte et al. 1995; Soong and Wang 1995; author's unpublished data) This figure excludes the large series reported by Duyao et al. (1993) and Kremer et al. (1994), as well as several studies that appeared to include previously reported patients. Repeat sizes in the range of 30–39 constitute <1% of normal alleles but almost 3% of HD alleles. A region of overlap extending over ~35–38 repeats is apparent.

Worldwide, ⩾82 normal alleles in the 31–39-repeat range (de Rooij et al. 1993*a,* 1993*b;* Goldberg et al. 1993*a;* MacMillan et al. 1993*b;* Myers et al. 1993; Rubinsztein et al. 1993, 1994; St. Clair 1994; Simpson et al. 1993; Snell et al. 1993; Stine et al. 1993; Zühlke et al. 1993*b;* Beilby et al. 1994; Davis et al. 1994; Legius et al. 1994; Novelletto et al. 1994; Trottier et al. 1994; Kremer et al. 1995; Tzagournissakis et al. 1995; author's unpublished data), as well as 137 upper alleles of ⩽38 repeats from HD-affected individuals, have been reported (Andrew et al. 1994*a;* Barron et al. 1993; Craufurd and Dodge 1993; de Rooij et al. 1993*b;* MacMillan et al. 1993*b;* Myers et al. 1993; Novelletto et al. 1993; Rubinsztein et al. 1993; Simpson et al. 1993; Snell et al. 1993; Stine et al. 1993; St. Clair 1994; Zühlke et al. 1993*a;* Ashizawa et al. 1994; Davis et al. 1994; Kieburtz et al. 1994; Legius et al. 1994; Dürr et al. 1995; Lucotte et al. 1995; Tzgournissakis et al. 1995). However, although the suggestions that some individuals with repeat lengths in the 30s live long lives without an illness recognizable as HD and that occasional patients with <37 repeats develop HD are not new, variations in laboratory methods and the sparse clinical information provided have prevented any firm conclusions from being drawn from these case reports.

In an attempt to burn through the fog surrounding this issue, Rubinsztein et al. studied, using uniform methods, 178 individuals with CAG repeat lengths in the 30–40 range. Of these, only 61 patients are discussed in their paper, and some of these appear to have been described elsewhere. Thus, it is not possible to adjust, compare, or combine the data depicted in figure 1 with the Rubinsztein data. In addition, one can only assume that the affected individuals with 36–38 repeats are truly affected with HD, since clinical aspects of their illness are not mentioned.

Apart from these reservations, the major findings are not disputable: (1) some patients with 36 repeats have HD; (2) some very old people with 36–39 repeats do not have recognizable HD; and (3) a small (but nonzero) number of individuals would be miscategorized if primers that fail to exclude the CCG repeat are used. These findings have several implications for clinical laboratories and clinicians who test patients for HD.

It was thought previously that the HD gene mutation is fully penetrant—that is, that all carriers of the gene mutation would develop the disease if they lived long enough. This study demonstrates that nonpenetrance for CAG repeat expansions exists, at least as defined by current clinical and pathological means. A new designation, the "range of incomplete penetrance", can be defined, referring to allele sizes that have been associated with HD in some but not all individuals. Although strict demonstration of nonpenetrance should include (1) death without symptoms after age 85 years and (2) absence of HD pathology on autopsy (criteria fulfilled by one patient in the Rubinsztein study), a less strict definition might be appropriate for the clinical setting. It should be emphasized that the intermediate allele range and the range of incomplete penetrance are not synonymous; although the ranges may overlap, they have different clinical definitions and implications. As defined by this study, the range of incomplete penetrance is 36–39 repeats. As noted above, previous studies suggesting that occasional patients with ≤35 repeats may manifest HD are difficult to interpret. It remains possible, however, that the HD phenotype may occasionally be penetrant in individuals carrying alleles with <36 repeats. Further studies are required to determine empirically the frequency of nonpenetrance for different allele sizes in the range of incomplete penetrance. Clinicians and laboratories should counsel new patients appropriately about the possibility of nonpenetrance of CAG expansions in the 36–39 repeat range, and they should consider recounseling or retesting, using CCG-excluding primers, any patients whose results were in or close to the 36–39 repeat range but who were studied with a CCG-inclusive primer set.

