Six Generations of *CHMP2B*-mediated Frontotemporal Dementia: Clinical Features, Predictive Testing, Progression and Survival

Running Title: Six Generations of CHMP2B

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ABSTRACT

Objectives

Chromosome 3-linked Frontotemporal Dementia (FTD-3) is caused by a c.532-1G>C mutation in the *CHMP2B* gene. It is extensively studied in a Danish family comprising one of the largest families with an autosomal dominantly inherited frontotemporal dementia (FTD).

This retrospective cohort study utilizes demographics to identify risk factors for onset, progression, life expectancy and death in *CHMP2B*-mediated FTD. The pedigree of 528 individuals in six generations is provided, and clinical descriptions are presented. Choices of genetic testing are evaluated.

Materials and Methods

Demographic and lifestyle factors were assessed in survival analysis in all identified *CHMP2B* mutation carriers (44 clinically affected FTD-3 patients and 16 presymptomatic *CHMP2B* mutation carriers). Predictors of onset and progression included sex, parental disease course, education and vascular risk factors. Life expectancy was established by matching *CHMP2B* mutation carriers with average life expectancies in Denmark.

Results

Disease course was not correlated to parental disease course and seemed unmodified by lifestyle factors. Diagnosis was recognized at an earlier age in members with higher levels of education, probably reflecting an early dysexecutive syndrome, unmasked earlier in people with higher work-related requirements.

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Carriers of the *CHMP2B* mutation had a significant reduction in life expectancy of 13 years. Predictive genetic testing was chosen by 20% of at-risk family members.

Conclusions

CHMP2B-mediated FTD is substantiated as an autosomal dominantly inherited disease of complete penetrance. The clinical phenotype is a behavioural variant FTD. The disease course is unpredictable, and life expectancy is reduced. The findings may be applicable to other genetic FTD subtypes.

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Key words

Frontotemporal Dementia

CHMP2B

Hereditary degenerative disorders

INTRODUCTION

The Danish FTD-3 family was initially described in 1987 in the family of a patient admitted to a psychiatric department in a rural area of Northern Jutland in Denmark [1]. With aunts and uncles previously admitted to the same department, the pattern of an autosomal dominant neurodegenerative disease appeared, and the disease was soon traced to the grandmother, a woman born in 1876 and the mother of twelve children. She developed personality changes at the age of 56 and died at the local psychiatric institution at the age of 68. Nine of her twelve children developed similar behavioural changes and cognitive decline in their sixth decade of life, while neither her parents nor siblings had presented symptoms of dementia. Her descendants now comprise one of the largest known kindreds of an autosomal dominant neurodegenerative disease. Clinical and pathological features in affected family members were first reported in 1987 and updated in 1995, 1999, 2002 and 2008 [1–5]. Disease onset is usually in the sixth decade of life, although longitudinal assessments have demonstrated a decline in executive functions years prior to behavioural symptoms [6]. The typical course of disease starts with subtle personality changes with disinhibited behaviour, apathy and lack of insight. With progression, a global dementia develops, including language impairment. Later, dystonia, myoclonus and parkinsonism develop followed by immobilization, bed confinement and death [4].

Through the extensive collaboration between researchers and family members, genetic linkage analyses located the disease gene to chromosome 3, coining the term chromosome 3-linked Frontotemporal Dementia or FTD-3 [2,3]. In 2005, the cause of disease was identified as a single-base mutation (c.532-1G>C) in the *CHMP2B* gene causing C-terminal truncation of the CHMP2B protein [7,8]. Identification of the disease-causing mutation enabled predictive genetic testing of at-risk family

members. The neuropathology of FTD-3 was characterized as FTLD-UPS [9–11] which is unique to this family.

The Danish FTD-3 family now comprises 528 individuals across six generations with 51 clinically affected by FTD and fewer presymptomatic mutation carriers identified. The phenotype and disease course are highly heterogeneous.

The aim of this study was to provide the first systematic description of the family since 2002 [4]. Further, by including all known *CHMP2B* mutation carriers in survival analysis, we aimed to identify predictors and modifiers of disease course in demographics, presymptomatic lifestyle and pharmaceutical and medical history. We hypothesized that clinical onset of FTD-3 could be predicted by demographic factors and parental disease course, while progression and death in FTD-3 could be predicted by onset and possibly modified by lifestyle factors.

MATERIALS AND METHODS

Study population

The FTD-3 family has been followed extensively for almost three decades by the **F**rontotemporal dementia **Re**search in **J**utland **A**ssociation (FReJA) consortium, and the continuing contact between researchers and family members has provided a comprehensive family history recording including clinical and genetic characterization of family members. In a research setting at the Danish Dementia Research Centre, Rigshospitalet, Denmark all family members are offered genetic counselling and possible predictive testing as well as diagnostic work-up and follow-up. The pedigree is well established, although some branches have been lost to follow-up due to lack of affected family members, and it appears likely that the mutation is not present in these branches.

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In this retrospective cohort study, all 51 family members affected by FTD-3 were described in terms of clinical features, Age at Onset (AAO), Age at Institutionalization (AAI) and Age at Death (AAD), and these endpoints of disease course were applied in survival analysis. AAO was defined as the time of first symptoms noticed by relatives. In this pedigree spanning more than a century, reports on disease course could vary between distant relatives in contact with the research group and older medical records with disease course provided by the closest relatives. In these cases, the reports on disease course provided in medical records were chosen and considered more credible than reports provided retrospectively, sometimes decades later. As institutionalization in Denmark is in some degree based on specified criteria, AAI was considered a more uniform and reliable measure of disease severity and progression. AAD was registered for all deceased cases.

We included information about lifestyle in terms of smoking, physical exercise, alcohol consumption and education, applying the International Standard Classification of Education (ISCED). When relevant, medical history and pharmacological treatments were described.

The presence of the *CHMP2B* mutation was established in 44 cases of disease by either direct genetic testing or by the presence of the mutation in descendants of the affected deceased family member. Further, 16 presymptomatic *CHMP2B* mutation carriers were identified and included in the analyses.

