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Increased effect of the *ApoE* gene on survival at advanced age in healthy and long-lived Danes: two nationwide cohort studies

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Summary

Studies of Nordic twins suggest an increased genetic influence on mortality with age. Contrary to this, the heterogeneity hypothesis predicts that the mortality of individuals carrying a 'frail' or 'risky' genotype in a population will approach that of noncarriers with age because of selection pressure. The *ApoE ε4* allele is associated with an increased mortality risk, and its effect has been suggested to decrease with age. Here, we investigated the effect of *ApoE ε4* allele on survival in a sample of the healthiest and long-lived Danes. The study population comprised Danes born in 1905 and a replicate sample of the 1895 cohort. For the 1905 cohort, a total of 350 carriers and 1256 noncarriers of the *ApoE ε4* allele were followed from 1998 until death or end of follow-up. Cox regression models were used for the analysis. Of the 1606 persons with known *ApoE ε4* status in 1998, 1546 had died at the end of the 10-year follow-up. Carriers of the *ApoE ε4* allele had an increased mortality compared to noncarriers, and the influence of *ApoE* status on mortality increased in the age interval 92–103. For the covariates sex and independency status, the difference in relative risk of death between groups decreased with advancing age. Our findings of increasing influence of *ApoE ε4* allele on mortality with age do not support previous findings of decreased influence *ApoE ε4* allele on mortality with age, and alternative models such as the multifactorial threshold models should be considered for understanding the genetic effects on mortality at advanced age.

Keywords

ApoE; frailty; heterogenity; longevity; mortality; oldest old

Introduction

One intrinsic question in human longevity research is how the human genome influences our longevity. Specific combinations of genes that lead to a long life and others that cause early death may exist, and the perspective of the identification of such genes for the understanding of the human life span is central.

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Danish and Swedish twin studies have found that genetic factors account for about a quarter of the variation in human life span in contemporary Nordic populations (Herskind *et al*., 1996; Skytthe *et al*., 2003). Combined studies of Nordic twins suggest an increased genetic influence on mortality with age (Hjelmborg *et al*., 2006). Contrary to this view, the heterogeneity hypothesis predicts that the mortality of individuals carrying a 'frail' or 'risky' genotype in a population will approach that of noncarriers with age because of a selection pressure on carriers (i.e., only the otherwise most robust individuals will survive with a 'frail' genotype) (Christensen *et al*., 2006; Ewbank, 2007). Also, the diversity of sufficient causes needed for death is expected to increase with age leading to competing risks of death.

ApoE polymorphisms are the most well-documented common gene variants that influence life span. The *ApoE ε4* allele is associated with an increased mortality risk throughout adulthood (Christensen *et al*., 2006). Although conceptually related, the term 'frailty' is used differently in genetics, medicine, and statistics. The APOE gene has been suggested to be a frailty gene rather than a longevity gene, i.e., a gene that affects the age-specific susceptibility to death, rather than total longevity (Gerdes *et al*., 2000). Although conceptually related, the term 'frailty' is used differently in genetics, medicine, and statistics. In health sciences, frailty is often described as an intermediate condition between morbidity and disability (Fried *et al*., 2001). The statistical concept 'frailty models' was coined by Vaupel in 1979 (Vaupel, 1988). The frailty model assumes that the hazard function for an individual with risk factor X can be written as $Z\alpha$ ($t;X$). Here, Z denotes the unobserved frailty variable. A 'frail' individual has a higher value of Z and thus a higher risk of dying.

