Penetrance of the Fragile X–Associated Tremor/Ataxia Syndrome in a Premutation Carrier Population

Sébastien Jacquemont, MD
Randi J. Hagerman, MD
Maureen A. Leehey, MD
Deborah A. Hall, MD
Richard A. Levine, PhD
James A. Brunberg, MD
Lin Zhang, MD, PhD
Tristan Jardini
Louise W. Gane, MS
Susan W. Harris
Kristin Herman, MD
James Grigsby, PhD
Claudia M. Greco, MD
Elizabeth Berry-Kravis, MD, PhD
Flora Tassone, PhD
Paul J. Hagerman, MD, PhD

Context Premutation expansions (55-200 CGG repeats) of the fragile X mental retardation 1 (*FMR1*) gene are frequent in the general population, with estimated prevalences of 1 per 259 females and 1 per 813 males. Several articles have recently described the presence of late-onset neurological symptoms in male carriers of premutation (*FMR1*) alleles. The main clinical features described in this newly identified syndrome are cerebellar ataxia and intention tremor. Additional documented symptoms include short-term memory loss, executive functional deficits, cognitive decline, parkinsonism, peripheral neuropathy, lower-limb proximal muscle weakness, and autonomic dysfunction.

Objective To study the penetrance of the fragile X–associated tremor/ataxia syndrome (FXTAS) among premutation carriers.

Design, Setting, and Participants Family-based study of 192 individuals (premutation carriers and controls) whose families belong to the Northern or Southern California Fragile X Associations. Data were collected (March 2002-April 2003) through a survey and a standardized neurological examination, which was videotaped and subsequently scored in a blinded fashion.

Main Outcome Measures Penetrance of intention tremor and ataxia among adult carriers (aged ≥50 years) of premutation expansions of the *FMR1* gene.

Results Data from the survey of 192 individuals demonstrated an age-related penetrance of the combination of reported intention tremor and gait ataxia in male carriers (17%, 38%, 47%, and 75% [lower-bound estimates] for participants aged 50-59, 60-69, 70-79, and ≥80 years, respectively). The male carrier group had an age-adjusted 13-fold increased risk (95% confidence interval, 3.9-25.4; $P=.003$) of combined intention tremor and gait ataxia when compared with male controls. The clinical examination data from 93 individuals demonstrated that male carriers experienced more difficulties on each of 3 standardized neurological rating scales compared with controls ($P<.05$). Female carrier scores were also higher than those of female controls ($P<.05$) on 2 of the 3 neurological rating scales, but no participant was identified with probable or definite FXTAS.

Conclusions The study demonstrates that older male carriers of premutation alleles of the *FMR1* gene are at high risk of developing FXTAS. Since male premutation carriers are relatively common in the general population, older men with ataxia and intention tremor should be screened for the *FMR1* mutation, especially if these signs are accompanied by parkinsonism, autonomic dysfunction, or cognitive decline, regardless of family history.

JAMA. 2004;291:460-469 www.jama.com

PREMUTATION EXPANSIONS (55-200 CGG repeats) of the fragile X mental retardation 1 (*FMR1*) gene are frequent in the general population, with estimated prevalences of 1 per 259 females and 1 per 813 males.[1,2] The CGG-repeat element is located in the 5′ untranslated region of the *FMR1* gene[3] and is prone to expansion when passed from mothers to their children. A CGG-repeat expansion in excess of 200 repeats (full mutation) generally results in partial or complete silencing of the gene, with subsequent deficiency or absence of *FMR1* protein (FMRP); absence or deficiency of the latter is responsible for fragile X syndrome.[3-5]

Author Affiliations: MIND Institute (Drs Jacquemont, R. Hagerman, and Herman, Mr Jardini, and Mss Gane and Harris) and Departments of Pediatrics (Dr R. Hagerman) and Neurology (Dr Zhang), University of California Davis Medical Center, Sacramento; Departments of Neurology (Drs Leehey and Hall) and Medicine (Dr Grigsby), University of Colorado Health Sciences Center, Denver; Department of Mathematics and Statistics, San Diego State University (Dr Levine), San Diego, Calif; Departments of Radiology (Dr Brunberg), Pathology (Dr Greco), and Biological Chemistry (Drs Tassone and P. Hagerman), University of California, Davis, School of Medicine, Davis; and Department of Pediatrics, Rush-Presbyterian-St Luke's Medical Center, Chicago, Ill (Dr Berry-Kravis).
Corresponding Author and Reprints: Paul J. Hagerman, MD, PhD, Department of Biological Chemistry, University of California, Davis, School of Medicine (Tupper Hall), One Shields Avenue, Davis, CA 95616 (e-mail: pjhagerman@ucdavis.edu).

Carriers of the premutation typically have a normal IQ, although emotional problems such as anxiety are common.[6-11] Approximately 20% of women who carry premutation expansions of the *FMR1* gene experience premature ovarian failure, a clinical finding that is unique to premutation alleles.[12,13]

We recently reported a distinctive form of clinical involvement in some older male carriers of the fragile X premutation.[14-17] These carriers, aged in their 50s and older, develop progressive intention tremor and ataxia. These movement disorders are often accompanied by progressive cognitive and behavioral difficulties, including memory loss, anxiety, deficits of executive function, reclusive or irritable behavior, and dementia. More variable features include parkinsonism, peripheral neuropathy, lower-limb proximal muscle weakness, and autonomic dysfunction (urinary/bowel incontinence and impotence). This disorder has been designated fragile X–associated tremor/ataxia syndrome (FXTAS).[16]

Magnetic resonance imaging in patients with FXTAS reveals approximately symmetric increases in T2-weighted signal intensity in the middle cerebellar peduncles (MCPs) and adjacent cerebellar white matter.[15] This unusual radiological finding, visible in the majority of published cases of premutation carriers with the tremor/ataxia disorder, is not observed in controls. The MCP finding thus potentially serves as a major diagnostic feature of FXTAS.[16]

The most striking neuropathological feature associated with FXTAS, present in all 8 brains examined to date from patients who died with this disorder, is the presence of eosinophilic, intranuclear inclusions in both neurons and astrocytes.[18,19] These inclusions are found throughout the cortex and brain stem, with the greatest densities of inclusions located in the hippocampus and frontal cortical regions. No inclusions have been detected in the Purkinje cells of the cerebellum, although there is evident Purkinje cell dropout and cerebellar axonal degeneration.[18,19] The inclusions are polyglutamine-negative, distinguishing FXTAS from CAG-repeat ataxias. Moreover, no tau or α-synuclein proteins have been found within the inclusions. Intranuclear inclusions that are morphologically similar to the inclusions observed in the FXTAS cases[18] have also been generated in a transgenic mouse model with expanded CGG repeats in the premutation range (approximately 100 CGG repeats in the *Fmr1* gene).[20]

The penetrance of FXTAS among fragile X premutation carriers is unknown, although the frequency of premutation alleles within the general population suggests that this disorder could represent a significant and currently unidentified contribution to the adult tremor and ataxia patient populations. For example, a recent screen of adult ataxia patients in the United Kingdom found that approximately 5% were previously undiagnosed premutation carriers.[21] Therefore, controlled studies are necessary to assess the relationships among the motor, cognitive, and autonomic symptoms and the premutation. To this end, we have conducted a community-based survey and a standardized neurological evaluation on members of families affected by fragile X syndrome in California.