Anticipation, new mutations, intermediate alleles, and nonpenetrance having all now been demonstrated, a number of molecular and clinical details remain to be elucidated. Adjacent intragenic sequences, regional chromosomal haplotypes, the normal allele, interactions of the huntingtin protein with other proteins, or other environmental factors may be important modifiers of these aspects of disease expression. It is not yet clear whether an expanding CAG repeat sequence acquires its two abnormal properties (meiotic instability and the ability to cause disease) simultaneously, in a related or coordinated fashion, or independently. Identification of family-specific or ethnic factors that modify the dynamic and disease-related aspects of CAG expansions would be particularly important to clinicians, as would population-based studies from which empiric risk figures for penetrance and allele expansion could be derived.

From a clinician's perspective, the dearth of clinical detail about normal and affected individuals with alleles in the range of incomplete penetrance and about other "HD" patients with two unexpanded alleles is unsettling, particularly since these may represent up to 5% of HD patients. Although clinical oversight, misdiagnosis, or laboratory error may account for many such cases, it remains possible that there is a clinical phenotype of HD (e.g., among the very elderly) that is not yet recognized, that a low-frequency mutational mechanism other than CAG repeat expansion is responsible for HD in a small percentage of cases (as recently shown for another trinucleotide repeat disease, Friedreich ataxia [Campuzano et al. 1996]), or that other, as yet unnamed, phenocopies of HD exist. Only by careful clinical, pathological, and genetic study of these unusual patients can these clinical issues be resolved.

Finally, the lessons learned from HD may also be applicable to other trinucleotide repeat diseases. HD is one of a growing group of "CAG repeat" disorders that also include the less common diseases spinocerebellar ataxia-1 (SCA 1), Machado-Joseph disease (MJD, or spinocerebellar ataxia-3 [SCA 3]), Kennedy disease (spinobulbar muscular atrophy [SBMA]), and dentatorubro-pallidoluysian atrophy (DRPLA). Although several genetic revelations about HD were evident after the molecular genetic analysis of the first few hundred HD alleles, other issues and their resolution have become apparent only as several thousand normal and expanded HD alleles have been studied. Although it may be possible to obtain sufficient empiric data to guide clinicians in the use of the HD gene test, it may not be possible to analyze an equivalent number of patients with less common CAG repeat diseases. Knowing that the normal and abnormal ranges, sensitivities, specificities, and other disease-specific details for such "orphan tests" are based on a small sample of patients, clinicians should utilize caution and an open mind when they apply these tests to clinical use.

References

Andrew SE, Goldberg YP, Kremer B, Squitieri F, Theilmann J, Zeisler J, Telenius H, et al (1994*a*) Huntington disease

without CAG expansion: phenocopies or errors in assignment? Am J Hum Genet 54:852–863

Andrew SE, Goldberg YP, Kremer B, Telenius H, Theilmann J, Adam S, Starr E, et al (1993) The relationship between trinucleotide (CAG) repeat length and clinical features of Huntington's disease. Nat Genet 4:398–403

Andrew SE, Goldberg YP, Theilmann J, Zeisler J, Hayden MR (1994*b*) A CCG repeat polymorphism adjacent to the CAG repeat in the Huntington disease gene: implications for diagnostic accuracy and predictive testing. Hum Mol Genet 3: 65–68

Ashizawa T, Wong L-JC, Richards CS, Caskey CT, Jankovic J (1994) CAG repeat size and clinical presentation in Huntington's disease. Neurology 44:1137–1143

Barron LH, Rae A, Holloway S, Brock DJH, Warner JP (1994) A single allele from the polymorphic CCG rich sequence immediately 3′ to the unstable CAG trinucleotide in the IT15 cDNA shows almost complete disequilibrium with Huntington's disease chromosomes in the Scottish population. Hum Mol Genet 3:173–175

Barron LH, Warner JP, Porteous M, Holloway S, Simpson S, Davidson R, Brock DJH (1993) A study of the Huntington's disease associated trinucleotide repeat in the Scottish population. J Med Genet 30:1003–1007