Presence of the pathogenic *CHMP2B* mutation was identified as previously described [5]. In seven deceased cases, no biosamples or autopsies were available, and no descendants tested positive for the mutation. Although similar in clinical symptoms, these seven cases were excluded from the subsequent analysis.

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Ethics Statement

Data was provided by family members. Subjects alive after 1985 gave informed consent to the inclusion of data in large studies, and data on subjects deceased prior to 1985 was provided by close relatives, who gave their consent to the inclusion. It was implied that subjects would not object to the inclusion. The study was approved by the Ethics Committee of the Capital Region of Denmark (H-1-2012-041).

Statistical analysis

Data were analyzed using SAS software (Enterprise Guide 7.1, 2014, SAS Institute Inc, Cary, NC, USA). Groups of predictive tested family members were evaluated using chi-square calculations including only individuals alive and above the age of 18 at the time period of accessible analysis (2005 until October 2017).

Analyses of risk factors for AAO, AAI and AAD were carried out by survival analysis and included 60 cases (44 affected family members with confirmed FTD-3 and 16 presymptomatic *CHMP2B* mutation carriers). Multivariable regression Cox analyses included genetic risk factors (variables were sex of the subject, sex of the affected parent and parental AAO, AAI and AAD respectively) and life style factors (variables were smoking, physical exercise, alcohol consumption and education). Life style factors were analyzed dichotomously as 1) have subject ever smoked, 2) has subject performed physical exercise on a regular weekly basis and 3) have subject ever had a daily intake of alcohol? When available, estimated weekly alcohol consumption were also included. In a subsequent analysis, ApoE genotype was analyzed as a risk factor.

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The small number of observations necessitated that genetic risk factors (sex, sex of affected parent and parental AAO, AAI and AAD respectively) were analyzed separately from life style factors (smoking, physical exercise, alcohol consumption and education). As ApoE genotype was available in only 33 mutation carriers, this analysis included only ApoE genotype and sex.Although all individuals were members of the same family, subjects in sibships were considered correlated, necessitating the use of a Cox analysis with robust standard errors allowing for non-independent observations (The PHREG procedure with the COVS AGGREGATE option in SAS®) [13,14]. Subjects were grouped in sibships by a parent identification number (parent-ID). We recently reported AAO correlated to family branch in a smaller study [12], so by nesting subjects by parent-ID, we adjusted for both branch and sibship (the ID statement in SAS®). This approach has been successfully applied in similar analyses of genetic susceptibilities in families [12,15]. The possibility of confounding was further reduced by including year of birth as a covariate in the Cox model. All identified *CHMP2B* mutation carriers were included in this study.

Results are given as Hazard Ratios (HR) with 95% Confidence Intervals (CI). All p-values are stated without correction for multiple comparisons, and $p<0.05$ is considered significant.

Estimates on anticipation and life expectancy included all family members, applying an Age at End of Study (AES). Anticipation of AAO was assessed using the mean of the difference: AAO – parental AAO, or in presymptomatic mutation carriers AES – parental AAO, assuming that anticipation difference followed a normal distribution, but allowing for the fact that some differences where right censored [16].

In establishing the life expectancy of *CHMP2B* carriers, we chose to match each confirmed *CHMP2B* carrier to the general Danish population. Tables of the average expected lifetime in Danes are

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available through the public institution Statistics Denmark (Danmarks Statistik) and provide age and sex specific mortality rates. We modeled the age and sex specific mortality rates in *CHMP2B* carriers as proportional to those of the Danish population with a hazard ratio describing the size of the increased mortality. Since *CHMP2B* carriers had to be recognized as carriers to be included, we developed a model with delayed entry. Thus, subjects were under risk of dying only from their age when the presence of the *CHMP2B* mutation was apparent. For affected family members in the early generations this would most often be AAO, while in later generations and in presymptomatic carriers this would be the age at genetic testing. To illustrate the increased mortality of the carriers, we used the survival model to determine the mean remaining lifetime at age 20 in different years.

Data availability

The data supporting the findings of this study is available from the FReJA collaboration upon reasonable request including local ethical approval. The data are not publicly available as genetic status of subjects cannot be disclosed.

RESULTS

Clinical update

The pedigree of the FTD-3 family is shown in Figure 1. It comprises 528 individuals across six generations. Affected members are indicated, while the genetic status of unaffected family members cannot be disclosed. The low numbers of confirmed mutation carriers in the fifth and sixth generations are due to young age in these generations, and the number is likely to increase in the coming years.

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All 51 clinical cases are presented in Table 1, and the 44 cases of confirmed carriers of the *CHMP2B* mutation are stated. Clinical descriptions of twelve selected cases are given in the Supplementary Material.

The clinical presentation was found highly heterogeneous including both cognitive and behavioural symptoms at an early stage of disease. Apathy, disinhibited behaviour and loss of empathy were common features, while aggressiveness and hypersexuality was less commonly reported. Cognitive deficits included dyspraxia and overall loss of executive functions, while memory was often preserved. Loss of insight was an early feature of almost every affected FTD-3 patient. Language impairments were common, often presenting as non-fluent aphasia and progressing into mutism. Extrapyramidal symptoms of bradykinesia and rigidity were commonly reported. With progression came gait impairment and immobilization. Fasciculations were reported in a few patients.
Cause of death as stated in medical records was most often ascribed to the immobilization caused by the neurodegenerative process.

Predictive and diagnostic genetic testing

Since the identification of the disease-causing *CHMP2B* mutation in 2005 [7], 49 family members have been tested, identifying 25 *CHMP2B* mutation negative and 24 *CHMP2B* mutation positive family members (Table 2).

During the period with genetic testing available, 147 family members with affected parents (50% risk individuals) have had the possibility of genetic testing. When including second and third generation children of affected patients, the number of at-risk family members total 356. Of these,

34 unaffected family members have been referred to genetic counseling and predictive testing. Among 50% risk individual, nine were tested mutation positive and 21 were tested mutation negative. When compared to the expected fraction, the number of mutation carriers in this group (29%) was surprisingly low, although this was not statistically significant (X^2=2.5, p=0.11). In four instances, predictive testing was requested by second-degree relatives of affected FTD-3 patients, and among those, only one was tested mutation positive. In one instance, predictive testing was requested by a third-degree relative who tested negative.