METHODS

All aspects of this study were reviewed and approved by the institutional review board of the University of California, Davis.

Determination of *FMR1* Carrier Status

All parents or grandparents of each fragile X syndrome proband whose genetic status was not previously known through prior genetic testing or pedigree analysis underwent *FMR1* genotyping to document their carrier status. Since CGG repeat number was not a covariate in the current investigation, participants underwent genotyping only when their *FMR1* status (premutation carrier or normal) was not known. Genotype analysis was used to establish carrier status in 50 of 99 carriers (CGG repeats were available for 42 patients; in 8 cases, individuals were tested in other laboratories and exact repeat numbers were not available). The status of all remaining carriers (49/99) was established by pedigree analysis, which documented obligate carrier status in individual pedigrees through additional genotyped children with fragile X syndrome. Genomic DNA was isolated from peripheral blood leukocytes using standard procedures (Puregene kit, Gentra Inc, Minneapolis, Minn). Southern blot and polymerase chain reaction (PCR) analyses were performed on each sample, per Taylor et al,[22] with the exception that for some of the samples the analysis was performed using a digoxygenin-labeled probe (dig-11ddUTP, Roche Diagnostics, Indianapolis, Ind). For Southern blot analysis, the enzymes *Eco*RI and *Nru*I were used for DNA digestion, and StB12.3 was used for the *FMR1*-specific probe.[23] The number of CGG repeats was determined by PCR analysis using primers 1 and 3.[24]

Ascertainment

This study is based on the ascertainment of all fragile X families in California who are members of either the Northern or the Southern California Fragile X Associations. All member families became affiliated with the associations through a child who was diagnosed as having fragile X syndrome (proband); that is, no family was identified through a premutation carrier with neurological problems. All premutation carriers who were members of families residing in California and who were at least 50 years of age qualified for this study. The control groups for both male and female carriers were composed predominantly of the spouses of the carriers (85% of male controls were spouses; 82% of female controls were spouses). The remaining controls were siblings of the carriers who either tested negative for premutation alleles or who were obligate controls (eg, since men cannot inherit an X chromosome from their father, sons of carrier men are obligate controls). Genetic screening was lim-

Table 1. Individuals Satisfying Initial Inclusion Criterion (Aged ≥50 Years) Within Families Identified Through the Southern and Northern California Fragile X Associations

	Age Range, y				
	50-59	60-69	70-79	≥80	Total
Qualifying family members					
Participants					
Male premutation carrier	7	12	17	4	**40**
Male control	22	17	15	5	**59**
Female premutation carrier	26	13	13	7	**59**
Female control	4	16	11	3	**34**
Total					**192**
Unreachable carriers and spouses					
Male premutation carrier	5	1	0	0	**6**
Female premutation carrier	4	2	5	4	**15**
Spouse					**21**
Total					**42**
Nonqualifying family members (absence of genetic information)					
Men					**28**
Women					**36**
Total					**64**

ited to parents and grandparents of the proband for whom status was initially unknown. Uncles, aunts, granduncles, and grandaunts older than 50 years were not included for the initial estimates of penetrance if their genetic status was unknown. Ninety percent of both male and female carriers were parents or grandparents of the fragile X probands. The remaining carriers were uncles, aunts, granduncles, or grandaunts of the proband.

Survey

One hundred twenty-three families belonging to either the Northern or the Southern California Fragile X Association were asked to participate in the study via a letter from their respective association. Upon receiving each family's permission through the associations, we contacted the families by telephone. A total of 23 families were excluded. Four families refused to participate without providing a reason; in 18 families the child with fragile X syndrome, or a parent, was adopted; and in 1 family the individual had lost contact with all members of the family aged 50 years or older. We were unable to contact an additional 11 families (incorrect telephone information).

In the remaining 89 families, 298 individuals aged 50 years or older were identified on the premutation side (predominantly the mother's side) of the family. Sixty-four individuals (28 men and 36 women) did not qualify for participation in the study because they were granduncles or grandaunts whose genetic status was unknown. Of the 234 individuals who did qualify for the study, we were unable to contact 21 known carriers (6 men and 15 women) and their 21 spouses (controls). The 6 male carriers were included in a more conservative penetrance calculation. Thus, 192 (82%) participated in the study (40 male carriers, 59 male spouse controls, 59 female carriers, and 34 female spouse controls) (TABLE 1).

All qualifying participants self-reported symptoms on a survey completed by telephone or in person. For the purpose of the survey, symptoms were scored as present if noticed by the respondent or spouse, with clarification of the questions or characterization of the symptoms provided by the interviewing physician as necessary. The participant provided the final answers. Patients signed informed consent approved by the institutional review board before participating. Those contacted by telephone were read the consent form and gave oral consent.

The survey comprised 54 questions exploring 6 domains: (1) tremor: questions were asked regarding the presence, characteristics, and time of onset of tremors; (2) gait and lower extremities: questions were asked related to the onset of balance problems, recent falls, walking distance, and use of a cane, walker, or wheelchair. Sensory issues were also explored with questions about lower extremity numbness, loss of sensation, and pain; (3) cognitive: questions were asked of the participant or his/her spouse regarding short-term memory loss and episodes of confusion and disorientation; (4) emotional: questions explored anxiety, depression, mood instability, and medications used for those symptoms; (5) other medical issues: questions explored any other medical problem and treatment (in particular, heart problems, stroke, autonomic symptoms, hearing loss, diabetes, and all medications taken by the participant); and (6) family history: questions addressed the presence of a family history positive for neurological disorders, including Parkinson disease, Alzheimer disease, other dementias, and multiple sclerosis.

Responses for cognitive or emotional impairment were not used further for this study.

Finally, participants completed the Tremor Disability Rating Scale (TDRS), designed and validated by Louis et al.[25] This rating scale has 31 questions exploring fine motor tasks. The final (summed) score, converted to a percentage, is the extent of impairment related to the loss of fine motor skills. Zero percent represents absence of impairment and 100% reflects inability to perform any fine motor tasks associated with daily living.

Clinical Assessment

To document the findings of the survey, all participants who were geographically available for clinical assessment (n=93; 24 male carriers, 25 male controls, 26 female carriers, and 18 female controls) underwent a detailed neurological examination that included pertinent items of 3 standardized scales: the Clinical Rating Scale for

Tremors (CRST),[26] the motor section of the Unified Parkinson's Disease Rating Scale (UPDRS),[27] and the International Cooperative Ataxia Rating Scale (ICARS).[28] Whereas the CRST and ICARS are unvalidated, the UPDRS has undergone extensive clinometric analysis. The neurological examination was videotaped and subsequently rated by a neurologist blinded to the patient's diagnosis and DNA study results. The clinician administering the video protocol was not blinded in all cases. However, this individual did not appear on the videotape. The duration of each task was standardized, and the administering clinician did not insist on a longer task interval than what is specified by the protocol. The neurologist rating the videotapes did not use the soundtrack except for the 3 tasks requiring vocalizations.