Beilby J, Chin CY, Porter I, Walpole IR, Goldblatt J (1994) Improving diagnosis of Huntington's disease by analysis of an intragenic trinucleotide repeat expansion. Med J Aust 161:356–360

Britton JW, Uitti RJ, Ahlskog JE, Robinson RG, Kremer B, Hayden MR (1995) Hereditary late-onset chorea without significant dementia: genetic evidence for substantial phenotypic variation in Huntington's disease. Neurology 45:443–447

Campuzano V, Montermini L, Moltò MD, Pianese L, Cossée M, Cavalcanti F, Monros E, et al (1996) Friedreich's ataxia: autosomal recessive disease caused by an intronic GAA triplet repeat expansion. Science 271:1423–1427

Craufurd D, Dodge A (1993) Mutation size and age at onset in Huntington's disease. J Med Genet 30:1008–1011

Davis MB, Bateman D, Quinn NP, Marsden CD, Harding AE (1994) Mutation analysis in patients with possible but apparently sporadic Huntington's disease. Lancet 344:714–717

de Rooij KE, de Koning Gans PAM, Losekoot M, Bakker E, den Dunnen JT, Vegter-van der Vlis M, Roos RAC, et al (1993*a*) Borderline repeat expansion in Huntington's disease. Lancet 342:1491–1492

de Rooij KE, de Koning Gans PAM, Skraastad MI, Belfroid RDM, Vegter-van der Vlis M, Roos RAC, Bakker E, et al (1993*b*) Dynamic mutation in Dutch Huntington's disease patients: increased paternal repeat instability extending to within the normal size range. J Med Genet 30:996–1002

Dürr A, Dodé C, Hahn V, Pêcheux C, Pillon B, Feingold J, Kaplan J-C, et al (1995) Diagnosis of "sporadic" Huntington's disease. J Neurol Sci 129:51–55

Duyao M, Ambrose C, Myers RH, Novelletto A, Persichetti F, Frontali M, Folstein S, et al (1993) Trinucleotide repeat length instability and age of onset in Huntington's disease. Nat Genet 4:387–392

Goldberg YP, Andrew SE, Theilmann J, Kremer B, Squitieri F, Telenius H, Brown JD, et al (1993*a*) Familial predisposition to recurrent mutations causing Huntington's disease: genetic risk to sibs of sporadic cases. J Med Genet 30:987–990

Goldberg YP, Kremer B, Andrew SE, Theilmann J, Graham RK, Squitieri F, Telenius H, et al (1993*b*) Molecular analysis of new mutations for Huntington's disease: intermediate alleles and sex of origin effects. Nat Genet 5:174–179

Goldberg YP, Zeisler J, Almqvist E, Sillence D, Richards F, Buchanan J, Telenius H, et al (1995) Increased instability of intermediate alleles in families with sporadic Huntington disease compared to intermediate alleles in the general population: implications for genetic counseling. Am J Hum Genet Suppl 57:A28

Harper PS (1991) Huntington's disease. WB Saunders, London

Hayden MR (1981) Huntington's chorea. Springer, New York

Huntington's Disease Collaborative Research Group (1993) A novel gene containing a trinucleotide repeat that is expanded and unstable on Huntington's disease chromosomes. Cell 72:971–983

Illarioshkin SN, Igarashi S, Onodera O, Markova ED, Nikolskaya NN, Tanaka H, Chabrashwili TZ, et al (1994) Trinucleotide repeat length and rate of progression of Huntington's disease. Ann Neurol 36:630–635

Kieburtz K, MacDonald M, Shih C, Feigin A, Steinberg K, Bordwell K, Zimmerman C, et al (1994) Trinucleotide repeat length and progression of illness in Huntington's disease. J Med Genet 31:872–874

Kremer B, Almqvist E, Theilmann J, Spence N, Telenius H, Goldberg YP, Hayden MR (1995) Sex-dependent mechanisms for expansions and contractions of the CAG repeat on affected Huntington disease chromosomes. Am J Hum Genet 57:343–350

Kremer B, Goldberg YP, Andrew SE, Theilmann J, Telenius H, Zeisler J, Squitieri F, et al (1994) A worldwide study of the Huntington's disease mutation: the sensitivity and specificity of measuring CAG repeats. N Engl J Med 330: 1401–1406

