Neurol Sci (2002) 23:S107–S108

CAG mutation effect on rate of progression in Huntington's disease

F. Squitieri (✉) · M. Cannella · M. Simonelli
Neurogenetics Unit, IRCCS Neuromed, Pozzilli (IS), Italy

Abstract Huntington's disease (HD) is progressively invalidating and caused by a CAG expanded mutation. We tested the effect of the mutation length on the rate of progression in a cohort of 80 patients clinically followed-up and genetically characterized. Two patients presenting an infantile and aggressive HD form starting under 10 years had over 90 repeats; the other patients did not show any influence of the CAG expanded number on the rate of progression. In conclusion, the CAG expanded repeat affects the disease progression only at a very upper pathological range and in rare cases initiating very early in the life, while it does not seem to affect in any way the severity of the phenotype in most HD patients. Other factors affecting the motor symptom progression, other than the expanded repeats, therefore have to be investigated.

Huntington disease's (HD), dominantly transmitted and caused by a CAG expanded mutation beyond 36 repeats [1], is highly and progressively invalidating. The severity of the symptom progression is variable among patients, with juvenile (onset <20 years) [2] and homozygous subjects (F. Squitieri, unpublished results) showing the most accelerated course towards high disability. However, although the age at onset is affected by the CAG mutation length [3] (the low expanded number is associated with a variable penetrance of the mutation [4]), no genetic factors have so far been described as possible modifiers of the disease severity [5]. In this study we analyzed the possible contribution of the expanded CAG repeat length on symptom progression in a cohort of 80 patients of identical ethnic origin, whose genetic and clinical data of age at onset and follow-up were available from the Italian HD Databank [5].

All 80 patients (37 males and 43 females) were seen at the Neurogenetics Unit of the IRCCS Neuromed of Pozzilli (IS), Italy and clinically analyzed on the Unified Huntington Disease Rating Scale (UHDRS), which includes motor, behavioral, cognitive and independence assessments. The rate of disability was assessed with the Total Functional Capacity (TFC) [6] and by the disability scale (DS) [7] and calculated as loss of units per year (TFC, mean 0.6±0.5, range 0.2–3.2; DS, mean 3.4±2.8, range 0.8–13.7). The onset of disease was defined as the time when clinical motor manifestations (i.e. choreic movements) first became noticeable (mean 41.3±14.0 years, range 3–73). To investigate the size of the CAG repeat expanded mutation, a molecular genetic test on patients' DNA was performed, after informed consent, at Neuromed by published techniques [8] (mean 46.3±10.5 CAG repeats, range 39-100). For statistical analysis, a nonparametric test (Mann-Whitney U) and a linear regression approach were used (significance at $p<0.05$). Among these 80 patients, two had an infantile form (starting at an age less than 10 years) with age at onset at three and seven years, 100 and 95 repeats, respectively, and a particularly devastating progression towards a high loss of independence (mean loss of units per year was 2.7±0.7 (range 2.2-3.2) at TFC and 12.7±1.4 (range 11.7–13.7) at DS). The mean loss-of-unit values per year obtained from these two little patients was significantly different from the cohort of juvenile (onset under 20 years, n=4, mean 0.7±0.7 units per year at TFC, $p=0.016$ and 4.7±3.9 at the DS; $p=0.017$) and adult subjects (0.6±0.3 units per year at TFC, $p=0.008$ and 3.0±2.3 at DS, $p=0.009$). In order to examine a possible influence of the mutation length on the HD symptom progression, we plotted the expanded CAG repeat number with the mean loss of units per year at TFC and DS and found a significant correlation (CAG/TFC, $p=0.02$; CAG/DS, $p=0.01$) if the two patients were included in the cohort of all 80 subjects (Fig. 1, panels A and B). Conversely, there was no significant correlation after excluding the two patients with the infantile form (Fig. 1, $p>0.05$, panels C and D).

Discussion

We found an influence of the mutation length on HD severity only when the mutation was much expanded, the CAG repeat number at the upper edge of the pathological range so far described [1]. In these rare cases, the increased toxic effect of the mutation likely contributes to the devastating severity of the phenotype. This was not the case of the adult affected population whose accurate clinical follow-up consented to obtain data of symptom progression from our databank. The juvenile patients (onset between 11 and 20 years), whose assessment of HD progression was available, were only four and did not differ substantially from the adults. After the discovery of the gene mutation, only few reports studied the influence of the very expanded CAG repeat number on HD severity [10]. This was likely due to the difficulty of studying large affected HD cohorts of patients both accurately followed-up and genetically characterized. Our study confirms the effect of large CAG expansions on the rate of HD progression [10]. Differently from Illarioshkin *et al*. [10], we did not find any significant correlation after excluding from statistical analysis patients carrying the most expanded mutations (Fig. 1). It likely depended on either the different genetic background of the patient cohorts analyzed and on the diverse clinical methodology used to calculate the rate of progression. We have recently reported the occurrence of a worse HD prognosis in case of at onset atypical