All patients available for clinical testing underwent the video protocol, which included only half (93/192) of the survey participants. Among the 93 individuals participating in the videotape protocol, 36% had reported 1 or more neurological symptoms on the survey, similar to the 30% figure for all 192 survey participants. Moreover, 58% of the male carriers available for the videotaping had self-reported 1 or more neurological symptoms on the survey, identical to the 58% figure for all 40 male carriers participating in the study. Therefore, there does not appear to be a substantial bias in the population participating in the video protocol.

Throughout this article, we use FXTAS to denote probable or definite FXTAS, as defined by Jacquemont et al.[16] Probable FXTAS requires the presence of both tremor and gait ataxia, whereas definite FXTAS requires a positive MCP finding on magnetic resonance imaging as well as at least 1 major clinical finding (tremor or gait ataxia).

Statistical Methods

Only 16 items on the 54-question survey were subjected to further analysis. This reduction was obtained by (1) removing all questions related to cognition and emotional difficulties, as the evaluation of these symptoms requires the use of appropriate psychometric instruments, which were not a part of the current study; and (2) removing all questions that required more than a closed yes/no answer. Logistic regression analyses were performed on the results of the remaining 16 questions, comparing male carriers with male controls and female carriers with female controls. Results are presented as odds ratios (ORs) and 95% confidence intervals (CIs), with *P* values gauging the significance of the OR for each question. For each item, the analyses are adjusted for age, ie, age is included as a covariate in the regression model. All analyses were 2-tailed.

We tested for interaction, collinearity, and overfitting in the logistic regression and found no violations of regression model assumptions due to collinear variables. The Holm method[29,30] and the Benjamini false discovery rate procedure[31] were applied to control for familywise (multiple outcomes) error rates for the 16 analyses. Since there were only 2 pairs of siblings in the group of 40 male carriers, any correlation of outcomes between these 2 pairs of family members would not bias the conclusions of our regression analyses. The summed TDRS score was analyzed separately. A nonparametric Mann-Whitney test, stratified by age group, was performed to compare the scores obtained by the carriers and controls for each sex.

The clinical rating scale scores were analyzed in 2 ways. First, the results were analyzed according to the total scores of each of the 3 neurological scales. Regression analyses were performed on the summary scores, comparing the square root–transformed mean score of male carriers with male controls and female carriers with female controls. The square root transformation was used to ensure that the assumptions of homoscedastic and Gaussian errors in the regression model are not violated. For these 3 analyses, adjustment for multiple outcomes was unnecessary. Second, the results were analyzed by 12 different subscores for the 3 neurological scales. Three CRST subscores (postural, action, and rest tremor) were computed[26] for the 3 tremor types identified within this scale. Four ICARS subscores were computed for the 4 domains described within this scale.[28] Six UPDRS subscores were also computed for the 6 clinically distinct disability domains previously identified by factor analysis within the UPDRS motor examination.[32,33] However, 1 of the subscores (rigidity) was removed because data on this subgroup could not be obtained in a blinded fashion. Comparisons between the carrier and control groups for each sex were made by the nonparametric Wilcoxon rank sum test. The Holm method[29] and the Benjamini false discovery rate procedure[31] were applied to control for familywise error rates on the basis of the 12 analyses and the tandem walk task, as described by Singer et al.[34]

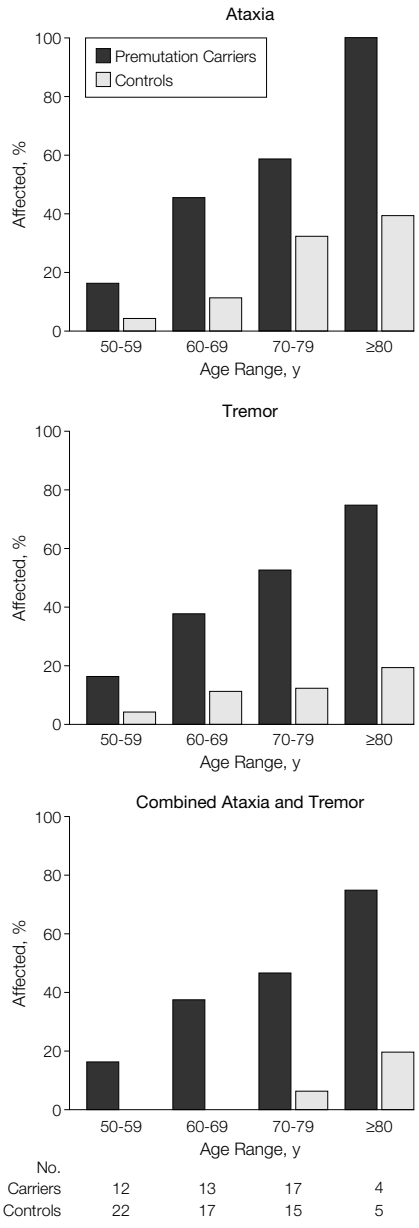
Statistical analyses were performed using S-Plus software, version 6.1 (Insightful Corp, Seattle, Wash). $P<.05$ was considered statistically significant throughout all analyses.

RESULTS

Questionnaire

Among men, in the premutation carrier group (n=40), the prevalence of reported gait instability, tremor, and both symptoms combined increased with age from 50 years to 80 years or older. The penetrance of these symptoms in carriers and controls is displayed in FIGURE 1. To produce more conservative estimates for these penetrance data, we also included the additional 6 male carriers (Table 1) who were unable to participate, stipulating that these cases were all asymptomatic for the lower-bound estimates. The resulting estimates are 17%, 38%, 47%, and 75% of male carriers with self-reported frequencies of combined tremor and gait ataxia within the 50 to 59, 60 to 69, 70 to 79, and 80 years or older age ranges, respectively. The frequency of these reported symptoms was significantly increased in the premutation group rela-

Figure 1. Percentage of Men With Self-Reported Symptoms of Gait Disturbance, Intention Tremor, or Combined Ataxia and Tremor



The percentages in the 50-59 year and 60-69 year age ranges are lower-bound estimates based on the conservative assumption that the 6 male premutation carriers who were unreachable (5 and 1, respectively, for the 2 age ranges; Table 1) were clinically uninvolved.

tive to the control group. For tremor, the age-adjusted OR was 6.8 (95% CI, 2.3-20.5; $P=.001$). For gait instability, the OR was 5.1 (95% CI, 1.9-14; $P=.001$). For both symptoms combined, the OR was 21.2 (95% CI, 4.3-103; $P=.003$) (TABLE 2). Only 2 male controls reported combined tremor and gait ataxia (3%).

The severity of gait difficulty was also greater in the male carrier group. Thirty-five percent of male carriers reported episodes of unexplained falling as opposed to 3% in the male control group (OR, 12.9; 95% CI, 2.6-62.4; $P=.001$) (Table 2). Twenty percent of the male carriers reported using a cane, with a mean onset at 71 years of age, as opposed to 5% in the male control group (at 53, 54, and 80 years). Five male carriers (13%) required a wheelchair at a mean age of 77 years as opposed to 1 male control at 53 years. After multiplicity adjustment using 2 different methods at the .05 level,[29,31] 4 items remained significant: intention tremor, gait instability, falling more than once in the past month without an obvious reason, and the combination of tremor and gait instability.