Here, we investigate the age dependency of *ApoE* genotype at the highest age by following the Danish 1905 birth cohort. For replication and to increase sample size at the highest ages, persons from the Longitudinal Study of Danish Centenarians (LSDC) (Danes born in 1895 aged 100 and above) were followed from age 100 until death (Andersen-Ranberg *et al*., 2001). At this age span, the selection pressure is very high. For the 1905 birth cohort, the chance of survival from birth to age 92 is about 1/15 and the chance of surviving from age 92 to 100 is again 1/15, so half of the selection from birth to age 100 takes place in the age interval under study here. Based on predictions from our initial work (Bathum *et al*., 2006), results from many previous studies (Ewbank, 2002, 2004, 2007), and the heterogeneity hypothesis, we would expect the effect of *ApoE* genotype on mortality to decline with age. On the other hand, casual field models (multifactorial threshold models) and the Nordic twin studies cited earlier (Hjelmborg *et al*., 2006) suggest an increasing mortality risk of *ApoE ε4* allele with age.

Materials and methods

Study population

The study was based on a Danish cohort of persons born in 1905 (1905-cohort) and a replication cohort of persons born in 1895–1896 (LSDC).

The Danish 1905 Cohort Survey is a nationwide longitudinal survey consisting of all individuals born in Denmark in 1905 identified through the Danish Civil Registration System. At baseline in 1998, a total of 3600 persons aged 92–93 years were still alive in the cohort, and 2262 of these persons participated in the intake survey (63%). The baseline interview and successive follow-ups consisted of a personal interview in the respondents' home and included information on sociodemographic variables, health, and self-reported activities of daily living (ADL). Trained interviewers from the Danish National Institute of Social Research carried out the survey. Since then, in-person follow-ups have been conducted every 2–3 years, the most recent one taking place in 2005. A nonresponse analysis showed no difference between responders and nonresponders with respect to gender (women, 84% vs. 89%, $P = 0.16$), median number of hospitalizations ($P = 0.56$), or median number of bed days ($P = 0.71$) during the

years 2003 and 2004 (Engberg *et al*., 2008). This cohort was used for the simple descriptions of the observed data on age-specific mortality rates calculated from life table statistics (Preston *et al*., 2001). The 1905 birth cohort study was further used for the rest of the statistical modeling. Data on age-specific mortality for all Danes born in 1905 (i.e., participants plus nonparticipants in the present study) were retrieved from the Danish Central Person Registry, which holds information on all Danes and their death and migrations.

The Longitudinal Study of Danish Centenarians comprises all individuals reaching the age of 100 years in the period from April 1, 1995, through May 31, 1996. A total of 207 of 276 eligible centenarians identified through the Danish Civil Registration System participated in the intake survey (75%). No difference between responders and nonresponders with respect to gender, housing, and mean number of hospitalizations in the last 18 years was found in a nonresponder analysis (Andersen-Ranberg *et al*., 2001). The same geriatrician and geriatric nurse visited all respondents. The interview consisted of sociodemo-graphic variables, health, and self-reported ADL. Since the baseline survey, in-person follow-ups have been conducted once every year. The study has previously been described in more detail (Andersen-Ranberg *et al*., 2001). This cohort was included to increase sample size in the age range of 100+. The data were used for the analysis of age-specific mortality rates calculated from life table statistics (Preston *et al*., 2001) (see Fig. 1).

DNA analysis and interview content

In both the cohorts (i.e., 1905 cohort and LSDC), DNA samples were taken from persons who were able to give informed consent. DNA was separated using QIAamp DNA Mini kit (Qiagen, Hilden, Germany), and the Taqman technology was used to genotype the two polymorphisms at amino acid residue 112 and 158 allowing for grouping into *ApoE ε4* carriers and noncarriers. Carriers were defined as anyone carrying at least one *ApoE ε4* allele. Primers and probes were designed using Primer Express software (Applied Biosystems, Foster City, CA, USA).

The participants in the two studies were invited to a home-based 2-h multidimensional interview, covering a wide range of questions and (Nybo *et al*., 2001). Activities of daily living scale was made using five items (bathing, dressing, toileting, transfer, and feeding) covering Katz's ADL index (Katz *et al*., 1963). The two levels were ‘independent’ defined as independent in all items, and ‘dependent’ defined as dependent in one or more items (Andersen *et al*., 2002). We included the variables’ dependency status as a possible confounder for the association between *ApoE* status and mortality. Sex was included and also used as an example of a traditional risk factor expected to have a decreasing effect with age.

