Original Research Article

Dementia and Geriatric Cognitive Disorders

Dement Geriatr Cogn Disord 2005;19:246–255
DOI: 10.1159/000084549

Accepted: September 9, 2004
Published online: March 18, 2005

Presymptomatic Signs in Healthy CJD Mutation Carriers

Ariela Gigi[a] Eli Vakil[b] Ester Kahana[c] Uri Hadar[d]

[a]Psychobiology Research Unit and [d]Cognitive Unit, Department of Psychology, Tel-Aviv University, Tel Aviv; [b]Neuropsychology Department, Bar-Ilan University, and [c]Neurology Department, Barzilai Medical Center, Ashkelon, Israel

Key Words

Creutzfeldt-Jacob disease · Anxiety · Dementia · Preclinical signs · PRNP gene · Neuropsychology · Prion mutation

Abstract

Creutzfeldt-Jacob disease (CJD) is a rapidly progressing dementia with neurological, psychiatric and cognitive symptoms. We focused our study on the familial CJD form among Libyan Jews (the E200K mutation), trying to identify preclinical neuropsychological signs in mutation carriers to facilitate early diagnosis of the disease. A wide range of neuropsychological tests was administered to 27 healthy volunteers, all first-degree relatives of genetic CJD patients. Thirteen of our participants were gene mutation carriers (E200K) and 14 controls. The healthy mutation carriers reported significantly lower Trait and higher State anxiety scores. Repeated Measure analysis showed statistical significance. The Anxiety Index (State-Trait Anxiety Score) progressed with age in the carriers' group but not in the controls. Since this was more pronounced in the older subjects, we suggest that abnormal stress mechanisms precede the clinical onset of CJD. Cognitive differences have also been found between carriers and controls, especially in visual recognition of pictured objects. Both kinds of differences (anxiety levels and cognitive deficits) were most pronounced in elderly subjects. This study is the first to show any dysfunction in healthy CJD mutation carriers.

Copyright © 2005 S. Karger AG, Basel

Scientific Background

Creutzfeldt-Jacob Disease (CJD) is a fatal neurodegenerative disorder, the most prevalent of the human prion diseases. CJD often has a sudden onset and it is ultimately terminal, usually within a few months [49]. The clinical characterization of CJD is heterogeneous and includes diverse neurological and psychiatric signs, such as motor deficits, visual disturbances, visual hallucinations, personality changes and vigilance disturbances, as well as anxiety and depression. However, the major clinical symptom is a rapidly progressing dementia [31, 32, 35, 55].

The pathogenesis of CJD is believed to relate to the accumulation of an abnormal prion protein isoform (PrP^{Sc}) of the normally occurring cellular protein (PrP^{c}) in the central nervous system [49]. The disease may occur sporadically or due to infection, but a certain percentage of the known cases is associated with a familial autosomal dominant point mutation in the PRNP gene [35, 37]. Several different mutations in the PRNP gene have been

KARGER

Fax +41 61 306 12 34
E-Mail karger@karger.ch
www.karger.com

© 2005 S. Karger AG, Basel
1420–8008/05/0196–0246$22.00/0

Accessible online at:
www.karger.com/dem

Dr. Ariela Gigi
The Joseph Sagol Neuroscience Research Center, The Chaim Sheba Medical Center
Tel Hashomer 5621 (Israel)
Tel. +972 3 5304753, Fax +972 3 5304752
E-Mail Ariela.Gigi@sheba.health.gov.il

shown to concentrate in pedigrees in which familial prion diseases are common. One of the largest clusters of CJD is reported among Jews of Libyan decent. In this community, the incidence of CJD is 100 times higher than usual and is the highest in the world [33]. The disease in this community is linked to a dominant point mutation at codon 200 of the PRNP gene (E200K) [7, 24]. The cumulative risk for developing CJD among E200K mutation carriers is negligible at the age of 30 and reaches 80–100% by the age of 80 [6]. Still, the variability of age at disease onset and the sudden and very rapid onset of the disease make it difficult to study the factors that enable the expression of, or trigger, the familial form of the disease.

Dementia accompanies many disorders with different etiologies and does not constitute a homogeneous diagnostic entity. Irrespective of the underlying pathological process, the symptoms are usually determined by the locus of brain dysfunction rather than the type of the disease [28, 36]. Of course, certain types of dementia may compromise particular groups of cells and produce consistent cognitive profiles, but even here there may be considerable cognitive variability as, for example, in Alzheimer's disease (AD), which selectively affects mesolimbic memory areas early in the course of the disease [e.g. 1]. Clinically, it has become customary to distinguish between cortical and subcortical dementia [30]. Cortical dementias (such as AD) are characterized by deficits in abstraction, orientation, judgment and memory functions [17, 32]. In addition, the affected patients may have incomplete lesions in those association areas subserving language, skilled movement and sensory interpretation. This may result in aphasia, apraxia or agnosia, which are usually of limited severity due to the diffuse and incomplete nature of the related cell loss. The prominent feature in subcortical dementia (such as Huntington's (HD) or Parkinson's (PD) diseases) is a gradual decline in cognitive abilities but without notable signs of cortical dysfunction [32, 36]. These kinds of distinctions could have been useful in identifying the affected regions in CJD, yet no reliable cognitive profiles exist for early CJD. This lack originates in the clinical nature of CJD: by contrast to the relatively slow progression of symptoms in the aforementioned types of dementia, the dementia in CJD progresses very rapidly, so symptoms and signs become manifest only weeks or months prior to loss of consciousness or death [1, 2]. Differential diagnosis at the early stages of the disease is also difficult and often gives rise to the diagnosis of AD or certain psychiatric disorders. Therefore, rapid onset and development of cognitive impairment are themselves diagnostic criteria for CJD, which leaves little time for a detailed cognitive investigation of the related patterns [9, 33, 35, 55].