Fifteen cases had a diagnostic genetic test due to early behavioural changes. In one of these cases the mutation was not found, and subsequent clinical evaluation found no evidence of a neurodegenerative disease. In the remaining 14 cases, the positive finding of the pathogenic mutation in the *CHMP2B* gene facilitated the early diagnosis of FTD in an early symptomatic family member.

Disease course

Disease course was evaluated among all *CHMP2B* mutation carriers, including 44 clinically affected FTD-3 patients and 16 presymptomatic *CHMP2B* mutation carriers. Survival analyses was applied to Age at Onset (N=44), Age at Institutionalization (N=22), and Age at Death (N=30).

Results are presented in the text and in a table in Supplementary Materials.

Age at Onset (AAO)

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Mean AAO was 58.9 (SD = 5.9) and did not differ between generations (HR: 1.01, CI: 0.64-1.58, p=0.98). Anticipation of AAO from parent to child varied with a large range from –12.6 years to +15.2 years with an estimated mean difference of 1.44 years (95%CI: -1.47; 4.36, p=0.34).

The relationship between AAO and parental AAO is shown in Figure 2; Cox analysis including all *CHMP2B* mutation carriers could not support a correlation between AAO and parental AAO (HR: 0.97, CI: 0.89-1.06, p=0.47).

When sex was analyzed as a single independent factor, AAO seemed neither influenced by the sex of the affected person, nor by the sex of the parent from whom the mutation was inherited. However, when allowing for interaction between sex and the sex of the mutation-carrying parent, a possible risk factor for early onset appeared in women having inherited the pathogenic mutation from their father (Figure 3). The five affected women with a paternally inherited mutation were dispersed in only two sibships, but in Cox analysis accounting for sibship and year of birth, the risk for early onset was significant (HR: 4.27, CI: 1.70-10.73, p=0.002).

AAO was not correlated to either smoking or exercise (HR: 1.07, CI: 0.48-2.38, p=0.86 and HR: 0.58, CI: 0.17-2.00, p=0.38 respectively). Educational level seemed influential in AAO (HR: 1.64, CI: 1.06-2.55, p=0.028), with higher levels of education having an earlier onset. There was a significant correlation to daily alcohol consumption (HR: 4.00, CI: 1.52-10.00, p=0.005), and a correlation between AAO and the amounts of alcohol consumed per week (HR: 1.20, CI: 1.10-1.35, p=0.004). A medical history of depression or stress was correlated to an earlier AAO (HR: 2.50, CI: 1.16-5.26, p=0.018).

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Age at Institutionalization (AAI)

Twenty-two of the 44 *CHMP2B* confirmed FTD-3 cases were institutionalized as disease progressed. Mean AAI was 65.1 (SD = 7.6) and significantly correlated to AAO (HR: 0.83, CI: 0.77-0.90, p<0.0001), meaning that early onset led to early institutionalization. In only six cases, institutionalized patients could be matched to an institutionalized parent, so although survival analysis showed significant correlation between AAI and parental AAI (HR: 0.77, CI: 0.73-0.82, p<0.0001), this result should be interpreted with caution.

Including only individuals who had been institutionalized (N=22), mean duration from AAO to AAI was 5.7 years (SD=3.7) and unaffected by lifestyle factors like smoking, exercise, alcohol consumption and educational level (HR: 2.49, CI: 0.31-20.33, p=0.39; HR: 3.99, CI: 0.73-21.79, p=0.11; HR: 0.71, CI: 0.24-2.15, p=0.55; and HR: 1.68, CI: 0.81-3.51, p=0.17 respectively).

In the subsequent analysis of ApoE genotype, the ε4/ ε4 carriers had a significantly delayed AAI (HR: 0.26, CI: 0.08-0.80, p=0.019), suggesting a protective role of ε4.

Age at Death (AAD)

Of the 44 affected family members, 30 had died at the time of analysis.

In early generations, cause of death was recorded as cerebral insult (two cases) and cardiac failure (two cases). In five later cases, medical records stated cause of death as concomitant cancer illnesses. All other deaths were associated with the immobilization in late stage neurodegenerative disease, most often ascribed to pneumonia.

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Mean AAD was 68.2 (SD = 8.0). When including only individuals who had died (N=30), mean duration from AAO to AAD was 10.0 years (SD=7.1), but with huge variability from less than six month to more than 27 years. 14 had been institutionalized prior to death, permitting the analysis of correlation between AAI and AAD. AAD was significantly correlated to AAI, but not to AAO (HR: 0.84, CI: 0.74-0.95, p=0.006 and HR: 1.12, CI: 0.93-1.35, p=0.25 respectively). AAD was not significantly correlated to parental AAD (HR: 1.03, CI: 0.93-1.13, p=0.57).

Of the 30 deceased, 19 were women and 11 were men. AAD was unrelated to sex (HR: 0.69, CI: 0.35-1.37, p=0.29). Women seemed to live longer with the disease from AAO to AAD, but not from AAI to AAD (HR: 0.40, CI: 0.20-0.78, p=0.008 and HR: 0.56, CI: 0.19-1.63, p=0.29 respectively). At the time of analysis, all women having inherited the pathogenic mutation from their father (N=5) had also died. Nevertheless, the risk for women having inherited the pathogenic mutation from their father was not reproduced in terms of AAD (HR: 0.93, CI: 0.43-2.01, p=0.85), as the earlier onset was compensated by longer duration from AAO to AAD (HR: 0.39, CI: 0.21-0.75, p=0.005).