Among women, the survey did not reveal any significant difference between carriers and controls, either before or after multiplicity adjustment. All frequencies, ORs, and 95% CIs for the survey are reported in Table 2.

Tremor Disability Rating Scale

In men, the mean score obtained on the TDRS (reflecting the mean degree of impairment in fine motor tasks associated with daily living) by the carrier group was 7% among men aged 50 to 59 years and 41% among those aged 80 years or older. In the male control group, the degree of impairment was steady at between 1% and 2% throughout all age ranges. We performed a nonparametric Mann-Whitney test to compare carriers and controls. The age groups of 50 to 59 years and 80 years or older showed significantly higher TDRS scores among the male carriers than among the controls ($P=.01$ and $P<.01$, respectively). The severity of impairment was greater in the male carrier group across all age ranges. A logistic regression analysis was performed comparing impaired premutation carriers with impaired controls after adjusting for age. Impairment was considered as a 2-level factor (normal or mild, score ≤10; severe, score >10). Carriers and controls differed significantly after adjusting for age (OR, 6.45; 95% CI, 1.26-33; $P=.02$).

The scores on the TDRS for women did not differ significantly between the premutation and control groups.

The TDRS has been validated with a control population (aged ≥65 years) and the mean score was 1.6%,[35] which reflects a quasi absence of impairment. This score is not significantly different from our control population (mean scores, 2% for the male control group and 0.3% for the female control group).

Standardized and Videotaped Clinical Protocol

The distribution of the scores for all 3 neurological scales is represented in FIGURE 2. Male premutation carriers showed significant impairment on the 3 scales that measured tremor (CRST), cerebellar dysfunction (ICARS), and parkinsonism (UPDRS). Regression analysis indicated that the average score on a CRST item (ranging from 0 to 3) was significantly higher in the male carrier group relative to the male control group (the relationship was defined through an estimated regression coefficient of 0.18; 95% CI, 0.02-0.35; $P=.04$). Significant differences were also observed on the ICARS (coefficient=0.2; 95% CI, 0.05-0.37; $P=.01$) and UPDRS scales (coefficient=0.2; 95% CI, 0.06-0.37; $P=.01$). In all 3 scales, age was a significant covariate ($P<.01$ for the CRST, $P=.01$ for the ICARS, and $P<.01$ for the UPDRS).

We investigated a total of 12 subscores within the 3 neurological scales, as well as an additional tandem walking task described by Singer et al,[34] to identify tasks that were the most sensitive to the neurological phenotype of the male carriers (TABLE 3). The subscores were compared between the carrier and control groups using the Wilcoxon rank sum test. We examined the 3 components of tremor on the CRST[26]:

Table 2. Results of Age-Adjusted Logistic Regression Analyses of 16 Self-Reported Survey Items*

	Men				Women			
	Frequency, No. (%)				Frequency, No. (%)			
Symptoms	Carriers (n = 40)	Controls (n = 59)	OR (95% CI)	*P* Value	Carriers (n = 59)	Controls (n = 34)	OR (95% CI)	*P* Value
Intention tremors	19 (48)	6 (10)	6.8 (2.3-20.5)	.001†	5 (8)	1 (3)	3.5 (0.3-32.0)	.26
Gait difficulties								
Gait instability	22 (55)	10 (17)	5.1 (1.9-14.0)	.001†	11 (19)	6 (18)	1.1 (0.3-3.7)	.81
Falls‡	14 (35)	2 (3)	12.9 (2.6-62.4)	.001†	8 (14)	1 (3)	5.1 (0.5-43.8)	.13
Cane use	8 (20)	3 (5)	3.5 (0.9-14.9)	.08	4 (7)	2 (6)	1.0 (0.1-7.0)	.98
Wheelchair use	5 (13)	1 (2)	6.0 (0.6-56.3)	.11	1 (2)	0		
Tremor and gait combined	18 (45)	2 (3)	21.2 (4.3-103.3)	.003†	3 (5)	0		
Other medical problems								
Heart problems	8 (20)	4 (7)	3.1 (0.8-11.4)	.09	6 (10)	4 (12)	0.91 (0.2-3.6)	.89
Hypertension requiring treatment	9 (23)	19 (32)	0.3 (0.1-1.0)	.06	19 (32)	18 (53)	0.5 (0.2-1.2)	.13
Stroke	4 (10)	1 (2)	5.4 (0.5-52.1)	.14	2 (3)	0		
Impotence	11 (28)	8 (14)	1.9 (0.6-5.5)	.22	NA	NA		
Incontinence	10 (25)	5 (8)	2.9 (0.9-9.8)	.07	8 (14)	7 (21)	0.7 (0.2-2.2)	.54
Loss of sensation in lower extremities	9 (23)	9 (15)	1.3 (0.4-3.7)	.63	7 (12)	5 (15)	0.9 (0.2-3.1)	.87
Seizures	0	1 (2)			0	0		
Hearing loss	13 (33)	19 (32)	0.7 (0.3-1.8)	.54	4 (7)	9 (26)	0.1 (0.05-0.77)	.02
Diabetes requiring treatment	8 (20)	5 (8)	2.3 (0.6-8.0)	.17	3 (5)	3 (9)	0.53 (0.1-2.9)	.47
Family history of neurological disorder	9 (23)	7 (12)	2.4 (0.7-7.5)	.12	21 (36)	7 (21)	2.1 (0.7-5.7)	.14

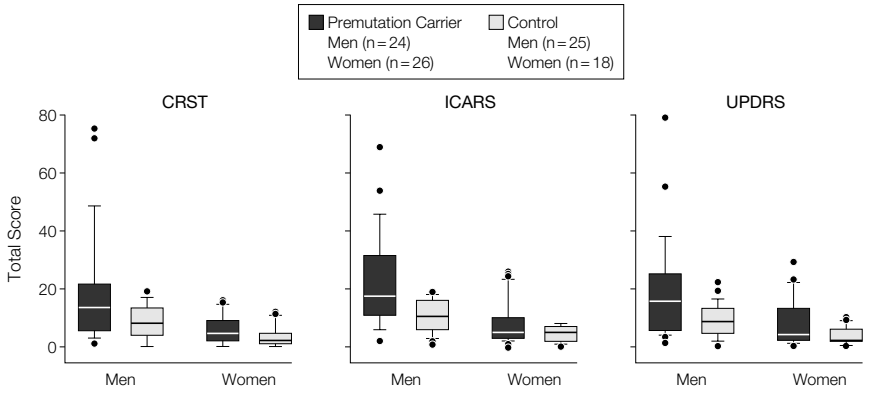
Abbreviations: CI, confidence interval; NA, not applicable; OR, age-adjusted odds ratio of symptom frequencies in carriers vs controls.
*Age was included as a covariate in the logistic regression model. Data are based solely on individuals who participated in the survey (Table 1). Items with too few scores (most or all controls) to yield meaningful *P* values do not show ORs, CIs, or *P* values.
†Items were statistically significant at the .05 level after multiplicity correction on the basis of 16 separate outcome measures, using both the Holm method[29] and the Benjamini false discovery rate.[31]
‡Fell more than once in the past month without an obvious reason.

rest, postural, and kinetic tremors (Table 3). In contrast with the total CRST score, male carriers did not show significant impairment in any of the 3 subscores.