Usually, the neuropsychological assessment of dementia is performed within a wide context of the clinical evaluation of patients, aimed at the diagnosis of neurological, psychiatric and physical dysfunction. In cases where neuropsychology made a significant contribution to dementia research, the clinical tests have been extended in a systematic and focused manner, so as to allow the theoretically motivated extension of the baseline clinical data. This allowed the achievement of better early diagnosis, often achieved with largely presymptomatic subjects, especially in AD and HD [16, 51]. In CJD, by contrast, cognitive assessment has not been sufficiently detailed so far, due to rapid evolution of the disease. Neuropsychological research in clinical CJD has also been hampered by the fact that it consisted, almost exclusively, of single case reports over a wide range of neuropathological conditions [e.g. 8, 20]. Consequently, misdiagnosis remained common and no coherent body of data has accumulated which could indicate the possible direction for extended research.

At the heart of the reported research is the neuropsychological evaluation of PRNP E200K mutation carriers. The study aimed to identify subtle subclinical abnormalities in healthy carriers in comparison to non-carriers, in order to understand the very early processes and pathogenesis of the disease. The participants in the study were all healthy first-degree relatives of Libyan Jewish patients, who died from the familial CJD form of the disease. Therefore, all participants had the same cultural background and lived in the same environmental conditions. Furthermore, they were familiar with the symptoms of CJD and the genetic implications of the disease. However, none of the participants were aware of their specific genetic status (carriers or non-carriers) prior to and during the study. This allowed us to conduct the study as a double-blind procedure (the researchers did not obtain the genetic status information during the administration and analysis of the neuropsychological tests). Since the E200K is a dominant mutation [35, 37] it was expected that around half of our participants would be carriers of this mutation. After obtaining the genetic data the participants were assigned into one of the two study groups: 'carriers' or 'controls'.

We gave the subjects a wide range of neuropsychological tests (including anxiety assessment). This battery of tests considered two possible outcomes: First, if no differences are found between healthy carriers and matched

Table 1. List of the cognitive tests that were used for evaluating the cognitive performance

Cognitive parameter	Neuropsychological tests	References
General evaluation of cognitive potential	Mini-Mental State Evaluation (MMSE) Wechsler Adult Intelligence Scale (WAIS-R)	Folstein et al., 1975 [15] Wechsler, 1981 [65]
Attention	Cancellation (tests A and B); Color Trail Making (tests A and B) Digit Span and Digit to Symbol (from WAIS-R)	Reitan, 1958 [50] Bornstein, 1985 [3]
Memory	Rey-Auditory Verbal Learning Test (AVLT) ; Visual Paired Association (from WMS-R) Delayed Recall of Rey-Autrich (FCT); Word and Sentence Repetition	Vakil and Blachstein, 1994 [62] Vakil and Blachstein, 1997 [63] Wechsler, 1987 [66] Spree and Strous, 1991 [57]
Visual perception	Copy Rey-Autrich (FCT); Picture Completion (from WAIS-R); Picture Recognition (usual and unusual views); Visual Paired Association (from WMS)	Spree and Strous, 1991 [57] Warrington and Taylor, 1973 [67]
Language	Object naming[1]; Word and Sentence Repetition; Verbal Fluency	Lezak, 1995 [41]
Executive functions	Wisconsin Card Sorting Test (WCST); Verbal Fluency (category and initial phoneme); Tower of Hanoi (time and number of trials); Similarities, Picture Arrangement and Block Design (from WAIS-R)	Heaton et al., 1981 [27] Hadar and Rose, 1990 [26]

[1] Object naming was assessed by asking subjects to name 12 real objects. This test is commonly used clinically to assess dementia (especially AD). Another assessment for object naming was done by using 20 photographed objects that were shown from usual views. The performance in this test was compared to object recognition, using the same 20 objects photographed from unusual views.

controls, this would mean that CJD has, indeed, a sudden, all-or-none onset, and that the clinical symptoms are, in fact, the earliest dysfunctions. Alternatively, carriers could differ, neuropsychologicaly, from controls. This would mean, of course, that CJD is preceded by presymptomatic dysfunctions, which may have differential rates of progression. Slow evolution of symptoms may also be indicated by large variability of performance profiles, which could also imply that a mild progressive cognitive decline appears in the carrier group and precedes the clinical symptoms and the disease onset by a fairly large margin. This study focused on the early symptoms of CJD in healthy subjects. The fact that the E200K familial form of the disease is very similar to its sporadic form [33] enhances the potential contribution of our study to the understanding of the preclinical processes that underlie the progress of the disease in the sporadic population as well.

Methods

Subjects and Testing Conditions

Twenty-seven healthy volunteers participated in the study. They were recruited through the national research roster for patients with CJD in Israel. All were first-degree relatives of genetically symptomatic patients who died from familial CJD and all were of Jewish Libyan decent. Inclusion criteria were language skills (fluent Hebrew), age (45–75), and the absence of neurological abnormalities or psychiatric illnesses. The institutional ethics committee approved the study, and each subject signed an informed consent to participate. Blood samples for genetic evaluation [6] were taken from all subjects in another session. The study was conducted as a double-blind procedure, neither the subjects nor the examiner nor any of the investigators were aware of the participants' genetic status (carrier/control). Since this study applied a double-blind procedure, some group parameters could not be precontrolled. This included group size, subjects' age and level of education. Thirteen of our volunteers (6 males and 7 females) were found, post-assessment, to be carriers of the E200K point mutation ('carriers') and the other 14 (7 males and 7 females) were not, constituting the control group ('controls').