Information on date of death in the study populations up to 2008 was retrieved from the Central Population Registry which holds information for the total Danish population since 1968 (Pedersen *et al*., 2006) and is linked to the data using a unique identification number given to all Danish citizens. Therefore, a complete and accurate follow-up was performed for all participants.

To address the question of a possible selection bias in the study, we compared the carriers of the *ApoE ε4* allele with the rest of the Danish population born in 1905.

Statistical analysis

For the initial descriptive analysis, we calculated age-specific mortality from life tables and made Kaplan–Meier survival curves (Preston *et al*., 2001).

Several analytical models were used to investigate the effect of the explanatory variables, written in compact form as X, on the survival. All the models focused on the hazard rate, which we denote by $\acute{a}(t;X)$. The traditional Cox model,

$$\acute{a}(t;X) = \acute{a}_0(t)\exp\left(a_1X_1 + \cdots + a_pX_p\right) \quad (1)$$

was rejected for these data using the score process test of (Lin *et al*., 1993) suggesting time-changing covariate effects. As an exploratory tool to investigate for such behavior, we applied the Aalen additive hazards model (Aalen, 1989). The Aalen additive hazards model specifies the hazard function as

$$\acute{a}(t;X) = \acute{a}_0(t) + \acute{a}_1(t)X_1 + \cdots + \acute{a}_p(t)X_p,$$

where the regression coefficients, $\acute{a}_j(t)$, are allowed to change with time hence not assuming a constant effect of the corresponding covariate. Guided by this analysis, an extended Cox model was applied allowing the covariate effects to change after 2 years from the start of study and again 5 years after the start of study. The effect, a_1X_1, of X_1 in (1) is thus changed to

$$a_{11}X_1 + a_{12}X_{12}(t) + a_{13}X_{13}(t) \quad (2)$$

where $X_{12}(t) = X_1I(2 \le t)$ and $X_{13}(t) = X_1I(5 \le t)$ where $I(B)$ is the indicator of the event B and similarly with the other explanatory variables. The relative risk associated with X_1 is therefore

$$a_{11} \quad t<2 \quad (3)$$

$$a_{11}+a_{12} \quad 2 \le t<5 \quad (4)$$

$$a_{11}+a_{12}+a_{13} \quad 5 \le t \quad (5)$$

and can thus change with time in this specified way. This extended model can be seen as a Cox model with time-varying explanatory variables, which is clear from the expression (2) and can thus be fitted using standard software. The score process test (Lin *et al*., 1993) was now insignificant, and we therefore proceeded with this model. To see whether the potential time-changing covariate effects can be explained as selection effects, the Cox frailty model (Nielsen *et al*., 1992) was applied still allowing for time-changing effects as described earlier. The Cox frailty model is specified as

$$a(t;X;Z) = Z\,\acute{a}(t;X),$$

where Z denotes the frailty variable taken to be gamma distributed. The relative risks given by (3–5) are now the relative risks keeping all other explanatory variables fixed and also the frailty, thus comparing two individuals being equally frail. In this way, the frailty model controls for the potential selection.

Results

Overall findings

For the 1905 cohort, information on *ApoE* status was available for 1606 persons, and 1546 (96%) were dead at the end of the study period (Table 1). About a quarter were carriers of the *ApoE ε4* allele, and ¾ were women in the study population (Table 1). Slightly more than half of the participants were dependent at study start. For the 207 participants from the LSDC, there were 26 carriers of the *ApoE ε4* polymorphism, and 162 were women (Table 1).

From the study start in 1998, both carriers and noncarriers had a lower mortality than the total population born in 1905 and 1895 (Fig. 1). For the noncarriers, the difference in mortality with the total population born in 1905 and 1895 was equal over age. The age-specific mortality of carriers increased over age and was generally higher than that of the mortality of all Danes born in 1905 and 1895. The median survival time for noncarriers was 237 days longer than for carriers.