Comparing the life expectancy of *CHMP2B* mutation carriers to the general Danish population established a significant increase in mortality rates (HR: 3.84, CI: 2.55-5.79, p<0.0001). The effect did not depend significantly on sex and birth year. The increased mortality was illustrated as a reduction in expected remaining life time at age 20. The life expectancies after the age of 20 are illustrated throughout the century in Figure 4, comparing *CHMP2B* mutation carriers to the general Danish population. In 2015, these life expectancies were 63 and 59 years for Danish women and men respectively, while in *CHMP2B* mutation carriers the expected life time after age 20 was 50.5 and 45.5 years respectively, corresponding to an expected loss of about 13 years.

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DISCUSSION

Clinical presentation

Most cases in the Danish family fulfilled diagnostic criteria for FTD. In addition to behavioural symptoms, FTD-3 patients suffered early executive dysfunctions. This was in accordance with previous reports of a global cognitive decline in otherwise presymptomatic *CHMP2B* mutation carriers [6]. Dystonia and signs of motor neuron involvement were observed at an early stage. In the later stages of disease, extrapyramidal features were prominent with bradykinesia, rigidity, gait impairment and immobilization. The clinical presentations are heterogeneous, but characteristic of an FTD with extrapyramidal features and motor neuron involvement. The pedigree and the segregation of the disease with the *CHMP2B* mutation substantiate the autosomal dominant inheritance with complete penetrance.

Predictive genetic testing

Molecular genetic testing for disease-causing mutations in genes associated with FTD is possible, and in families with a known genetic etiology, predictive testing can be offered to family members at risk of having inherited a pathogenic mutation. Among the persons in the Danish family having a 50% disease risk, a relatively low fraction of 20% has requested genetic counseling and subsequent predictive testing. The low fraction is consistent with similar estimates in other familial FTDs and Huntington's disease, in which less than 20% of at-risk persons choose predictive testing [17,18]. That so many at-risk family members choose to live with the uncertainty rather than the knowledge of a future disease may in part be explained by the fact that neither treatment nor prevention is currently available in these diseases [19–21].

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Among family members requesting genetic counseling and predictive testing, only 29% carried the pathogenic mutation. The most obvious explanation for this imbalance is that at-risk persons postpone testing to a late stage in life, when development of disease seems unlikely to them. Although speculative, another possible reason for this imbalance could be an early pathological lack of insight among mutation carriers, leaving the at-risk person unaware or indifferent to the consequences of carrying the disease-causing mutation. In concordance with this possible mechanism, neuropsychological evaluation of mutation carriers has shown an early decline of cognition prior to onset of behavioural changes [6]. Additionally, structural changes and biochemical signs of neurodegeneration appear early in presymptomatic carriers of FTD causing mutations [22–26].

Risk factors

The heterogeneity of FTD-3 is reflected in the finding that onset and progression appeared unpredictable. A correlation between AAO and parental AAO could not be established. With differences in AAO between parent and child as high as 15 years, and with differences in AAO within sibships as high as 17 years, the AAO appeared unpredictable based on parental AAO. Similarly high variability of AAO has been found in a large Belgian family of *GRN* mediated FTD [27]. The obvious assumption that AAO would correlate to AAO of the affected parent has also been refuted in a larger study of *MAPT*, *GRN* and *C9orf72* FTD, although mean AAO seemed to be correlated within families [25].

The sex of the mutation carrier was not in itself found to be a risk factor of early AAO, AAI or AAD. However, women with a paternally inherited mutation appeared to have an early onset but were not institutionalized or died at an earlier age than the rest of the cohort. Consequently, the early

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onset was followed by a proportionately prolonged disease duration. To the best of our knowledge this is the first report of an interaction between sex and parental sex; whether this is related to disease specific pathomechanisms remains speculative.

Known non-genetic risk factors for FTD are scarce. Demographics and presymptomatic illnesses are without influence in predicting onset and prognosis [28–30], although one study found that a former head trauma correlated with early AAO [31]. Another study reported an increased risk of autoimmune disease with FTD, pointing to an inflammatory component in the pathogenesis [32]. Earlier studies have suggested an inflammatory component in FTD-3 [33], but in the cases presented here, no obvious risk factors in medical history were identified.

Neither onset, nor progression, nor death was correlated to the vascular risk factors of smoking and lack of physical exercise. Although earlier imaging has suggested a vascular pathology in FTD-3 [24], the findings presented here were in accordance with earlier reports finding no differences in vascular risk factors in cases of sporadic FTD compared to non-FTD dementia cases [31,34].

The role of education in the progression of FTD is difficult to establish. Some studies have found higher education and cognitive reserve to seemingly play a role in progression [35,36], while another found education of no influence on survival [28]. The finding in the FTD-3 cohort that individuals with higher education had an apparent earlier onset than individuals with short education should not imply causality. Rather, this association probably reflects an early dysexecutive syndrome in FTD-3, unmasked earlier in people of high education and accordingly higher work-related requirements. This would lead to an early withdrawal from work and thus attention to other changes in behaviour and cognition. Certainly, in several cases, AAO was given as the time of withdrawal from work.

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Alcohol consumption was significantly correlated to AAO in the sense that daily alcohol consumption lowered the AAO. This association should however be interpreted with caution, as over consumption of alcohol can be an early feature of FTD.

A medical history of depression or stress seemed to reduce AAO, but as in the case of alcohol consumption, this correlation does not prove causality. Rather, depression should be considered a first symptom of onset, probably reflecting apathy or emotional impairment.

Overall, we did not identify any convincing predictors of disease onset.

Progression

Progression from onset to institutionalization and death appeared unpredictable. It was hypothesized that an early onset would imply a more severe and rapidly progressing disease, but this could not be demonstrated. Duration from AAO to AAI and to AAD was independent of AAO, meaning that an early onset did not predict a more rapid progression of disease. The high variability in duration from AAO to AAI and AAD further underlined the unpredictability of the disease (Supplementary figure 1). These findings are in accordance with earlier studies showing no predictability of disease severity from time of onset [37], and a high variability in survival after diagnosis [38]. The expectation that young onset FTD would be more favorable in life expectancy (as demonstrated in one review [39]) was not reproduced here.