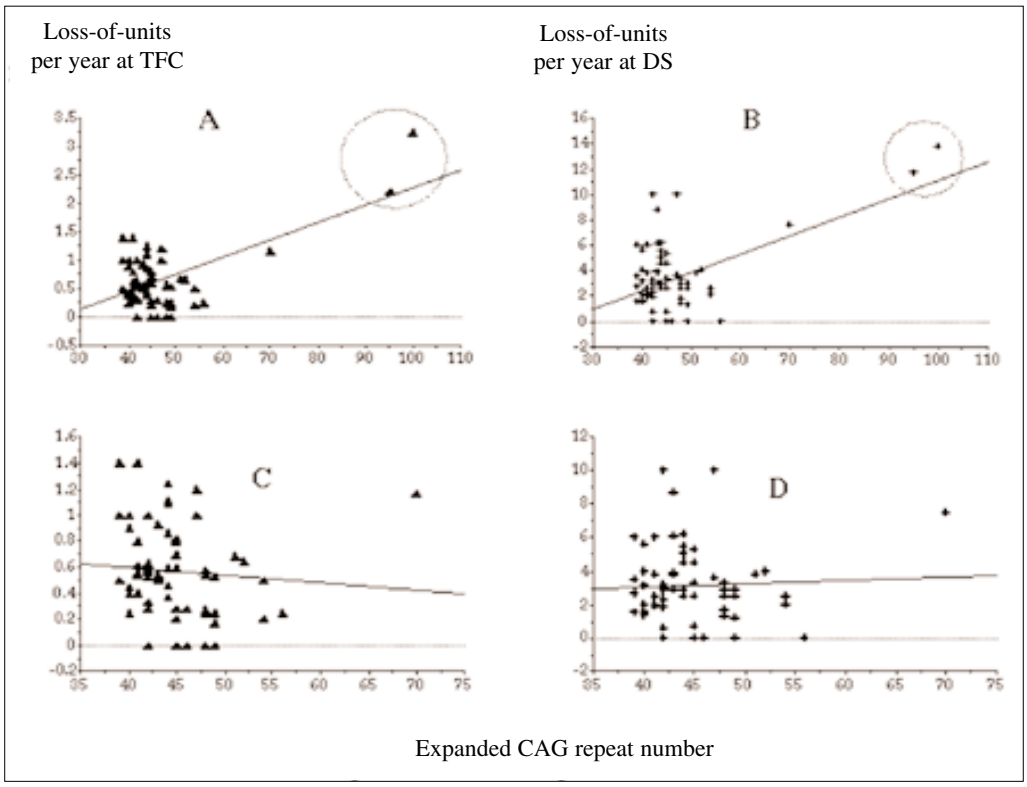

Fig. 1a-d. Linear regression analysis between expanded CAG repeat number (x-axis) and loss of units per year (y-axis) at the TFC and DS. **a,b** Analysis including the two patients with infantile form. **c,d** Excluding the 2 patients with infantile Huntington's disease. The infantile patients are highlighted by a circle in **a** and **b**

movement disorder [9]. In that study, we found mean expanded repeats longer in the juvenile rigid patients than in the juveniles initiating with chorea, predicting therefore the occurrence of an influence of large expansions on HD phenotype. We confirm such hypothesis as both patients affected with infantile form here described presented at onset movement disorder other than chorea, characterized by rigidity in one and dystonia and limb ataxia in the other. In these cases, as in the rigid juvenile patients, the particularly expanded and toxic mutation leads to a highly invalidating phenotype. Conversely, in most patients the CAG repeat contributes to the age at onset for about 50%–60%, the remaining percentage is influenced by other genetic factors probably of familial origin [3], but not to the rate of disease progression. Other factors affecting the severity of the phenotype have to be studied on cohorts of patients well characterized clinically and genetically. The discovery of factors influencing the symptom progression and, possibly predicting the HD prognosis, will offer new opportunities in the therapeutic strategies for this devastating disease.

References

1. Kremer B, Goldgerg P, Andrew SE et al (1994) A worldwide study of the Huntington's disease mutation. N Engl J Med 330:1401–1406
2. Nance MA (1997) The US HD Genetic Testing Group: genetic testing of children at risk for Huntington's disease. Neurology 49:1050–1053
3. Squitieri F, Sabbadini G, Mandich P et al (2000) Family and molecular data for a fine analysis of age at onset in Huntington disease. Am J Med Genet 95:366–373
4. Rubinsztein DC, Leggo J, Coles R 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
5. Squitieri F, Cannella M, Giallonardo P et al (2001) Onset and pre-onset studies to define the Huntington's disease natural history. Brain Res Bull 56:233–238
6. Shoulson I, Kurlan R, Rubin AJ et al (1989) Assessment of functional capacity in neurodegenerative movement disorders: Huntington's disease as a prototype. In: Munsat TL (ed) Quantification of neurological deficit. Butterworths, Boston, pp 271–283
7. Myers RH, Sax DS, Koroshetz WJ et al (1991) Factors associated with slow progression in Huntington's disease. Arch Neurol 48:800–804
8. Warner JP, Barron LH, Brock DJH (1993) A new polymerase chain reaction (PCR) assay for the trinucleotide repeat that is unstable and expanded on Huntington's disease chromosome. Mol Cell Probes 7:235–239
9. Squitieri F, Berardelli A, Nargi E et al (2000) Atypical movement disorders in the early stages of Huntington's disease: clinical and genetic analysis. Clin Genet 58:50–56
10. Illarioshkin NS, Igarashi S, Onodera O et al (1994) Trinucleotide repeat length and rate of progression of Huntington's disease. Ann Neurol 36:630–635