Similarly, we analyzed the 4 categories of the original ICARS rating tool[28]: postural/gait/stance, limb movement, speech, and oculomotor movements (Table 3). Male carriers were significantly more impaired relative to male controls in all subgroups except oculomotor movements.

Based on previously published factor analysis data on the UPDRS,[32,33] we examined 5 stable factors: right bradykinesia, left bradykinesia, midline bradykinesia/balance, rest tremor, and postural tremor. Only 1 subscore, midline/gait/balance, was significantly different between male carriers and controls. Right bradykinesia was nearly significant (*P* = .06). The tandem walking task described by Singer et al,[34] not part of any of the 3 scales, was analysed as a 13th subscore. It showed significant impairment in the male carrier group compared with controls (Table 3). However, after multiple-outcomes correction using the methods of both Holm[29] and Benjamini et al[31], only 1 item (tandem walking) remained significant at the .05 level.

Figure 2. Distribution of CRST, ICARS, and UPDRS Total Scores, by Sex and Carrier Status



CRST indicates Clinical Rating Scale for Tremors (score range, 0-120); ICARS, International Cooperative Ataxia Rating Scale (score range, 0-100); and UPDRS, Unified Parkinson's Disease Rating Scale (score range, 0-108). The horizontal line in the middle of each box indicates the median, while the top and bottom borders of the box mark the 75th and 25th percentiles, respectively. The whiskers above and below the box mark the 90th and 10th percentiles. The points beyond the whiskers are outliers beyond the 90th or 10th percentiles.

The distribution of the scores among women for all 3 neurological scales is represented in Figure 2. Only 3 women, aged 69, 81, and 84 years, showed very mild intention tremor and/or gait ataxia. However, the regression analysis indi-

Table 3. Summary of CRST, ICARS, and UPDRS Components by Carrier Status Among Men*

	Median (Range)		
	Carriers	Controls	*P* Value†
CRST			
Action/intention	11 (0-49)	8 (0-16)	.06
Postural tremor	2 (0-14)	1 (0-5)	.24
Rest tremor	0 (0-15)	0 (0-1)	.09
ICARS			
Limb ataxia	8 (0-25)	4 (0-12)	.02
Postural/gait/stance	5 (1-32)	3 (0-8)	.02
Oculomotor	1 (0-3)	2 (0-3)	.30
Speech	2 (0-6)	1 (0-4)	.04
UPDRS			
Midline/gait/balance	4 (0-32)	2 (0-8)	.01
Bradykinesia			
Right	4 (0-10)	2 (0-7)	.06
Left	4 (0-12)	3 (0-6)	.20
Postural tremor	2 (0-8)	2 (0-4)	.10
Rest tremor	0 (0-9)	0 (0-1)	.30
Tandem walking[34]	1 (0-4)	0 (0-2)	.003‡

Abbreviations: CRST, Clinical Rating Scale for Tremor; ICARS, International Cooperative Ataxia Rating Scale; UPDRS, Unified Parkinson's Disease Rating Scale.
*Summary data and analyses for women are not shown because of absence of significant differences between carriers and controls on all subscores.
†Analyzed by Wilcoxon rank sum test. This nonparametric analysis did not allow for age correction. The comparison of the age distribution using the χ^2 (parametric) and Kolmogorov-Smirnov (nonparametric) tests indicated no significant difference between the 2 groups (*P* values of .55 and .99, respectively).
‡This item remained statistically significant at the .05 level after multiplicity correction on the basis of 13 different analyses, using both the Holm method[29] and the Benjamini false discovery rate.[31]

cated that the average scores for 1 item from the UPDRS and ICARS (ranging from 0 to 3) were higher in the female carrier group and significantly different from the female control group (coefficient = 0.19; 95% CI, 0.04-0.34; *P* = .02 for the UPDRS; coefficient = 0.71; 95% CI, 0.1-1.28; *P* = .02 for the ICARS). Age was a significant covariate for both scales ($P<.01$). The CRST did not show significant differences between the 2 groups (coefficient = 0.12; 95% CI, –0.003 to 0.24; *P* = .07), and female carriers did not show significant impairment in any of the 12 subscores or on the tandem walking task.

To evaluate the reliability of the self-reported symptoms on the survey, we performed a Wilcoxon rank sum test comparing the scores obtained on the survey with scores of the neurological evaluation. Participants who reported tremor scored higher (median, 14; interquartile range [IQR], 11-24) on the CRST than those who reported no tremor (median, 7; IQR, 3-12; $P<.01$). The participants who reported gait instability scored higher (median, 20; IQR, 11-35) on the ICARS than those who reported no gait instability (median, 11; IQR, 6-15; $P<.01$). Participants who reported combined tremor and gait instability scored higher (median, 37; IQR, 24-66) on the combined CRST and ICARS than those who reported no tremor and no gait instability (median, 18; IQR, 9-27; $P<.01$).

COMMENT

The current investigation provides initial estimates for the penetrance of self-reported gait difficulties and intention tremor among carriers of the fragile X premutation. The penetrance of this tremor/ataxia disorder (corresponding to probable FXTAS as diagnosed clinically)[16] is defined as the fraction of carriers who self-reported intention tremor and gait instability combined. The data obtained from the survey, representing a total ascertainment of families enrolled in the Northern and Southern California Fragile X Associations, yielded a lower-bound estimate of the age-related penetrance of probable FXTAS in male carriers that ranged from 17% (aged 50-59 years) to 75% (aged ≥80 years) and averaged 39% for the male carrier group (aged ≥50 years) as a whole. However, the elevated penetrance of the group aged 80 years or older is based on a small sample size (n = 4) (Table 1). Applying the correction of Zhang and Yu[36] to the age-adjusted OR (21.2; 95% CI, 4.3-103.3) for combined tremor and gait ataxia (Table 2), the relative risk of combined major clinical symptoms defining the diagnosis of probable FXTAS is 13.2 times higher (95% CI, 3.9-25.4; *P* = .003) in the male carrier group than in the male control group. However, the CIs are large because of small sample size, especially in participants aged 80 years or older. Larger samples are required to generate more accurate penetrance data and ORs. The severity of symptoms was variable within the carrier group, and 25% of the participating male carriers experienced significant disability related to ambulation (requiring a wheelchair) and/or fine motor skills (significant writing impairment on the TDRS), as opposed to 3% in the male control group.

The data from the survey reveal that gait instability was the most common symptom reported by male carriers, involving 17% (aged 50-59 years) to 100% (aged ≥80 years) of that group, in agreement with an earlier report.[16] The penetrance and severity of intention tremor is higher in the male carrier group relative to controls across all age ranges (OR, 6.8; 95% CI, 2.3-20; $P<.01$). Again, the CIs are wide, and larger groups will be needed to produce more accurate ORs. In the male control group, the mean self-reported penetrance of intention tremor is 10%, which is similar to the results of epidemiological studies of essential tremor for the same age range.[37] In contrast, 48% of the male premutation group self-reported intention tremors (Table 2). Intention tremor is also a major finding in the study by Berry-Kravis et al.[38] In addition, as shown by the TDRS, the degree of impairment due to intention tremor reported in the control group was lower than that reported by the carrier group (*P* = .03).