In analyzing factors of AAI, the ApoE genotype ε4/ε4 appeared to delay institutionalization. The role of ApoE in FTD-3 has previously been described in detail [12], although the original study adjusted for branch only, while the present study adjusted for both branch and sibship. Nevertheless, the previously described protective role of ApoE ε4 is reproduced here.

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As demonstrated, no other investigated factors could predict disease severity and progression.

Death

FTD is a fatal neurodegenerative disease with reduced life expectancy [38–40]. Although estimates vary, FTD is considered to cause a relatively greater reduction in life expectancy than other types of dementia, not only due to the early onset, but also due to the severity and rapidity of neurodegeneration [40]. By matching each *CHMP2B* mutation positive case in the family with the general Danish population, we demonstrated a significant reduction in life expectancy of 13 years in *CHMP2B* mutation carriers. This estimate is consistent with similar estimates in bvFTD [38,39,41] and substantiates FTD-3 as a fatal disease.

Strengths and limitations

We included all reported cases in the clinical description, but excluded seven deceased cases in the statistical analysis, because the presence of a *CHMP2B* mutation could not be confirmed by biosamples or by genotype in their descendants. This criterion preselected family members with many children or with children who had asked for genetic testing, but it secured the genetic homogeneity of the cohort. The relatively low number of cases was somewhat compensated by the inclusion of presymptomatic carriers, although not all presymptomatic carriers in the family have been identified.

Information on AAO, AAI and AAD were available in all cases, and consequently these factors were chosen as the descriptive endpoints of disease course. With an onset of subtle personality changes, the transition from presymptomatic to behavioural symptomatic disease is difficult to assess and

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consequently, AAO was liable to reporting errors. AAI was considered a more uniform and reliable measure of disease severity and progression. AAD is indisputable, although not always related to FTD.

The multivariable regression Cox analysis allowed for multiple comparisons, and although the low number of observations necessitated the division of analyses into genetic and lifestyle factors respectively, p-values were stated without post hoc corrections. Establishing correlations in multiple regression necessitates large cohorts, and consequently possible modifiers of disease could have been overlooked.

In the assessment of life expectancy in *CHMP2B* carriers, the small sample size was compensated by comparing all verified *CHMP2B* mutation carriers to the general Danish population.

Conclusion

The identified pathogenic mutation in the *CHMP2B* gene causes an autosomal dominantly inherited FTD with complete and age-related penetrance, with a heterogeneous phenotype including behavioural changes, cognitive decline, extrapyramidal symptoms and motor neuron signs. Predictive genetic testing is chosen infrequently by at-risk persons. Onset and progression appear unpredictable. Life expectancy is reduced by 13 years.

The Danish FTD-3 family is a well described cohort, and the demographic and clinical characteristics provided here may be applicable to other FTDs, genetic as well as sporadic.

Conflict of Interest and Sources of Funding Statement

The authors have no conflict of interest to declare.

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Data Availability:

The data supporting the findings of this study is available from the FReJA collaboration upon

reasonable request including local ethical approval

TABLES

Table 1:

Clinical presentation of FTD-3

AAO: Age at Onset; AAI: Age at Institutionalization; AAD: Age at Death; MND: Motor neuron signs present; EPS: Extrapyramidal signs present.

Description available 1: In Gydesen et al. Neurology. 2002. Supplementary Material; 2: In Gydesen et al. Neurology. 2002; 3: In Lindquist et al. Eur J Neurol. 2008; 4: In Appendix

FTD-3 update								
Gene-ration	AAO	AAI	AAD	Clinical Description	MND	EPS	*CHMP2B* mutation	Description available
I	56		68	Apathy and mutism			By descendants	1
II	61		71	Aggressive behaviour			By descendants	1
II	60	63	64	Aggressive behaviour		X	By descendants	1
II	57	62	64	Apathy and aggressive behaviour			By descendants	1
II	57	60	65	Apathy and aggressive behaviour		X	By descendants	1
II	64		67	Euphoria and aggressive behaviour			By descendants	1
II	59		65	Anger and persevatory behaviour			By descendants	1
II	54		61	Aggressive behaviour			By descendants	1
II	67		71	Cognitive problems			By descendants	1
II	56		76	Aggressive behaviour and possible apraxia	X	X	By descendants	1
III	63		66	Irritable and restless, stereotypical behaviour			Unconfirmed	1
III	60	73	87	Apathy, disinhibeted and aggressive behaviour		X	Tested	1
III	65		69	Irritability and early dyscalculia			By descendants	1
III	63		64	Personality changes, short history due to early death by other cause			By descendants	
III	57	71	71	Disinhibeted behaviour and early dyspraxia			Unconfirmed	
III	51		59	Apathy and mutism, early loss of memory			By descendants	1
III	51		63	Aphasia, apraxia and visual agnosia			Unconfirmed	1
III	51		61	Impulsive behaviour			By descendants	1

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III	50	55	59	Restlessness, impulsive behaviour, later persevatory speech			Unconfirmed	1
III	64	73	83	Persevatory behaviour		X	By descendants	
III	54	61	78	Apathy, later aggression, loss of insight and disinhibited behaviour			By descendants	2
III	69		80	Unclear history			Unconfirmed	
III	54		69	Apathy and stereotypical behaviour			By descendants	2
III	61	66		Aggressive behaviour, later aphasia			By descendants	
III	62		75	Apathy and stubborness		X	By descendants	1
III	61	69	71	Emotional lability, early loss of memory		X	Unconfirmed	1
III	56		62	Angry and unsocial			Unconfirmed	
III	72	79	79	Loss of memory, persevatory behaviour, loss of insight			Tested	
III	53		70	Apathy and disinhibited behaviour		X	Unconfirmed	1
III	59	72	74	Initially manic episodes, later depression and inertia			Tested	
III	68	77		Apathy and disinhibited behaviour, later aphasia. Choreatic movements.	X	X	Tested	4
III	66		68	Personality changes, short history due to early death by other cause			By descendants	
III	67	71		Apathy, non-fluent aphasia	X	X	Tested	4
III	65			Grouchy and stubborn, possibly jealous, loss of personal hygiene			Tested	4
III	68			Uncritical, loss of insight			Tested	
III	59	61	63	Persevatory behaviour, word finding difficulties			Tested	
III	63	69		Loss of empathy, persevatory in subject of speech, loss of insight	X	X	Tested	4
III	57	64		Disinhibeted behavior, borderline mania, later stereotyped speech			Tested	4
III	51		61	Apathy, loss of insight, persevatory behaviour		X	Tested	1
III	57	61		Ill-tempered, early pronounced aphasia, later apathy and mutism	X	X	Tested	4
III	57	60		Lack of concentration, grouchy and stubborn, shadowing behaviour		X	Tested	4
III	47	49	70	Angry and dilusional		X	By descendants	3
IV	53			Apathy and carelessness, empathy preserved			Tested	4
IV	61			Inappropriate slandering, lack of empathy			Tested	
IV	48		49	Hot-tempered, possible persecutory delusions	X	X	Tested	
IV	58	63		Apathy, early lack of executive functions, novel empathy to relatives	X	X	Tested	4

