Huntington's disease: report on four Saudi families

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Abstract Objectives: To report on Huntington's disease (HD), rarely reported in the Arabian Peninsula.

Methods: Over a period of eight years, four families with typical features of HD were diagnosed. All families were of native origin and two were of "Bedouin" (nomadic) origin. The inheritance was autosomal-dominant in all. The disease onset occurred between age 25-35 years and was characterized by mood and personality changes followed by clumsiness and chorea. Apart from a history of one maternal grandmother who married into a Moroccan family, there were no known foreign antecedents.

Results: In all affected individuals, analysis of the trinucleotide repeat (CAG) in the Huntington gene IT15 confirmed the presence of an expanded repeat. The range was 41-55 (control range 12-28, median 17).

Conclusions: These findings support the hypothesis that a new mutation was responsible for the HD in Saudi Arabia. There may be more patients with HD in Saudi Arabia than previously thought.

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Huntington's disease is a fully penetrant genetic disease inherited in an autosomal dominant pattern. Most patients present in their early thirties with clumsiness, chorea and dementia. Death usually ensues some 15 to 20 years later.1-3 A subset of patients (10%) present before the age of 20. The juvenile onset cases are usually associated with paternal transmission1 and are characterized by spasticity, rigidity, intellectual decline and a rapidly progressive course.

Pathologically, there is loss of neurons and gliosis in the caudate and putamen (the striatum) resulting in atrophy. Medium-size neurons containing \( \gamma \)-aminobutyric acid (GABA) and enkephalin or GABA and substance P are predominantly affected.1-2 The biochemical basis for neuronal death in HD is unknown and there is no effective treatment.

In 1983, the gene responsible for HD was mapped to the short arm of chromosome 4 using restriction fragment length polymorphism technique.4 Recently the molecular defect was identified as an expansion of a polymorphic (CAG) repeat in the 5' part of IT15 gene.5 The gene spans about 210 kb of genomic deoxyribonucleic acid (DNA) and encodes a protein with a predicted size of 348 kDa, designated "Huntington". The repeat stretch is inversely correlated with age of onset.5,6 Huntington's disease occurs worldwide with an overall prevalence of 5-10 per 100,000 population.5,7 Recently two Saudi families with HD were reported,8 but no DNA linkage studies were carried out.

In this communication, we report four native Saudi families who presented to our tertiary care hospital with a clinical picture suggesting HD. In three families, the expanded trinucleotide repeat was demonstrated in all affected members and confirmed the diagnosis.

Family 1 The proband was a 40 year old male Saudi living in Qatif (an old city in the Eastern part of Saudi Arabia). He presented with a 10 year history of progressive intellectual deterioration

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and worsening choreiform movements affecting the face, tongue and all limbs. Four years into his illness he had to retire as a carpenter. The pedigree (Fig. 1) indicates an autosomal dominant inheritance; there was no known foreign antecedent. One sister and two paternal cousins were similarly affected. Their age of onset was in the late twenties.

The proband was underweight, demented, with deficient abstract thinking, impaired memory and an apathetic attitude. He was dysarthric and ataxic with choreic movements and hyper-reflexia. Routine laboratory investigations were normal. An electroencephalogram showed low amplitude activity (less than 5µV), with no focal or paroxysmal features. Magnetic resonant imaging (MRI) of the brain showed moderate atrophy. The heads of caudate nuclei were selectively affected with an increased bicaudate diameter of 29 mm (normal range 12.5-15 mm) indicating severe atrophy of the nuclei.9

Family II All members of this family were Saudis living in Makkah. The pedigree indicates autosomal dominant inheritance (Fig.1). An elderly sister (III.60) is married to a Moroccan, otherwise there was no foreign antecedent.

The proband (III.43) is a 43 year old Saudi female. She had a seven year history of forgetfulness, irritability, pathological jealousy, loss of interest in her surroundings, social isolation, with abnormal shaky movements, unsteadiness of gait, occasional incontinence of urine and stool, and loss of libido.

She was emaciated and demented but was able to follow simple commands. She had dysarthria, involuntary grinning, ataxia with hypoactive tendon reflexes but no weakness. Computed tomography (CT) and magnetic resonance imaging (MRI) of the brain indicated advanced atrophy of the striatum and generalized cortical atrophy with dilatation of the ventricles. The bicaudate diameter was 27 mm (Fig. 2).