Neuropsychological Tests and Procedure

All subjects were told that they had been selected to participate in a study of risk factors for the development of CJD due to their Libyan origin and being relatives of CJD patients. The test session lasted 4–5 h, and was conducted at a relaxed pace (coffee breaks, etc.). Each healthy participant completed a battery of cognitive tests that was based on various prevalent clinical findings [21, 22]. The test battery is summarized in table 1. Following cognitive assessment, each subject was evaluated with a Hebrew version of self-reported Trait and State Anxiety questionnaire [23].

Table 2. Subjects' demographic information (age and education) and general cognitive potential (MMSE and WAIS-R) (**a**, for the subject population generally; **b**, separating between young and elder subjects). There were no significant statistical differences between groups in any of the demographic parameters

a	Controls (14)	Carriers (13)	t-test	sig.
Age	59.4 ± 2.2	58.6 ± 2.0	0.33	0.74
Education	10.3 ± 1.2	9.9 ± 1.0	0.23	0.82
MMSE	27.3 ± 0.6	26.5 ± 0.8	0.85	0.41
WAIS-R	101.4 ± 3.2	98.8 ± 4.3	0.47	0.64

b	Young (11)				Elders (16)			
	controls (5)	carriers (6)	t-test	sig.	controls (9)	carriers (7)	t-test	sig.
Age	50.0 ± 1.8	52.3 ± 1.8	0.79	0.17	64.7 ± 1.2	64.0 ± 1.5	0.39	0.70
Education	13.6 ± 1.5	11.2 ± 0.8	1.46	0.45	8.4 ± 1.4	8.9 ± 1.7	1.87	0.85
MMSE	28.4 ± 0.6	27.7 ± 0.3	1.12	0.29	26.7 ± 0.75	25.4 ± 1.4	0.83	0.42
WAIS-R	103.6 ± 6.7	104.2 ± 4.5	0.72	0.99	100.1 ± 3.6	94.3 ± 6.8	0.81	0.43

Statistical Analysis

Data was analyzed using the SPSS for Windows statistical software (version 9). Mann-Whitney non-parametric t-test (for independent samples), MANOVA and repeated measures tests were used to assess differences between group parameters. All means are presented with 1 standard error of mean (mean ± SEM).

Results

Statistical analyses showed that the two healthy groups (carriers and controls) were similar in demographic mean parameters (age and education), and showed no significant differences in mean MMSE score (see table 2a). Splitting the data according to age groups did not achieve significant differences either (table 2b). By contrast, our results revealed three main presymptomatic neuropsychological differences between healthy CJD mutation carriers and controls: (1) Anxiety level among the mutation carrier group was significantly higher compared to the control group that was recruited from the same families. This difference increased with age (fig. 1). (2) The older healthy carrier group (age ≥ 60 – referred to as 'older carrier' henceforth) showed significant deficits in some cognitive tasks. These included object recognition, object naming, and the copy stage of the CFT test (table 3). In addition, the mean performance of verbal learning, verbal memory and verbal IQ were markedly lower in the 'older carrier' in comparison to old controls, although only the slope of the verbal learning task (AVLT; trials 1–5) reached significance. Analysis of the 'older carriers' data revealed 3–4 healthy carriers who seemed to contribute most to the significance of the cognitive differences. These 4 subjects were, also, the only ones to show an object-naming deficit. Moreover, all of them showed abnormal verbal WAIS-R IQ score (<85, which is 1 SD under the normal population mean) (table 4). Using MANOVA (or repeated measures for repeated trials) to compare these 4 healthy elder carriers with elder controls and the remaining elder carriers revealed that they were signifi-

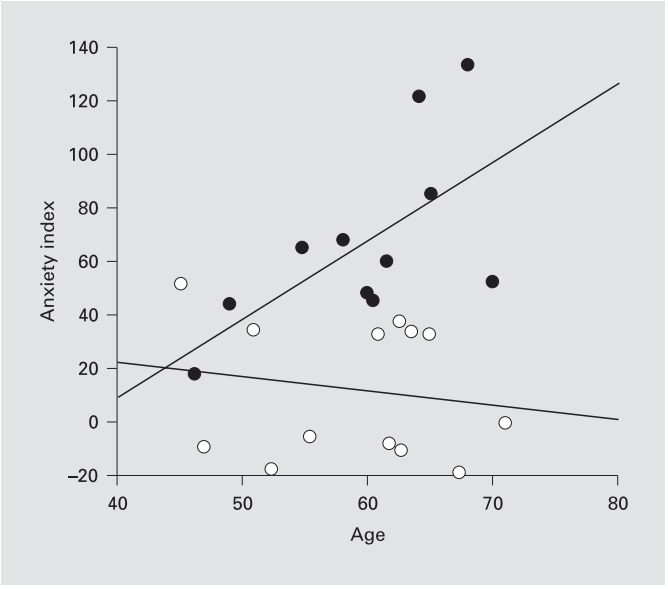

Fig. 1. Correlation between anxiety index (see Results) and age in carriers (●, r = 0.64; p = 0.03; Y = –111.27 + 2.99X) and controls (○, r = 0.16; n.s. Y = 42.46 – 0.53X).