The penetrance data in this study are all derived from the survey. To correlate the data obtained by the self-report method with the degree of clinical involvement, we analyzed the scores obtained on both the survey and the neurological evaluation using nonparametric regression. Significant correlations were demonstrated between self-reported tremor and the CRST score (100% increase in the summary scores of the CRST; $P<.01$) and between self-reported gait instability and the ICARS score (75% increase in the summary scores of the ICARS; $P<.01$). The high level of concordance between the survey and the clinical evaluation established that the self-report method in this study was clinically meaningful. Clinical follow-up of the 24 male carriers who had participated in the video protocol revealed that 10 of the 11 carriers who had self-reported the combination of tremor and gait instability demonstrated these symptoms on the clinical examination. One participant was asymptomatic on the clinical examination but claimed to have intermittent symptoms. This suggests that a small fraction of participants overreported symptoms or that an intermittent and mild presentation predates the appearance of clinically recognizable symptoms.

The results of the clinical assessment reported here confirm the preliminary findings of a study by Berry-Kravis et al[38] for a smaller group of individuals (n=37). Those investigators reported significantly higher scores in male carriers vs male controls on the ICARS and CRST. For their study, significance was reached on the UPDRS only when comparing the male carrier group with the 3 other groups combined (male controls, female carriers, and female controls), most likely because of the small sample size. The current study, with larger sample sizes, demonstrates a significant difference between male premutation carriers and controls for the UPDRS.

The clinical data also reveal that a limited number of subscores are sensitive enough to distinguish carriers from controls (Table 3). The subscores with significant *P* values reflect either cerebellar symptoms (ICARS) or gait difficulties (midline/gait/balance in the UPDRS and tandem walking[34]). None of the CRST subscores (postural, intention, or action tremor) was significantly different between groups. In addition, after correction for multiple outcomes, none of the individual subtests, other than tandem walking, retained significance at the .05 level. Larger populations will be necessary to assess significance for each of the subtests.

The statistical analysis demonstrates that the overall scores of the UPDRS and ICARS are significantly higher in female carriers than controls ($P=.02$ for UPDRS and ICARS); however, the mean scores of the female carrier group did not differ from those of the male control group. The subclinical neurological involvement of female carriers of the premutation thus needs to be evaluated further with larger clinical studies. We have documented the rare occurrence of FXTAS symptoms in women outside of California.[39-41] However, the occurrence of FXTAS in women appears to be far less common than in men.

The skewed sex ratio observed in this study supports the concept of an X-linked disorder,[14,16,38] although earlier reports[42-49] failed to reach a consensus regarding the sex ratios of intention tremor, gait ataxia, and parkinsonism in the general population. For essential tremor, several studies have reported contradictory results regarding sex ratio.[37,42-44] Most surveys with a personal (as opposed to medical record–based) screening for parkinsonism found no significant sex bias.[45-49]

The neurological disorder with the clinical presentation most closely resembling FXTAS is multiple system atrophy (MSA). Both MSA and FXTAS present with variable features of parkinsonism, gait ataxia, and autonomic dysfunction. Both also share the very distinctive neuroradiological sign of an increased T2 signal of the MCPs and adjacent cerebellar white matter. Multiple system atrophy is a relatively rare disorder. Prevalence in individuals aged 65 years or older is estimated to be approximately 4.4 per 100 000.[50] Therefore, a separate etiology leading to MSA in more than 1 of these patients would be extremely unlikely. However, the molecular underpinnings of FXTAS, related to the *FMR1* gene, may be contributing to the population of patients currently diagnosed as having MSA.[51]

The relatively small number of individuals (n=234) initially qualifying for this study reflects the distribution of the *FMR1* premutation alleles in a family: 1 of 4 grandparents is a carrier, and only 1 of 2 siblings of this grandparent is a carrier. No genetic screening was conducted with siblings of the probands' parents/grandparents; only granduncles/grandaunts who were identified as obligate carriers or who had been previously tested qualified for this study (this latter situation arises when granduncles/grandaunts also had grandchildren with fragile X syndrome).

There are at least 2 sources of known or potential ascertainment bias in the current family-based study. First, all premutation carriers were identified through families with at least 1 child with fragile X syndrome. This mode of ascertainment would bias the allele distribution toward larger premutation alleles, since there is a steeply increasing likelihood for allelic expansion into the full mutation range as the allele size increases from 60 CGG repeats to approximately 90 repeats; 59 repeats is the smallest allele known to expand to the full mutation range within 1 generation.[52] Alleles in the gray zone (41-54 repeats) are highly unlikely to result in full mutation alleles in 2 generations (grandchildren of the affected carriers).[52,53] The smallest allele we have observed thus far for a neurologically affected adult carrier within a known fragile X family is 69 CGG repeats,[16] and the smallest allele observed with the neuropathological changes associated with FXTAS is 70 repeats.[18,19] Both of these observations are close to the expected lower limit of approximately 59 CGG repeats for ascertainment within a fragile X syndrome–affected family.[52] Second, for premutation alleles much greater than 100 CGG repeats, there is a greater chance

of emotional, behavioral, and cognitive involvement in carriers, which may reduce their chances of having offspring or their willingness/availability for participation in screening studies.[54] We have not yet investigated the penetrance of FXTAS as a function of CGG repeat length. However, this last issue is of great mechanistic importance and is currently under study.

Based on the expectation that a measurable fraction of adults seen in adult neurology clinics with intention tremor and/or ataxia will be undiagnosed carriers, several groups have initiated screens of selected patients in movement disorders clinics. Macpherson et al[21] recently reported that 3 individuals (5%) within a group of 59 ataxia patients with no family history of fragile X syndrome possessed gray zone or premutation alleles (51, 66, and 81 CGG repeats). Those results provide significant evidence of association between the ataxia phenotype and expanded *FMR1* alleles. Further studies of tremor and/or ataxia populations are thus warranted for the purpose of better defining the neurological consequences of expanded *FMR1* alleles in the absence of biasing influences of ascertainment through known fragile X families.

CONCLUSION

The current study demonstrates that male carriers of the fragile X premutation are at high risk of developing FXTAS, a neurological disorder characterized by gait ataxia, intention tremor, parkinsonism, and cognitive decline. The penetrance of this disorder increases with age, suggesting that the preponderance of male carriers may eventually develop neurological dysfunction.

With a published prevalence of 1 in 813 (95% CI, 1/527 to 1/1781) for male carriers of premutation (*FMR1*) alleles in the general population,[2] the prevalence of FXTAS in men aged 50 years or older may approach approximately 1 in 3000, which would make this recently identified disorder one of the more common single-gene forms of tremor and ataxia in the aging population. Patients with late-onset ataxia and intention tremor (eg, MSA, prevalence of approximately 1/20 000[50]; or atypical Parkinson disease, approximately 1/3000[55]) should therefore be screened for premutation expansions of the *FMR1* gene.