The ordinary Cox model did not give an adequate fit ($P < 0.001$) indicating that the effects of the considered variables do not remain constant in the considered time span. This was confirmed by the Aalen hazards model fit, where straight lines correspond to constant effect. We see approximate straight lines in the three chosen intervals, but different slopes. Based on these visual inspections, two intervals were chosen of 2 and 5 years after study start. Thereafter, we extended the Cox regression analysis to allow for different effects in these intervals, and this model was accepted using the test that rejected the more simple and ordinary Cox model.

We next applied the Cox model allowing for these time-changing covariate effects. The score process test was nonsignificant indicating an adequate fit of the model. When fitting the statistical model, there was a general tendency for an increase in the relative risk of death with advancing age for carriers versus noncarriers of the *ApoE ε4* allele (Fig. 2). For the carriers of the *ApoE ε4* allele, 350 were at risk from study start, 121 died within the first 2 years of follow-up, 140 died during the next 3 years of follow-up, and 82 died after 5 years of follow-up. Seven carriers were still alive at the end of the study. The relative risk comparing carriers to noncarriers increased for the follow-up time interval of 0–5 years and more than 5 years (indicated by 5+ in Fig. 2) from 1.1 (95% CI: 0.9–1.2, $P = 0.26$) to 1.4 (95% CI: 1.1–1.8, $P = 0.007$).

The difference in relative risk of death between groups decreased with advancing age for the others risk factors, sex and independency status (Fig. 2). There was a significant decrease in the relative risk, comparing men to women (Fig. 2). In the follow-up time interval 0–5 years, the relative risk was estimated to 1.6 (95% CI: 1.4–1.8, $P < 0.001$) and to 0.9 (95% CI: 0.7–1.2, $P = 0.60$) in the later years. For independence status, there was also a significant decrease in the relative risk for independent compared to dependent from the time interval 0–2 years from start of follow-up to 2–5 years after start of follow-up with a decrease in relative risk from 2.2 (95% CI: 1.9–2.7, $P < 0.001$) to 1.5 (95% CI: 1.3–1.7, $P < 0.001$). For the time interval 2–5 to 5 years after the start of the follow-up, the decrease found was not significant. When using the frailty model that takes selection into account in the statistical model, this had limited effect on the results (solid lines versus dotted lines in Fig. 2), so the above conclusions also hold for this model.

Carriers of the *ApoE ε4* allele had a lower relative risk of dying compared with the rest of the Danish population born in 1905 in the first 2 years of follow-up (RR = 0.64, 95% CI: 0.50–0.80, $P < 0.001$). The relative risk increased to 0.96 (95% CI: 0.83– 1.10, $P = 0.56$) from 2 to 6 years of follow-up and ended with a higher relative risk of 1.24 (95% CI: 0.98, 1.56, $P = 0.07$) at 6+ years of follow-up.

To test whether the time-changing covariate effects may be described as selection effects (Vaupel *et al*., 1979; Vaupel, 1988), we fitted the Cox frailty model assuming the frailty term to be gamma-distributed. We found that the weakening of the effect of independence with time is partly explained by selection. There is still a weakening of the effect with time but it is not as pronounced after controlling for selection, and there is still a significant effect of this predictor 5 years after study start ($P = 0.02$). Figure 3 shows that sex was not a significant predictor 5 years after study start, and the frailty model indicates that this cannot be explained by selection. Controlling for selection had negligible effect on the *ApoE* results. The test for *ApoE ε4* for the Aalen model was $P = 0.14$ indicating an effect, though not significant.

Discussion

We found an increased mortality among the carriers of the *ApoE ε4* allele compared to noncarriers and the difference increased in the age interval 92–103. This increase was found for both men and women. Addition of a frailty component to our statistical model improved our model but had only a minor influence on the relative risk estimates.