Table 3. Mean (± SEM) cognitive performance of the various groups

Cognitive test	Young (11)		Elders (16)		Sig.
	controls (5)	carriers (6)	controls (9)	carriers (7)	
Attention					
Cancellation A, s	49.0 ± 5.8	46.0 ± 2.4	62.0 ± 6.6	71.4 ± 9.7	n.s
Cancellation B, s	60.2 ± 7.2	49.2 ± 3.1	60.2 ± 4.6	73.8 ± 8.3	n.s
Trail A, s	58.8 ± 9.6	53.0 ± 6.4	81.5 ± 13.5	91.0 ± 13.5	n.s
Trail B, s	162.6 ± 32.3	93.7 ± 5.3	187.2 ± 43.0	204.9 ± 45.9	0.03*
Digit to symbol	9.4 ± 1.5	9.2 ± 1.0	8.0 ± 0.6	7.1 ± 1.2	n.s
Digit span	8.0 ± 1.1	8.3 ± 0.8	8.1 ± 0.7	7.9 ± 1.3	n.s
Language and verbal memory					
Verbal IQ (WAIS-R)	104.4 ± 6.3	102.8 ± 4.3	98.7 ± 3.5	91.6 ± 7.2	n.s
Object naming (out of 12)	11.8 ± 0.2	12.0 ± 0.0	11.6 ± 0.2	10.0 ± 0.5	0.02*
Sentence repetition (max. = 38)	28.7 ± 1.8	31.4 ± 0.2	28.6 ± 1.0	21.3 ± 3.4	n.s
AVLT[1]					
Maximum words (out of 15)	11.4 ± 1.4	12.0 ± 1.0	10.6 ± 0.6	7.4 ± 1.3	n.s
Slope of learning [*(trial 5 – trial 1)/5*]	1.3 ± 0.1	1.4 ± 0.2	1.2 ± 0.06	0.6 ± 0.2	0.01*
Delayed recall	8.2 ± 1.8	10.2 ± 1.2	8.0 ± 0.8	6.5 ± 1.1	n.s
Visual perception and visual memory					
Recognition of usual views (max. = 20)	18.4 ± 0.4	19.3 ± 0.5	18.1 ± 0.5	15.4 ± 1.2	0.05*
Recognition of unusual views (max. = 20)	13.4 ± 2.3	16.0 ± 1.2	11.8 ± 0.9	8.3 ± 1.5	0.05*
CFT (copy)	34.3 ± 1.3	34.5 ± 0.7	27.6 ± 3.9	24.1 ± 3.7	0.02*
CFT (delayed recall)	16.3 ± 3.2	19.9 ± 4.4	11.4 ± 2.6	8.0 ± 1.6	n.s
Visual paired association (delayed recall)	4.0 ± 1.0	5.0 ± 1.0	2.3 ± 0.6	3.7 ± 0.8	n.s
Picture completion (from WAIS-R)	7.6 ± 1.3	9.2 ± 1.0	7.2 ± 0.9	6.8 ± 0.9	n.s
Executive functions					
Verbal fluency					
Category	52.4 ± 4.3	51.0 ± 4.2	43.2 ± 2.8	41.4 ± 1.9	n.s
Phonological	32.2 ± 1.7	37.2 ± 4.5	20.9 ± 3.8	20.6 ± 6.6	n.s
Similarities (from WAIS-R)	11.6 ± 1.3	11.0 ± 0.7	9.8 ± 0.7	9.3 ± 1.6	n.s
Block design	7.8 ± 1.2	8.0 ± 1.1	6.1 ± 1.4	4.9 ± 1.8	n.s
Tower of Hanoi	44.0 ± 5.9	41.2 ± 6.7	58.8 ± 21.8	54.7 ± 12.2	n.s
WSCT					
Categories	3.6 ± 1.0	5.4 ± 0.6	4.5 ± 0.6	4.1 ± 0.6	n.s
% perseverative errors	18.2 ± 4.0	12.4 ± 3.5	13.6 ± 2.3	22.6 ± 3.8	n.s
% nonperseverative errors	19.9 ± 7.2	10.2 ± 2.0	20.1 ± 4.0	16.1 ± 2.4	n.s

***** Significance is related to the Mann-Whitney t-test for non-parametric independent samples, comparing between the two elder groups (carriers and controls).

[1] The Auditory Verbal Learning Test (AVLT) included 15 words that were auditory presented at 5 times (each named a trial), and subjects were asked (at each trial) to recall as many words as they could remember. The learning curve was calculated as increment between trials 5 and 1 divided by number of recalled words in trial 5.

cantly lower in all cognitive parameters (post-hoc Scheffé, $p < 0.05$). These subjects were therefore considered a separate group and referred to as 'clinical presymptomatic'. (3) Unexpectedly, and despite their lower mean education compared to young controls, the young carrier group showed markedly better performance in almost all cognitive tasks. However, in only one task (Trail B time performance), the difference reached statistical significance.

Discussion

This is the first research that focuses on preclinical neuropsychological signs in healthy subjects that are at a genetic risk for CJD. Recent studies demonstrated neuropsychological presymptoms in other kinds of dementia such as AD and HD [1, 10, 14, 16, 18, 19, 25, 51, 56, 61] but, thus far, no research has evaluated either subjects at

Table 4. Mean (±SEM) WAIS-R IQ scores

	Young (11)		Elders (16)		
	controls	carriers	controls	carriers	4 pre-symptomatics[1]
Performance IQ					
Mean ± SEM	101.8 ± 6.6	105.7 ± 4.5	106.5 ± 6.0	109.7 ± 8.6	89.284.7
Minimun	87	96	86	97	76
Maximun	122	125	141	126	98
Verbal IQ					
Mean ± SEM	104.4 ± 6.3	102.8 ± 4.3	100.2 ± 3.6	108.7 ± 8.8	78.783.7
Minimun	92	88	88	92	68
Maximun	123	114	119	122	85

Four 'pre-symptomatic' elder carriers that were defined clinically as healthy, showed low IQ score (verbal and performance).