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IV	58			Hot-tempered, lack of executive functions, later inertia			Tested	4
IV	52			Loss of empathy, inappropriate behaviour, loss of insight	X	X	Tested	4
IV	63	67	70	Carelessness and loss of manners, later stereotyped pacing activity			Tested	4
IV	57	58	67	Disinhibited and aggressive behaviour			Tested	3
IV	49	51	55	Apathy and anger		X	Tested	3

1: In Gydesen S, Brown JM, Brun a, et al. Chromosome 3 linked frontotemporal dementia (FTD-3). Neurology. 2002;59(10):1585-1594, Supplementary Material

2: In Gydesen S, Brown JM, Brun a, et al. Chromosome 3 linked frontotemporal dementia (FTD-3). Neurology. 2002;59(10):1585-1594

3: In Lindquist SG, Braedgaard H, Svenstrup K, Isaacs a M, Nielsen JE. Frontotemporal dementia linked to chromosome 3 (FTD-3)--current concepts and the detection of a previously unknown branch of the Danish FTD-3 family. Eur J Neurol. 2008;15(7):667-670

4: In Appendix

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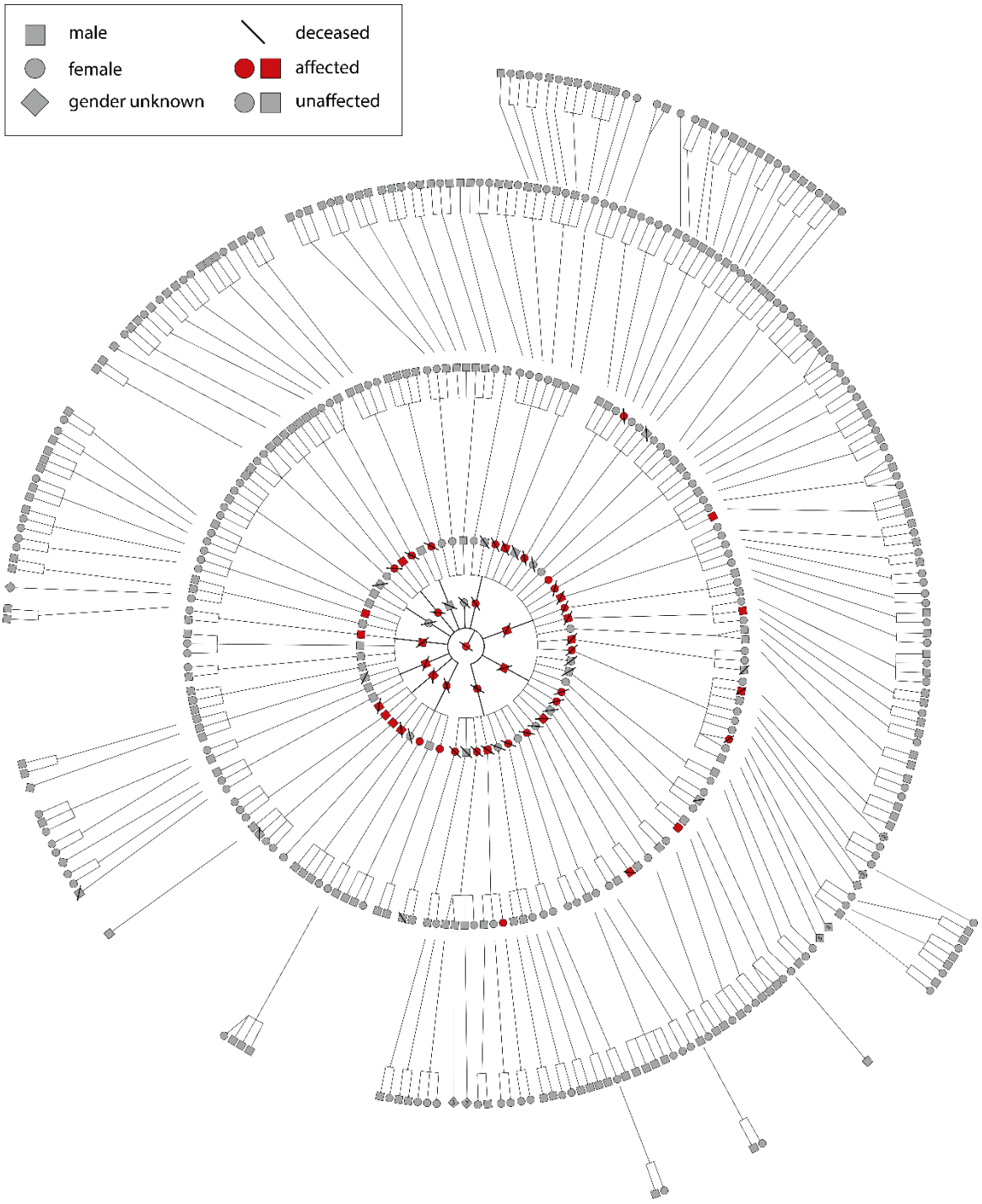
Table 2:

Genetic testing for the *CHMP2B* mutation in the Danish FTD-3 family

		Risk						Total	
		50%		25%		12.5%			
N (family members at risk)		147		126		83		356	
N (family members tested)		45		4		1		50	
Mutation positive (%)	Mutation negative (%)	23 (51)	22 (49)	1 (25)	3 (75)	0	1 (100)	24 (48)	26 (52)
Predictive testing		30		4		1		35	
Mutation positive (%)	Mutation negative (%)	9 (30)	21 (70)	1 (25)	3 (75)	0	1 (100)	10 (29)	25 (71)
Diagnostic testing (%)		15		0		0		15	
Mutation positive (%)	Mutation negative (%)	14 (93.3)	1 (6.7)					14 (93.3)	1 (6.7)

FIGURES

Figure 1: Pedigree of the Danish FTD-3 family.