Author Contributions: Dr P. Hagerman had full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.
Study concept and design: Jacquemont, R. Hagerman, Leehey, Levine, Jardini, Berry-Kravis, P. Hagerman.
Acquisition of data: Jacquemont, R. Hagerman, Leehey, Levine, Zhang, Jardini, Gane, Harris, Herman, Grigsby, Greco, Tassone.
Analysis and interpretation of data: Jacquemont, R. Hagerman, Leehey, Hall, Levine, Brunberg, Jardini, Grigsby, Greco, P. Hagerman.
Drafting of the manuscript: Jacquemont, R. Hagerman, Leehey, Levine, P. Hagerman.
Critical revision of the manuscript for important intellectual content: Jacquemont, R. Hagerman, Leehey, Hall, Levine, Brunberg, Zhang, Jardini, Gane, Harris, Herman, Grigsby, Greco, Berry-Kravis, Tassone, P. Hagerman.
Statistical expertise: Levine.
Obtained funding: R. Hagerman, Gane, Berry-Kravis, P. Hagerman.
Administrative, technical, or material support: R. Hagerman, Leehey, Brunberg, Zhang, Jardini, Harris, Grigsby, P. Hagerman.
Study supervision: R. Hagerman, Leehey, P. Hagerman.
Funding/Support: This work was supported by National Institute of Child Health and Human Development grant HD 36071 to Dr R. Hagerman for the study of genotype-phenotype relationships associated with the fragile X gene and National Institute of Neurological Disorders and Stroke grant NS 43532 to Dr P. Hagerman for the characterization of the tremor/ataxia disorder. The Lavoisier Fund, Paris, France, supported the first year of a fellowship for Dr Jacquemont and the University of California, Davis, MIND Institute provided additional fellowship support for Dr Jacquemont.
Role of the Sponsor: The funding sources had no role in the design and conduct of the study; the collection, analysis, and interpretation of the data; or the preparation, review, or approval of the manuscript.
Acknowledgment: We thank Elisabeth Becker for help with the organizational aspects of this research and for preparation of the manuscript and Lexin Li, PhD, for help with statistical analysis.

REFERENCES

1. Rousseau F, Rouillard P, Morel ML, Khandjian EW, Morgan K. Prevalence of carriers of premutation-size alleles of the *FMRI* gene—and implications for the population genetics of the fragile X syndrome. *Am J Hum Genet.* 1995;57:1006-1018.
2. Dombrowski C, Levesque S, Morel ML, Rouillard P, Morgan K, Rousseau F. Premutation and intermediate-size *FMR1* alleles in 10572 males from the general population: loss of an AGG interruption is a late event in the generation of fragile X syndrome alleles. *Hum Mol Genet.* 2002;11:371-378.
3. Verkerk AJ, Pieretti M, Sutcliffe JS, et al. Identification of a gene (*FMR-1*) containing a CGG repeat coincident with a breakpoint cluster region exhibiting length variation in fragile X syndrome. *Cell.* 1991; 65:905-914.
4. Tassone F, Hagerman RJ, Ikle DN, et al. FMRP expression as a potential prognostic indicator in fragile X syndrome. *Am J Med Genet.* 1999;84:250-261.
5. Hagerman RJ, Hagerman PJ, eds. *Fragile X Syndrome: Diagnosis, Treatment, and Research.* 3rd ed. Baltimore, Md: Johns Hopkins University Press; 2002.
6. Dorn MB, Mazzocco MM, Hagerman RJ. Behavioral and psychiatric disorders in adult male carriers of fragile X. *J Am Acad Child Adolesc Psychiatry.* 1994; 33:256-264.
7. Reiss AL, Freund L, Abrams MT, Boehm C, Kazazian H. Neurobehavioral effects of the fragile X premutation in adult women: a controlled study. *Am J Hum Genet.* 1993;52:884-894.
8. Franke P, Leboyer M, Gansicke M, et al. Genotype-phenotype relationship in female carriers of the premutation and full mutation of *FMR-1*. *Psychiatry Res.* 1998;80:113-127.
9. Sobesky WE, Taylor AK, Pennington BF, et al. Molecular/clinical correlations in females with fragile X. *Am J Med Genet.* 1996;64:340-345.
10. Loesch DZ, Huggins RM, Bui QM, Epstein JL, Taylor AK, Hagerman RJ. Effect of the deficits of fragile X mental retardation protein on cognitive status of fragile X males and females assessed by robust pedigree analysis. *J Dev Behav Pediatr.* 2002;23:416-423.
11. Riddle JE, Cheema A, Sobesky WE, et al. Phenotypic involvement in females with the *FMR1* gene mutation. *Am J Ment Retard.* 1998;102:590-601.
12. Schwartz CE, Dean J, Howard-Peebles PN, et al. Obstetrical and gynecological complications in fragile X carriers: a multicenter study. *Am J Med Genet.* 1994;51:400-402.
13. Allingham-Hawkins DJ, Babul-Hirji R, Chitayat D, et al. Fragile X premutation is a significant risk factor for premature ovarian failure: the International Collaborative POF in Fragile X study—preliminary data. *Am J Med Genet.* 1999;83:322-325.
14. Hagerman RJ, Leehey M, Heinrichs W, et al. Intention tremor, parkinsonism, and generalized brain atrophy in male carriers of fragile X. *Neurology.* 2001; 57:127-130.
15. Brunberg JA, Jacquemont S, Hagerman RJ, et al. Fragile X premutation carriers: characteristic MR imaging findings of adult male patients with progressive cerebellar and cognitive dysfunction. *AJNR Am J Neuroradiol.* 2002;23:1757-1766.
16. Jacquemont S, Hagerman RJ, Leehey M, et al. Fragile X premutation tremor/ataxia syndrome: molecular, clinical, and neuroimaging correlates. *Am J Hum Genet.* 2003;72:869-878.
17. Leehey MA, Munhoz RP, Lang AE, et al. The fragile X premutation presenting as essential tremor. *Arch Neurol.* 2003;60:117-121.
18. Greco CM, Hagerman RJ, Tassone F, et al. Neuronal intranuclear inclusions in a new cerebellar tremor/ataxia syndrome among fragile X carriers. *Brain.* 2002; 125:1760-1771.
19. Hagerman P, Iwahashi C, Babineau B, et al. Fragile X-associated Tremor/Ataxia Syndrome (FXTAS): a common heritable neuronal inclusion disorder [abstract]. *Neurology.* 2003;60:A469.
20. Willemsen R, Hoogeveen-Westerveld M, Reis S, et al. The *FMR1* CGG repeat mouse displays ubiquitin-positive intranuclear neuronal inclusions; implications for the cerebellar tremor/ataxia syndrome. *Hum Mol Genet.* 2003;12:949-959.
21. Macpherson J, Waghorn A, Hammans S, Jacobs P. Observation of an excess of fragile-X premutations in a population of males referred with spinocerebellar ataxia. *Hum Genet.* 2003;112:619-620.
22. Taylor AK, Safanda JF, Fall MZ, et al. Molecular predictors of cognitive involvement in female carriers of fragile X syndrome. *JAMA.* 1994;271:507-514.
23. Oberle I, Rousseau F, Heitz D, et al. Instability of a 550-base pair DNA segment and abnormal methylation in fragile X syndrome. *Science.* 1991;252: 1097-1102.
24. Fu YH, Kuhl DP, Pizzuti A, et al. Variation of the