[1] One-way ANOVA and Scheffé's post-hoc showed significance of $p < 0.05$ each (see text).

the very early stages of CJD or healthy individuals at risk for CJD. Our accessibility to this genetic population, at risk for developing the disease, offered a unique opportunity to investigate the question of whether the genetic mutation has an effect on preclinical cognitive function. Resolving this question could illuminate other aspects of this enigmatic disease, and may enable therapeutic treatment at diminished brain degenerated stages. All of our subjects (carriers and controls) were from the same lineage (Libyan Jewish), first-degree relatives of patients who were carrying the specific mutation (E200K) and who died from CJD. Therefore, our experimental population was homogeneous and the controls were tightly matched. In addition, the study was carried out in a double-blind manner.

Our main finding relates to anxiety levels. These were shown to be nearly normal until about 50 years of age, with a rapid increase thereafter (fig. 1). It is well known that stressful life events can be risk factors for expressing psychiatric (schizophrenia and major depression) and neurological (epilepsy, Alzheimer's, Parkinson's) diseases. Certain psychopathologic data point to the possibility that several psychiatric and neurological diseases are based on inherited brain defects which, together with stressful events, lead to the expression of the disease [39]. Likewise, there are some indications that the expression of the CJD mutation can be accelerated by psychological or physiological stress. Indeed, it has been shown that the expression in transgenic mice with abnormal human PRNP requires additional cellular factors for the conversion process [60], and it has been suggested that these factors might be activated by biological stress factors [59]. Interestingly, 49% of Branded's [4] CJD patients (27 of 55) had experienced emotional disturbances between 1 and 5 years before the onset of the disease, due to a stressful life event.

Perceiving an event as stressful depends not only on the nature of the event but also on individual propensity. The pathological defending expression for a stressful event may be depression or anxiety (as well as other responses). In our study we show no difference in depression scores between the carriers and the controls, but a very significant state anxiety difference. Therefore, we suggest that the PRNP E200K mutation carriers are chronically anxious, and perceive daily situations as stressful. The accumulation of stressful situations may enhance the conversion of the mutant protein to the abnormal protein isoform later in life. The fact that the elder carrier subjects showed higher abnormality supports this suggestion.

In humans, cortical areas have an important place in the perception of an event as stressful and in the control of cognitive influences on brain structures involved in emotion (e.g., limbic system). Evidence from brain imaging implicates specific brain structures such as the amygdaloid complex and septal area, as well as the cingulate, orbitofrontal and prefrontal cortices as mediators of the broad range of behavioral and physiological responses associated with anxiety [54]. Other studies suggest that the neuropathology of anxiety involves specific neuroreceptors, such as catecholaminergic and benzodiazepines. Anxiety states are also characterized by high arousal and

vigilance and are associated with hyperactivity of the hypothalamic-pituitary-adrenal (HPA) axis, which is further enhanced by morphologic and functional linkage between the corticotropin-releasing hormone and the sympathetic system [47]. However, it is still controversial whether perceiving the same external event as differently stressful accounts for the bulk of individual differences in the HPA axis activity or, alternatively, the HPA axis hyperactivity is secondary to the cognitive perception of an event as stressful [53]. In any of these possibilities, our results raise the question of whether anxiety is a preclinical sign resulting from the accumulation of very little amounts of the abnormal PRNP, or if it is present prior to the PrP^{Sc} conversion to the abnormal isoform. The latter possibility may have two explanations. Firstly, these familial carriers may have an inherited brain defect in pathways that mediate anxiety or stress responses (e.g., in the HPA axis or cortical areas involved in the perception of emotions, as well as limbic structures). Secondly, abnormal anxiety could be a secondary manifestation of the deficit in PrP^{c}. Recent biochemical studies showed a correlation between the cellular amounts of PrP^{c} and the resistance of brain cells to chemical stress [5]. At the same time, other studies suggest that the amount of heat-shock proteins is related to the phenotypic expression of PrP^{Sc} in transgenic mice [4, 59]. Even though the massive conversion to the abnormal isoform may start only later in life, carriers may exhibit less functional PrP^{c} and less resistance to metabolic stress. Defective cellular resistance to chemical injury may lead to the intracellular accumulation of biological stress factors [4] and thus to neuropharmacological changes and enhanced anxiety.

Two other findings are both remarkable and unexpected: one is the better cognitive performance of the young carriers compared to young controls, and the other is the cognitive differences between elder and young carriers, which were often greater than the disparity between carriers and controls. These two findings may be related to the main reported abnormality among our healthy at-risk subjects, who demonstrated elevated levels of anxiety and stress [23], accompanied by decreased levels of cognitive performance. Two possible explanations may apply here. At-risk subjects may respond with anxiety to the experience of subtle cognitive deterioration (although they do not report it). A similar explanation has recently been offered for clinically healthy subjects at risk for HD [10, 14]. The first suggestion, therefore, is that in CJD, the cognitive deficits are the primary consequences of the genetic mutation, while stress and anxiety are secondary responses. This idea, however, does not explain the superior cognitive performance of the young at-risk subjects. Alternatively, cognitive dysfunction may be the consequence of abnormal anxiety and chronic hyperarousal. Anxiety and arousal are modulated by steroid and adrenaline concentration through the HPA, usually following the perception of an event as a stressful one [13, 34, 46]. Slightly elevated arousal is well known to improve cognitive performance [13, 44]. However, prolonged high levels of these substances may hasten cell death and ultimately impair cognitive function [53]. This suggestion implies that abnormal stress levels are the primary impairments of the PRNP genetic mutation. These may involve abnormal function of either lower cerebral structures (HPA axis) or higher structures that modulate the perception of an event as stressful (primarily the amygdaloid complex, the orbitofrontal pathways and the cingulate cortex) [e.g. 2]. In this assumption, the cognitive impairments are subsequent to high anxiety levels. The superior cognitive performance of the young at-risk subjects may originate in the initial low levels of increased anxiety and arousal, while further increased anxiety level, both in magnitude and duration, eventually causes cognitive deterioration in the elder at-risk subjects. This explanation is partly supported by recent retrospective studies, which reported excessive proportion of stressful life events in patients who died from CJD [4, 40, 64]. Excessive stress in these patients may have accelerated the neuropathological process.