Six Generations of CHMP2B

Figure 2: Relationship between Age at Onset (ordinate) and parental Age at Onset (abscissa). Symbols represent sibships.

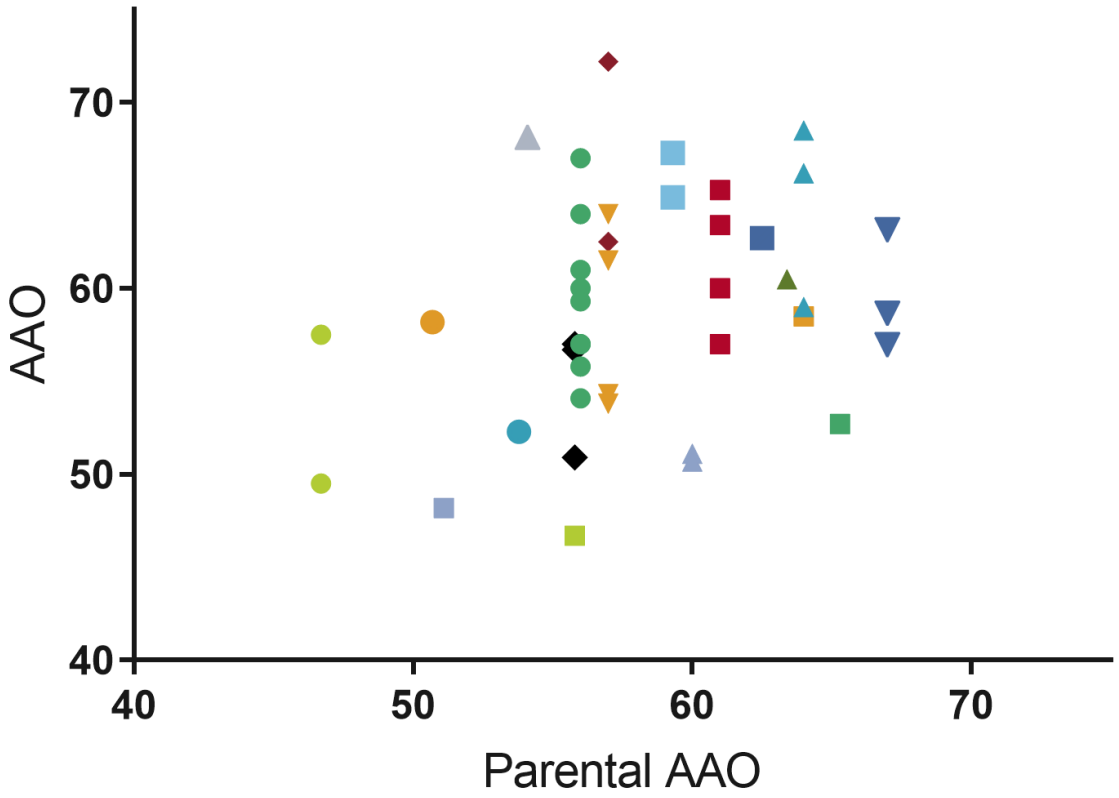


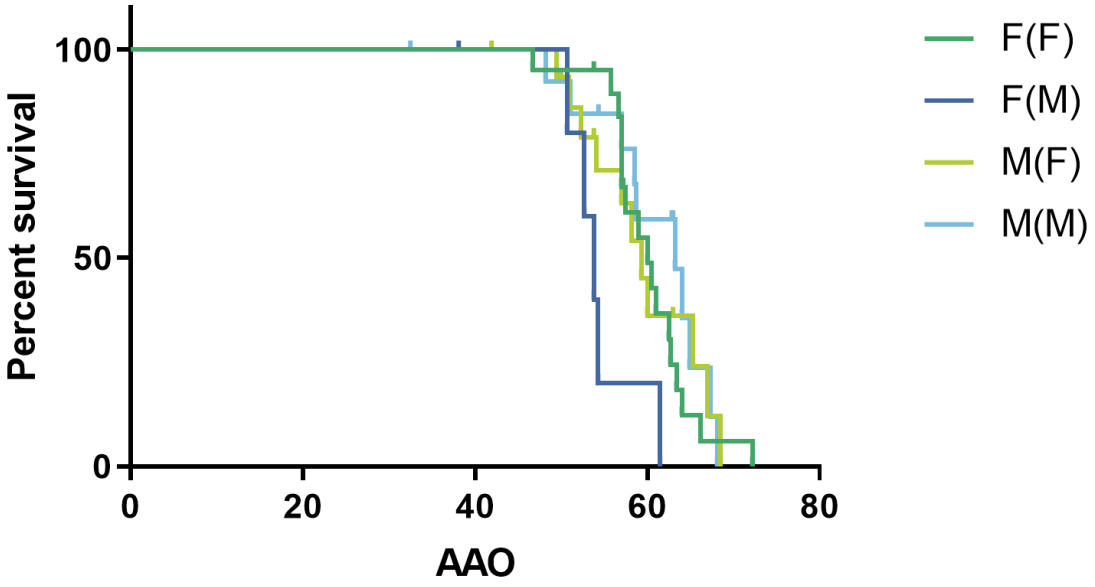
Figure 3: Age at Onset associated to the interaction of sex and the sex of the mutation carrying parent.