The patient’s brothers, sister and nephew were similarly demented and ataxic, with personality changes and generalized chorea.

Family III The members of the third kindred were Bedouin (nomads), living in the Northern part of the Arabian peninsula. There were no known foreign ancestors. The family pedigree supported autosomal dominant inheritance in three successive affected generations.

The proband, a 52 year old female, presented with psychiatric problems during her last pregnancy when she was 37 years old. She was
paranoid, had a history of suicide attempts, delusions and morbid jealousy. She developed choreoathetoid movements in her lower limbs and grimacing later. She was ataxic with hyperactive tendon reflexes. MRI of the brain showed moderate cortical atrophy.

The patient’s mother was clumsy and demented. She died at the age of 45. The patient’s maternal aunt was diagnosed to have HD at the age of 48.

**Family IV** The family members lived in Riyadh but originated from Qassim (approximately 330 km north of Riyadh). The proband, a 35 year old man, presented with anxiety. His psychiatric problems started in his mid-twenties and necessitated admission to a psychiatric hospital. Later he became fidgety, dysarthric and socially withdrawn. Subsequently, seizures occurred. An older brother became bedridden at the age of 45 with similar problems. The brother was severely demented, mute and emaciated with weak, flexed limbs. The mother died at the age of 42 and was reported to have chorea. Further details of the family history were not available.

A CT brain of the proband showed moderate frontal atrophy with an increased bicaudate diameter of 24 mm. All routine hematological and biochemical studies were normal.

**PCR assay and assessment of (CAG)\(_n\) repeat** Genomic DNA was extracted from venous blood sample or cultured EBV transformed cell lines as previously described.\(^{10-11}\)

For PCR analysis, the published primers of the (CAG)\(_n\) repeat were used. The PCR assay was modified as follows. PCR was performed in a reaction volume of 25 μl using 100 ng of genomic DNA, 100 pmol of each primer, 10 mmol/l Tris-HCl (pH 8.3), 5 mmol/l KCl, 2 mmol/l MgCl\(_2\), 100 μmol/l dNTP (with a 1:1 ratio of dGTP and 7-deaza-dGTP), 10% DMSO, 2.0 μCi of α[\(^{32}\)P]-dCTP (Amersham), and 1.25 U of AmpliTaq (Cetus). After heating for 95°C for 10 minutes, 40 cycles of one minute at 95°C, one minute at 65°C, two minutes at 72°C were performed in a DNA Thermal Cycler (Perkin Elmer Cetus). The addition of 7-deaza-dGTP proved essential to generate an interpretable signal and low background.

PCR products were mixed with an equal volume of 95% formamide loading buffer, denatured for five minutes at 95°C, and separated on 6% denaturing polyacrylamide gels. Autoradiography of fixed and dried gels was one to four days at room temperature. Since the band below the upper band of the PCR product was in most cases the major band using these primers and PCR conditions, this band was taken to estimate allele sizes relative to M13 sequencing ladders, not incubated with 7-deaza-dGTP. Incubation of the marker with 7-deaza-dGTP did not alter its electrophoretic mobility. The proximity of markers proved essential for accurate size determination. DNA samples from 38 members of families I, II and III were assessed for CAG repeat. An expanded number of CAG repeats was demonstrated in the Huntington gene IT15 (4p16.3) in all clinically affected individuals and ranged from 41 to 55 (median 47).

In all three families it was possible to compare the repeat length on HD chromosome with the same chromosome in elderly unaffected individuals. The range of repeat length observed in unaffected members of the families was 17-28 units (median 17).

**Discussion** The diagnosis of HD in these four unrelated families was based on a typical clinical presentation and an autosomal dominant inheritance pattern. Neuro-imaging confirmed the striking caudate atrophy in affected members examined. There was no evidence to suggest other conditions, such as, benign familial chorea, acanthocytosis, tardive dyskinesia or Wilson’s disease.