Irrespective of the above possible explanations, our results clearly show that mild, yet specific, cognitive deficits (and possibly a gradual decline) may precede the clinical onset of CJD in the at-risk population. The most noticeable cognitive deficits in our at-risk subjects were in the tasks of object naming and recognition. The very fact of presymptomatic deficits is not surprising since presymptomatic cognitive deficits were shown in other dementias, e.g., HD and AD [1, 10, 14, 18, 25, 56, 61]. Our results, however, are theoretically problematic, for if the observed deficits originate in extended arousal levels, which is the more likely explanation, then they should be diffuse and span a wide range of cognitive functions and perhaps even be of a psychiatric nature [41]. Yet, this is not the pattern of our results, where object semantics seems the prime problem (originating either in impaired visual recognition or in impaired lexical semantics). Thus, in the elder at-risk subjects, object naming was lower by about 15% compared to matched controls. Object recognition, assessed by performance from unusual views, was lower by 30% (table 3). The more severe problem from unusual views could tilt the balance in favor of a recogni-

tion problem, rather than a lexical retrieval problem. However, the difference between 15 and 30% deficit is not so remarkable and could be explained simply by the greater difficulty, even for normal subjects, to recognize objects from unusual views [12, 38, 42, 48, 58].

In addition, there is considerable evidence that associates aging and dementia with naming problems [11, 38, 45]. This is more commonly observed in AD patients, but has also been reported in HD patients [61]. Here, again, controversy about processing origins is nourished by the ambiguity of the available data. Thus, the performance deficits of HD patients were ascribed to impaired recognition, while the performance profiles of AD patients were ascribed to conceptual, or even lexical, semantic deficits, but even that was not left uncontested [e.g. 61]. On the present data, CJD profiles could indicate either visual recognition or linguistic impairments and more evidence is required to decide between the two explanations. We incline to favor the hypothesis of a recognition problem as underlying the deficits seen in our carrier population. Firstly, this population showed more clearly visual perception problems in performing the Rey-Complex Figure Test (table 3). Here they were similar to those of the preclinical HD carriers, for whom a visual recognition problem seemed very plausible. Indeed, visual problems are one of the common clinical CJD characteristics [33, 35]. Also, the Trail B task requires adequate visuospatial abilities [41], whose impairment may underlie the poor performance of the elder carriers. However, some of our evidence may be interpreted as indicating a linguistic deficit. Four healthy elder carriers displayed naming deficits in the real objects naming task. These subjects also displayed abnormal verbal IQ scores, reaching lower than 1 SD below the mean population score (<85, n.s.; table 4). These 4 carriers were responsible for the decreased ability to name objects pictured from usual views, indicating a linguistic deficits, but they were also the ones who contributed most to the low performance on objects pictured from unusual views. These 4 subjects could well be considered as 'presymptomatic'.

Considering the entire data, we may conclude that the progress of CJD in the carrier population involves three stages that precede the clinical onset. The initial stage involves alterations in stress or anxiety levels. We propose these to be the primary causes of preclinical deterioration in carriers and suggest that they indicate the existence of subcortical changes [23] prior to the cortical (cognitive) ones. The subcortical irregularity can be a direct consequence of the PRNP gene mutation, while the cortical irregularity may be a consequence of the subcortical dysfunction, by analogy to mechanisms suggested for other disorders (such as PD or HD [18]).

In between the stages of subcortical stress elevation and cortical deficits there may be a stage of first enhanced and then impaired attention and concentration. This may be indicated by the comparative overall performance profiles of, respectively, the younger and the elder carrier groups. The declining performance of the elder population on the memory tasks of the AVLT (table 3) may reflect the remains of this attentional deficit. However, if we place the subcortical damage at the amygdaloid complex, we may also explain the impaired higher visual functions as a direct consequence of the lower impairment. It is now well proven that amygdaloid activity directly modulates neuronal activity in higher visual cortex [29, 42, 52].

To summarize, we demonstrated in this study, for the first time, that preclinical deterioration precedes the rapid progressive dementia seen in clinical CJD. While the evidence that could lead to clear theoretical conclusions is still missing, our results were specific enough to venture into plausible speculations about the origins and course of this deterioration. This offers a challenging scope for both future research and, possibly, future therapeutic interventions. The data summarized here may offer new leads for the study of CJD, suggesting, among other things, that stress reduction may delay the onset of CJD in the carrier population. Studies using dynamic neuroimaging may help resolve the above issues by specifying the implicated brain regions, as well as the related neuropharmacology.

Acknowledgements

The study reported here was supported by a grant from the Israel Academy of Sciences and Humanities. We thank Prof. Amos Korczyn for allowing us to use his laboratory and Dr. Mira Birenbaum for executing some of the DNA tests.