CGG repeat at the fragile X site results in genetic instability: resolution of the Sherman paradox. *Cell.* 1991; 67:1047-1058.
25. Louis ED, Barnes LF, Wendt KJ, et al. Validity and test-retest reliability of a disability questionnaire for essential tremor. *Mov Disord.* 2000;15:516-523.
26. Fahn S, Tolosa E, Marin C. Clinical Rating Scale for Tremor. In: Jankovic JJ, Tolosa E, Scott-Connor CEH, eds. *Parkinson's Disease and Movement Disorders*. Baltimore-Munich: Urban & Schwarzenberg; 1998: 225-234.
27. Fahn S, Elton RL, for the UPDRS Development Committee. Unified Parkinson's Disease Rating Scale. In: Fahn S, Marsden CD, Calne DB, Goldstein M, eds. *Recent Developments in Parkinson's Disease*. Florham Park, NJ: Macmillan Health Care Information; 1987:153-164.
28. Trouillas P, Takayanagi T, Hallett M, et al, for the Ataxia Neuropharmacology Committee of the World Federation of Neurology. International Cooperative Ataxia Rating Scale for pharmacological assessment of the cerebellar syndrome. *J Neurol Sci.* 1997;145: 205-211.
29. Holm S. A simple sequentially rejective multiple test procedure. *Scand J Stat.* 1979;6:65-70.
30. Aickin M, Gensler H. Adjusting for multiple testing when reporting research results: the Bonferroni vs Holm methods. *Am J Public Health.* 1996;86:726-728.
31. Benjamini Y, Drai D, Elmer G, Kafkafi N, Golani I. Controlling the false discovery rate in behavior genetics research. *Behav Brain Res.* 2001;125:279-284.
32. Stebbins GT, Goetz CG. Factor structure of the Unified Parkinson's Disease Rating Scale: motor examination section. *Mov Disord.* 1998;13:633-636.
33. Stebbins GT, Goetz CG, Lang AE, Cubo E. Factor analysis of the motor section of the unified Parkinson's disease rating scale during the off-state. *Mov Disord.* 1999;14:585-589.
34. Singer C, Sanchez-Ramos J, Weiner WJ. Gait abnormality in essential tremor. *Mov Disord.* 1994;9: 193-196.
35. Louis ED, Wendt KJ, Albert SM, Pullman SL, Yu Q, Andrews H. Validity of a performance-based test of function in essential tremor. *Arch Neurol.* 1999; 56:841-846.
36. Zhang J, Yu KF. What's the relative risk? a method of correcting the odds ratio in cohort studies of common outcomes. *JAMA.* 1998;280:1690-1691.
37. Louis ED, Marder K, Cote L, et al. Differences in the prevalence of essential tremor among elderly African Americans, whites, and Hispanics in northern Manhattan, NY. *Arch Neurol.* 1995;52:1201-1205.
38. Berry-Kravis E, Lewin F, Wuu J, et al. Tremor and ataxia in fragile X premutation carriers: blinded videotape study. *Ann Neurol.* 2003;53:616-623.
39. Tassone F, Orrico A, Galli L, Jacquemont S, Hagerman RJ, Hagerman PJ. Spastic paraparesis and ataxia in two sisters with the fragile X premutation. Paper presented at: 8th International Fragile X Conference; July 17-21, 2002; Chicago, Ill.
40. Hagerman R, Zhang L, Brunberg J, et al. Two female cases of the fragile X premutation tremor/ataxia syndrome: cognitive, molecular, and radiological studies [abstract]. *Neurology.* 2003;60:A331.
41. Leehey MA, Grigsby J, Rubinstein D, et al. Parkinsonism, tremor and ataxia in a female fragile X carrier [abstract]. *Mov Disord.* 2002;17:S351.
42. Rajput AH, Offord KP, Beard CM, Kurland LT. Essential tremor in Rochester, Minnesota: a 45-year study. *J Neurol Neurosurg Psychiatry.* 1984;47:466-470.
43. Haerer AF, Anderson DW, Schoenberg BS. Prevalence of essential tremor: results from the Copiah County study. *Arch Neurol.* 1982;39:750-751.
44. Bharucha NE, Bharucha EP, Bharucha AE, Bhise AV, Schoenberg BS. Prevalence of essential tremor in the Parsi community of Bombay, India. *Arch Neurol.* 1988;45:907-908.
45. Schoenberg BS, Anderson DW, Haerer AF. Prevalence of Parkinson's disease in the biracial population of Copiah County, Mississippi. *Neurology.* 1985;35: 841-845.
46. Tandberg E, Larsen JP, Nessler EG, Riise T, Aarli JA. The epidemiology of Parkinson's disease in the county of Rogaland, Norway. *Mov Disord.* 1995;10:541-549.
47. Kis B, Schrag A, Ben-Shlomo Y, et al. Novel three-stage ascertainment method: prevalence of PD and parkinsonism in South Tyrol, Italy. *Neurology.* 2002; 58:1820-1825.
48. Schrag A, Ben-Shlomo Y, Quinn NP. Cross sectional prevalence survey of idiopathic Parkinson's disease and Parkinsonism in London. *BMJ.* 2000;321: 21-22.
49. Ho SC, Woo J, Lee CM. Epidemiologic study of Parkinson's disease in Hong Kong. *Neurology.* 1989; 39:1314-1318.
50. Schrag A, Ben-Shlomo Y, Quinn NP. Prevalence of progressive supranuclear palsy and multiple system atrophy: a cross-sectional study. *Lancet.* 1999; 354:1771-1775.
51. Zhang L, Leehey M, Wheelock V, Tassone F, Hagerman R, Hagerman P. A subtype of multiple system atrophy with cerebellar ataxia (MSA-C): the fragile X-associated tremor/ataxia syndrome. *Neurology.* 2003; 60:A470.
52. Nolin SL, Brown WT, Glicksman A, et al. Expansion of the fragile X CGG repeat in females with premutation or intermediate alleles. *Am J Hum Genet.* 2003;72:454-464.
53. Murray A, Macpherson JN, Pound MC, et al. The role of size, sequence and haplotype in the stability of FRAXA and FRAXE alleles during transmission. *Hum Mol Genet.* 1997;6:173-184.
54. Johnston C, Eliez S, Dyer-Friedman J, et al. Neurobehavioral phenotype in carriers of the fragile X premutation. *Am J Med Genet.* 2001;103:314-319.
55. de Rijk MC, Tzourio C, Breteler MM, et al. Prevalence of parkinsonism and Parkinson's disease in Europe: the EUROPARKINSON Collaborative Study. *J Neurol Neurosurg Psychiatry.* 1997;62:10-15.

A belief is not true because it is useful.
—Henri Frederic Amiel (1821-1881)