Kremer B, Squitieri F, Andrew SE, Theilmann J, Spence N, Goldberg YP, Hayden MR (1993) Molecular analysis of late onset Huntington's disease. J Med Genet 30:991–995

Leeflang EP, Fan F, Losekoot M, van Ommen GJB, Arnheim N (1995) Single sperm analysis of the transition from high normal to diseased triplet repeat length at the Huntington's disease locus. Am J Hum Genet Suppl 57:A29

Legius E, Cuppens H, Dierick, Van Zandt K, Dom R, Fryns J-P, Evers-Kiebooms G, et al (1994) Limited expansion of the $(CAG)_n$ repeat of the huntingtin gene: a premutation? Eur J Hum Genet 2:44–50

Lucotte G, Turpin JC, Riess O, Epplen JT, Siedlaczk I, Loirat F, Hazout S (1995) Confidence intervals for predicted age of onset, given the size of $(CAG)_n$ repeat in Huntington's disease. Hum Genet 95:231–232

MacDonald ME, Barnes G, Srinidhi J, Duyao M, Ambrose CM, Myers RH, Gray J, et al (1993) Gametic but not somatic instability of CAG repeat length in Huntington's disease. J Med Genet 30:982–986

MacDonald ME, Novelletto A, Lin C, Tagle D, Barnes G, Bates G, Taylor S, et al (1992) The huntington's disease candidate gene exhibits many different haplotypes. Nat Genet 1:99–103

MacMillan JC, Morrison PJ, Nevin NC, Shaw DJ, Harper

PS, Quarrell OWJ, Snell RG (1993*a*) Identification of an expanded CAG repeat in the Huntington's disease gene (IT15) in a family reported to have benign chorea. J Med Genet 30:1012–1013

MacMillan JC, Snell RG, Tyler A, Houlihan GD, Fenton I, Cheadle JP, Lazarou LP, et al (1993*b*) Molecular analysis and clinical correlations of the Huntington's disease mutation. Lancet 342:954–958

Martin JB, Gusella JF (1986) Huntington's disease: pathogenesis and management. N Engl J Med 315:1267–1276

McGlennan RC, Allinson PS, Matthias Hagen VL, Parker TL, Lovell MA, Kelly TE (1995) Evidence of an unstable paternal 27 CAG repeat allele in the huntingtin gene giving rise to clinically overt Huntington disease in a patient with the genotype (17/38). Am J Hum Genet Suppl 57:A246

Merritt AD, Conneally PM, Rahman NF, Drew AL (1969) Juvenile Huntington's chorea. In: Barbeau A, Brunette JR (eds) Progress in neurogenetics. Excerpta Medica, Amsterdam, pp 645–650

Myers RH, MacDonald ME, Koroshetz WJ, Duyao MP, Ambrose CM, Taylor SAM, G Barnes G, et al (1993) De novo expansion of a $(CAG)_n$ repeat in sporadic Huntington's disease. Nat Genet 5:168–173

Nance MA, Westphal B (1996) Diagnosis of patients presenting to a Huntington's disease clinic without a family history of Huntington's disease. Neurology Suppl 46:A251

Nørremølle A, Riess O, Epplen JT, Fenger K, Hasholt L, Sørenson SA (1993) Trinucleotide repeat elongation in the Huntingtin gene in Huntington disease patients from 71 Danish families. Hum Mol Genet 2:1475–1476

Novelletto A, Persichetti F, Sabbadini G, Mandich P, Bellone E, Ajmar F, Pergola M, et al (1994) Analysis of the trinucleotide repeat expansion in Italian families affected with Huntington disease. Hum Mol Genet 3:93–98

Rubinsztein DC, Barton DE, Davidson BCC, Ferguson-Smith MA (1993) Analysis of the huntingtin gene reveals a trinucleotide-length polymorphism in the region of the gene that contains two CCG-rich stretches and a correlation between decreased age of onset of Huntington's disease and CAG repeat number. Hum Mol Genet 2:1713–1715