References

1 Albert MS: Cognitive and neurobiologic markers of early Alzheimer disease. Proc Natl Acad Sci USA 1996;93:13547–13551.
2 Bernard JF, Bandler R: Parallel circuits for emotional coping behavior: New pieces in the puzzle. J Comp Neurol 1998;401:429–436.
3 Bornstein RA: Normative data on selected neuropsychological measures from a nonclinical sample. J Clin Psychol 1985;41:651–659.
4 Brandel JP, Delasnerie-Laupretre N: Creutzfeldt-Jakob disease and stress. J Neurol Neurosurg Psychiatry 1997;62:541.
5 Brown DR, Besinger A: Prion protein expression and superoxide dismutase activity. J Biochem 1998;334:423–429.
6 Chapman J, Ben-Israel J, Goldhammer Y, Korczyn AD: The risk of developing Creutzfeldt-Jakob disease in subjects with the PRNP gene codon 200 point mutation. Neurology 1994; 44:1683–1686.
7 Chapman J, Korczyn AD: Genetic and environmental factors determining the development of Creutzfeldt-Jakob disease in Libyan Jews. Neuroepidemiology 1991;10:228–231.
8 Cochran EJ, Bennett DA, Cervenakova L, Kenney K, Bernard B, Foster NL, Benson DF, Goldfarb LG, Brown P: Familial Creutzfeldt-Jakob disease with a five-repeat octapeptide insert mutation. Neurology 1996;47:727–733.
9 Collinge J: Creutzfeldt-Jakob disease and other prion diseases; in Obrien J, Ames D, Burns A (eds): Dementia. London, Oxford University Press, 2000, pp 863–875.
10 Craufurd D, Thompson JC, Snowden JS: Behavioral changes in Huntington disease. Neuropsychiatry Neuropsychol Behav Biol 2001; 14:219–226.
11 Cubelli R, Speri V: Naming rotated pictures and the riddle of object-centred neglect. Cortex 2001;37:159–174.
12 Damian MF, Martin RC: Semantic and phonological codes interact in single word production. J Exp Psychol Learn Mem Cogn 1999;25: 345–361.
13 De Kloet ER, Oitzl MS, Joels M: Stress and cognition: Are corticosteroids good or bad guys? Trends Neurosci 1999;22:422–426.
14 Diamond R, White RF, Myers RH, Mastromauro C, Koroshetz WJ, Butters N, Rothstein DM, Moss MB, Vasterling J: Evidence of presymptomatic cognitive decline in Huntington's disease. J Clin Exp Neuropsychol 1992; 14:961–975.
15 Folstein MF, Folstein SE, McHugh PR: 'Mini-mental state'. A practical method for grading the cognitive state of patients for the clinician. J Psychiatr Res 1975;12:189–198.
16 Foroud T, Siemers E, Kleindorfer D, Bill DJ, Hodes ME, Norton JA, Conneally PM, Christian JC: Cognitive scores in carriers of Huntington's disease gene compared to noncarriers. Ann Neurol 1995;37:657–664.
17 Forstl H: What is Alzheimer's disease? In Obrien J, Ames D, Burns A (eds): Dementia. London, Oxford University Press, 2000, pp 371–383.
18 Fox NC, Warrington AL, Seiffer AL, Agnew SK, Rosser MN: Presymptomatic cognitive deficits in individuals at risk of familial Alzheimer's disease. Brain 1998;121:1631–1639.
19 Frank EM, McDade HL, Scott WK: Naming in dementia secondary to Parkinson's, Huntington's, and Alzheimer's diseases. J Commun Disord 1996;29:183–197.
20 Gass CS: Familial CJD: A neuropsychological case report. Arch Clin Neuropsychol 2000;15: 165–175.
21 Gigi A, Kahana E, Vakil E, Hadar U, Prohovnik I: Neuropsychological assessment in early Creutzfeldt-Jakob disease. Soc Neurosci Abstr 1999;25:739.12.
22 Gigi A, Kahana E, Vakil E, Hadar U, Prohovnik I: Neuropsychological assessment in early Creutzfeldt-Jakob disease. Neurosci Lett 1998(suppl 51):S13.
23 Gigi A, Kahana E, Hadar U: Healthy Creutzfeldt-Jakob disease mutation carriers have abnormality in anxiety reported scores (STAS). Society for Neuroscience Abstracts, 2001.
24 Goldfarb LG, Korczyn AD, Brown P, Chapman J, Gajdusek DC: Mutation in codon 200 of scrapie amyloid precursor gene linked to Creutzfeldt-Jakob disease in Sephardic Jews of Libyan and non-Libyan origin. Lancet 1990; 336:637–638.
25 Gomez-Tortosa E, del Barrio A, Sanchez PR, Benitez J, Barroso A, Garcia YJ: Cognitive assessment of asymptomatic Huntington's disease carrier. Neurologia 1997;12:226–231.
26 Hadar U, Rose FC: Neuropsychological assessment of cognitive change in dementia. Neuroepidemiology 1990;9:189–192.
27 Heaton RK, Chelune GJ, Talley JL, Kay GG, Curtiss G: Wisconsin Card Sorting Test Manual (revised and expanded). Lutz/Fla, Psychological Assessment Resources, Inc, 1993.
28 Heilman KM, Valentin E: Clinical Neuropsychology, ed 3. London, Oxford University Press, 1993.
29 Hendler T, Rotstein P, Hadar U: Emotion-perception interplay in the visual cortex: 'The eyes follow the heart'. Cell Mol Neurobiol 2001;21: 733–752.
30 Huber SJ, Shuttleworth EC, Pauslson GW, Bellchambers MJ, Clapp LE: Cortical and subcortical dementia: Neuropsychological differences. Arch Neurol 1986;43:392–394.
31 Ironside JW: Prion diseases: Update on Creutzfeldt-Jakob disease. Neuropathol Appl Neurobiol 1996;22:446.
32 Jacobs DM: The role of neuropsychological testing in neurological disease. Neurol Update 1998;3:139–143.
33 Kahana E, Zilber N, Abraham M: Do Creutzfeldt-Jakob disease patients of Jewish Libyan origin have unique clinical features? Neurology 1991;41:1390–1392.
34 Kasckow JW, Baker D, Geracioti TD: Corticotrophin-releasing hormone in depression and post-traumatic stress disorder. Peptides 2001; 22:845–851.
35 Knight R: Creutzfeldt-Jakob disease: Clinical features, epidemiology and tests. Electrophoresis. 1998;19:1306–1310.
36 Knopman DS: The initial diagnosis and recognition of dementia. Am J Med 1998;104:S2–S12.
37 Kovanen J: Clinical characteristics of familial and sporadic Creutzfeldt-Jakob disease in Finland. Acta Neurol Scand 1993;87:469–474.
38 Laiaona M, Luzzatti C, Zonca G, Guarnaschelli C, Capitani E: Lexical and semantic factors influencing picture naming in aphasia. Brain Cogn 2001;46:184–187.
39 Lalonde FM, Martin AM, Myslobodsky MS: Increased prevalence of septal cavitation in a nonschizophrenic sample: An MRI study of HIV-infected individuals. J Neuropsychiatry Clin Neurosci 1995;8:47–53.
40 Laske C, Gefeller O, Pfahlberg A, Zerr I, Schroter A, Poser S: The effect of stress on the onset and progression of Creutzfeldt-Jakob disease: Results of a German pilot case-control study. Eur J Epidemiol 1999;15:631–635.
41 Lezak MD: Neuropsychological Assessment, ed 3. London, Oxford University Press, 1995.
42 Levelt WJ, Roelofs A, Meyer AS: A theory of lexical access in speech production. Behav Brain Sci 1999;22:1–38.
43 Levin BE, Tomer R, Rey GJ: Cognitive impairments in Parkinson's disease. Neurol Clin 1992;10:471–485.
44 McEwen BS: Neuroendocrine interactions; in Bloom FE, Kupfer DJ (eds): Psychopharmacology. New York, Raven Press, 1995, chapt 62, p 705.
45 Margorie N, Loraine KO, Rhoda A, Martin LA: On the nature of naming errors in aging and dementia. Brain Lang 1996;54:184–195.
46 Mathew SJ, Coplan JD, Gorman JM: Neurobiological mechanisms of social anxiety disorder. Am J Psychiatry 2001;158:1558–1567.
47 Merola B, Longobardi S, Colao A, Di Somma C, Ferone D, Rossi E, Covelli V, Lombardi G: Hypothalamic-pituitary-adrenal axis in neuropsychiatric disorders. Ann NY Acad Sci 1994; 741:263–270.
48 Nicholson KG, Humphrey GK: Surface cues reduce the latency to name rotated images of objects. Perception 2001;30:1057–1081.
49 Prusiner SB: The prion diseases. Brain Pathol 1998;8:499–513.
50 Reitan RM: Validity of the Trail-Making test as an indicator of organic brain damage. Percept Memory Skills 1958;8:271–276.
51 Rosenberg NK, Sorensen SA, Christensen AL: Neuropsychological characteristics of Huntington's disease carriers: A double-blind study. J Med Genet 1995;32:600–604.
52 Rotstein P, Malach R, Hadar U, Graif M, Hendler T: Feeling or features: Different sensitivity to emotion in high-order visual cortex and amygdala. Neuron 2001;32:747–757.