Analysis of trinucleotide repeats in the Huntington’s gene IT15 (4p 16.3) by means of PCR, confirmed the presence of an expanded repeat within a range of 41-55 in affected members of families I, II and III. The findings in family I were included in the world-wide study of HD mutation.\(^6\) Similar findings were seen among other HD cases from different nations and ethnic groups.\(^5,6,12\) The CAG repeat located in the 5’ region of the HD gene is highly polymorphic in the general population, ranging from 8 to 37 copies, with the vast majority of people (99%) having less than 30 repeat.\(^4\) In contrast, patients with HD have a trinucleotide repeat size ranging from 36 to 121, with a median of 44 repeat.\(^5,12,13\)

Almost all patients with a clinical diagnosis of HD have an expanded CAG repeat.\(^6\) CAG repeat sizes, however, are in the normal range in patients with familial Alzheimer’s disease, schizophrenia, major depression, senile chorea, benign hereditary chorea, neuroacanthocytosis and dentatorubropallidolysian atrophy. CAG expansion in HD gene is a highly specific and sensitive marker for HD.\(^6\)

It is of interest that Scrimgeour and colleagues described two families with clinically diagnosed HD (no DNA linkage studies were done) and they
suggested that the disease gene was transmitted to the Saudi families by European visitors to the Red Sea or the Arabian Gulf. We think this is unlikely; such ethnic intermixing could not be substantiated in our cases. This is particularly unlikely in native nomads. Marriage outside the tribe is a very uncommon practice; it is more likely that a fresh mutation accounted for HD in these families as stated in our reply letter addressing Dr Scrimgeour’s hypothesis. 

The rate of mutation causing new HD, previously deemed to be exceedingly low, is now known to be responsible for up to 3% of affected persons.5,13,15,16 The affected individuals have no family history of the disorder but show typical HD symptoms and sometimes transmit the disorder to the next generation. DNA marker analysis has revealed in some cases that other family members possess the same chromosome 4 homologue but do not display any symptoms of HD. The difference between the unaffected individuals and the new case of HD appears to lie in the length of the CAG repeat. Analysis of DNA in these studies12,15,16 identified intermediate alleles which are CAG alleles greater than that usually seen in the general population (>30), but less than the range seen in patients with HD (<36). These intermediate alleles are sometimes unstable and through the male germline, may expand to the full mutation associated with the clinical phenotype of HD.5,17

The specificity and sensitivity of HD mutation testing has allowed an increasing number of new cases of HD to be recognized and identified all over the world.6 The prevalence of HD in Saudi Arabia is not known. However, in contrast to a previous report from a major teaching hospital18 stating that HD was not seen, this country may harbor many more Huntington’s patients than previously thought.

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References


ياسمه نجف: تقرير لأربع عائلات سعودية
سعودية بو هليقة، دونالد ماكلين، محمد زهير القايمي، صلاح عمر،
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الهدف: إن التقارير عن داء هنتنغتون (ده) من منطقة شبه الجزيرة نادرة.

المكان: مستشفى الملك فيصل التخصصي ومركز الأبحاث.

الدراسة: شُخصت حالات تتبع لأربع عائلات على أنها تحمل سمات (ده) الواسمة وذلك في مركز
رئيسي للعناية الطبية في المملكة العربية السعودية. كانت هذه العائلات جميعًا من أصل محلي واثنتان منها
من أصول البدو. كان الانئتقال الوراثي صفة قاهرة لدى الجمعيات بدأت أعراض المرض في أواسط
العشرونيات من العمر، وتميزت بتدلبات في المزاج والشخصية، تل ذلك تعرّض الحركات وحركات رقصية.

ولم يظهر وجود سوابق من أصل غير محلي إلا في قصة واحدة تزوجت من عائلة مغربية.

النتيجة: أظهر تحليل تكرار سلسلة الحمض النووي (CAG) في موضع مورقة داء هنتنغتون (ITIS) وجود
تزايد في تكرار السلسلة لدى جميع المصابين. كان مدى التكرار من 41 إلى 55 مرات مقارنة بالتكرار
لدى لفنة الضبطة والذي تراوح من 20-28 متوسط 17.

الاستنتاج: تؤيد هذه النتائج الفرضية القائمة بأن الطفرة المسؤولة عن (ده) ليست نادرة كما كان يظن
سابقاً، وأنه قد يوجد في المملكة العربية السعودية (ده) أكثر مما كان يعتقد سابقاً.