Rubinsztein DC, Leggo J, Coles R, Almqvist E, Biancalana V, Cassiman J-J, Chotai K, et al (1996) Phenotypic characterization of individuals with 30–40 CAG repeats in the Huntington disease (HD) gene reveals HD cases with 36 repeats and apparently normal elderly individuals with 36–39 repeats. Am J Hum Genet 59:16–22 (in this issue)

Rubinsztein DC, Leggo J, Goodburn S, Crow TJ, Lofthouse R, DeLisi LE, Barton DE, et al (1994) Study of the Huntington's disease (HD) gene CAG repeats in schizophrenic patients shows overlap of the normal and HD affected ranges but absence of correlation with schizophrenia. J Med Genet 31:690–693

St Clair D (1994) Expanded CAG trinucleotide repeat of Huntington's disease gene in a patient with schizophrenia and normal striatal histology. J Med Genet 31:658–659

Sánchez A, Rosich M, Castellví-Bel S, Milà M, Badenas C, Jiménez D, Estivill X (1995) Sporadic Huntington's disease due to maternal transmission. Am J Hum Genet Suppl 57: A227

Simpson SA, Davidson MJ, Barron LH (1993) Huntington's disease in Grampian region: correlation of the CAG repeat number and the age of onset of the disease. J Med Genet 30:1014–1017

Snell RG, MacMillan JC, Cheadle JP, Fenton I, Lazarou LP, Davies P, MacDonald ME, et al (1993) Relationship between trinucleotide repeat expansion and phenotypic variation in Huntington's disease. Nat Genet 4:393–397

Soong B-W, Wang J-T (1995) A study on Huntington's disease associated trinucleotide repeat within the Chinese population. Proc Natl Sci Counc Repub China B 19:137–142

Squitieri F, Andrew SE, Goldberg YP, Kremer B, Spence N, Zeisler J, Nichol K, et al (1994) DNA haplotype analysis of Huntington disease reveals clues to the origins and mechanisms of CAG expansion and reasons for geographic variations of prevalence. Hum Mol Genet 3:2103–2114

Stevens DL, Parsonage M (1969) Mutation in Huntington's chorea. J Neurol Neurosurg Psychiatry 32:140–143

Stine OC, Pleasant N, Franz ML, Abbott MH, Folstein SE, Ross CA (1993) Correlation between the onset age of Huntington's disease and length of the trinucleotide repeat in IT-15. Hum Mol Genet 2:1547–1549

Telenius H, Kremer B, Goldberg YP, Theilmann J, Andrew SE, Zeisler J, Adam S, et al (1994) Somatic and gonadal mosaicism of the Huntington disease gene CAG repeat in brain and sperm. Nat Genet 6:409–414

Telenius H, Kremer HPH, Theilmann J, Andres SE, Almqvist E, Anvret M, Greenberg C, et al (1993) Molecular analysis of juvenile Huntington disease: the major influence on $(CAG)_n$ repeat length is the sex of the affected parent. Hum Mol Genet 2:1535–1540

Trottier Y, Biancalana V, Mandel J-L (1994) Instability of CAG repeats in Huntington's disease: relation to parental transmission and age of onset. J Med Genet 31:377–382

Tzagournissakis M, Fesdjian CO, Shashidharan P, Plaitakis A (1995) Stability of the Huntington disease $(CAG)_n$ repeat in a late-onset form occurring on the island of Crete. Hum Mol Genet 4:2239–2243

Vogel F, Motulsky A (1986) Human genetics, 2d ed. Springer, New York

Warner JP, Barron L, Brock DJP (1993) A new polymerase chain reaction (PCR) assay for the trinucleotide repeat that is unstable and expanded in Huntington's disease. Mol Cell Probes 7:235–239

Zühlke C, Riess O, Bockel B, Lange H, Thies U (1993*a*) Mitotic stability and meiotic variability of the $(CAG)_n$ repeat in the Huntington disease gene. Hum Mol Genet 2:2063–2067

Zühlke C, Riess O, Schroeder K, Siedlaczck I, Epplen JT, Engel W, Thies U (1993*b*) Expansion of the $(CAG)_n$ repeat causing Huntington's disease in 352 patients of German origin. Hum Mol Genet 2:1467–1469