F(F): Female with mutation inherited from mother

F(M): Female with mutation inherited from father

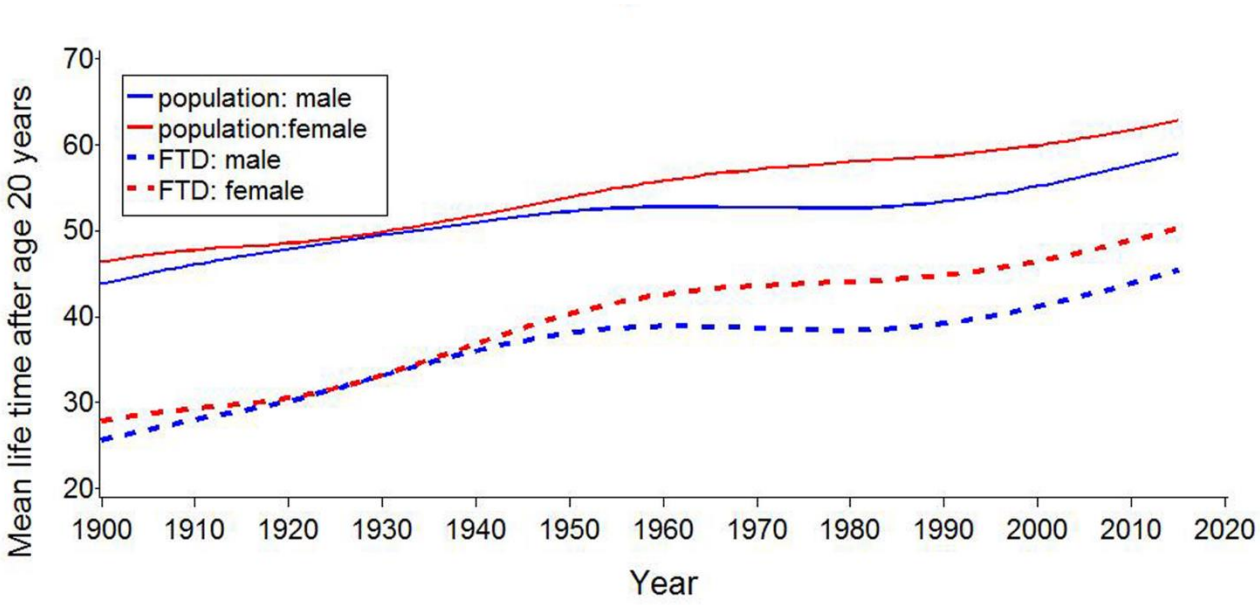
M(F): Male with mutation inherited from mother

M(M): Male with mutation inherited from father

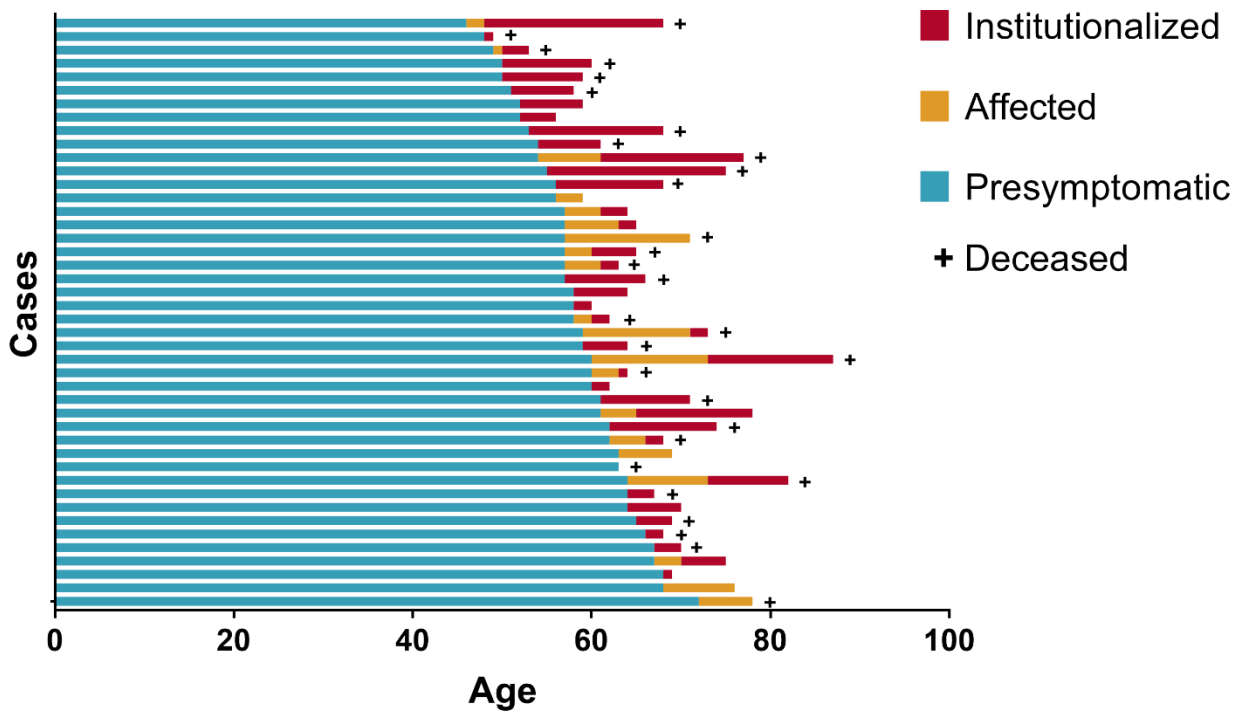


Six Generations of CHMP2B

Figure 4: Life expectancy of *CHMP2B* mutation carriers and the general Danish population.



Supplementary Figure 5: Age at Onset (AAO), Age at Institutionalization (AAI) and Age at Death (AAD) for each case (sorted by AAO). As illustrated, the prognosis is highly heterogeneous and unrelated to AAO.



Six Generations of CHMP2B