53 Sapolsky RM: Stress, the Aging Brain, and the Mechanisms of Neuron Death. London, MIT Press, 1992.
54 Scott LR, Cary RS, Nathaniel MA, Alan JF, Michael AJ: The functional neuroanatomy of anxiety: A study of three disorders using positron emission tomography and symptom provocation. Biol Psychiatry 1997;42:446–452.
55 Simon ES, Kahana E, Chapman J, Treves TA, Gabizon R, Rosenmann H, Zilber N, Korczyn AD: Creutzfeldt-Jakob disease profile in patients homozygous for the PRNP E200K mutation. Ann Neurol 2000;47:257–260.
56 Small GW, Ercoli LM, Silverman DH, Hung SC, Komo S, et al.: Cerebral metabolic and cognitive decline in persons at risk for Alzheimer's disease. Proc Natl Acad Sci USA 2000;97:6037–6042.
57 Spree O, Strous EA: Compendium of Neuropsychological Tests. New York, Oxford University Press, 1991.
58 Stuss DT, Sarazin FF, Leech EE, Picton TW: Event-related potentials during naming and mental rotation. Electroencephalogr Clin Neurophysiol 1983;56:133–146.
59 Tatzelt J, Voellmy R, Welch WJ: Abnormalities in stress proteins in prion diseases. Cell Mol Neurobiol 1998;18:721–729.
60 Telling GC, Scott M, Hsiao KK, Foster D, Yang SL, Torchia M, Sidle KCL, Collinge J, DeArmond SJ, Prusiner SB: Transmission of Creutzfeldt-Jakob disease from humans to transgenic mice expressing chimeric human-mouse prion protein. Proc Natl Acad Sci USA 1994;91:9936–9940.
61 Tierney MC: Cognitive tests that best discriminate between presymptomatic AD and those who remain nondemented. Neurology 2001;57:163–164.
62 Vakil E, Blachstein H: A supplementary measure in the Rey AVLT for assessing incidental learning of temporal order. J Clin Psychol 1994;50:240–245.
63 Vakil E, Blachstein H: Rey AVLT: Developmental norms for adults and the sensitivity of different memory measures to age. Clin Neuropsychol 1997;11:1–14.
64 Van Duijn CM, Delasnerie-Laupretre N, Masullo C, et al: Case-control study of risk factors of Creutzfeldt-Jakob disease in Europe during 1993–1995. Lancet 1998;351:1081–1085.
65 Wechsler D: WAIS-R Manual. New York, Psychological Corp, 1981.
66 Wechsler D: WMS-R Manual. New York, Psychological Corp, 1987.
67 Warrington EK, Taylor AM: The contribution of the right parietal lobe to object recognition. Cortex 1973;9:152–164.