Previously, we reported no increased risk with age for carriers of the *ApoE ε4* allele after 5 years of follow-up in the 1905 cohort (Bathum *et al*., 2006). The addition of 3 years of follow-up, the inclusion of additional data from the LSDC, and the comparison with all Danes born in 1905 in this study made the power and validity of our analysis stronger than the previous study whereby we now find an effect of the *ApoE ε4* allele on total difference in mortality and an indication that this effect increases with age.

A recent meta study by Ewbank (2007) using data from cohort studies and case–control studies reported that the mortality by genotype diminished at the oldest ages. Ewbank (2002) also reported a decline in the risk over age for carriers of the E4 allele when using a statistical model incorporating heterogeneity (i.e., assuming a gamma distribution). Both studies were based on results from combination of cross-sectional, case–control and cohort studies from different countries and including a large number of individuals (Ewbank, 2002, 2007). In our study, the frailty component (i.e., heterogeneity) had little influence on the relative risks between carriers and noncarriers at very old age.

When viewed in the multifactorial threshold model perspective, the difference between this study and the studies reviewed by Ewbank (2002, 2007) may be because of the very old age groups analyzed in the present studies where many other components leading to death may be fulfilled, increasing the influence of having an E4 genotype. Furthermore, the reason for the difference in the findings may be the rather large variability in study populations and designs used in the studies by Ewbank (2002, 2007) when compared to the more homogeneous study population used in the present cohort study. Also, the large sample size in this cohort study and the reduction in secular induced noise from changes in competing risk factors, as well as the minimizing of the differences in selection pressure between genetic groups when using the same ethnic homogeneous birth cohort, make us more confident that no decline in the risk between *ApoE ε4* carriers and noncarriers with age is present in the Danish 1905 birth cohort.

The difference we found for a change with increasing age between carriers and noncarriers of the *ApoE ε4* allele is in line with the findings of Hjelmborg *et al*. (2006) an increased genetic influence on mortality with age. This is in contrast to the view of the *ApoE* gene being a traditional frailty gene (Gerdes *et al*., 2000; Fried *et al*., 2001; Christensen *et al*., 2006). In the demographic perspective, a frailty gene effects the age-specific susceptibility to death (Christensen *et al*., 2006). Mortality of individuals carrying the ‘frail’ genotype in a population will approach that of noncarriers with age because of a selection pressure on carriers (Christensen *et al*., 2006), the reason being that the surviving carriers possess other factors

(i.e., heterogeneity becomes a more important factor) that compensate for the more frail genotype. Rather, our study supports a multifactorial threshold models where many single factors with small effect add up and contribute to a disorder/condition (here death) until a threshold after which the disorder occurs. This model imposes the possibility of a scenario where there is an increasing frequency in causes needed for an event (e.g., death) to happen with age thus leading to greater influence of a frail genotype. This does not exclude the effect of competing risks as an increasing amount of sufficient causes may be expected with increasing age. In our study, this model fits well with the observed pattern of the oldest age groups having 'filled up' factors leading to death and thereby increasing the influence of the E4 genotype.

In the present study, sex status was chosen to illustrate traditional risk factors expected to have a decrease in the difference in mortality over age. This was also found in this study, supporting that compensating factors for these two traits are present. Such a decrease in influence at advanced age has been seen for several well-known risk factors including marital status, education, smoking, obesity, consumption of alcohol, and number of self-reported diseases (Nybo *et al*., 2003) supporting that the selection pressure is leaving only persons alive with other genetic and/or environmental characteristics that protect them.

The cohort study design of the present study overcomes the problem of a similar case–control design, comparing mortality of different birth cohorts. However, the participation rate of 63% could indicate a potential selection problem. Previous studies on the same cohort have found that participants, when compared to nonparticipants, are a fairly nonselected group (Nybo *et al*., 2003). Among middle-aged men, 31% carried the *ApoE ε4* variant in Denmark (Gerdes *et al*., 2000; Nybo *et al*., 2001), suggesting that the percentage of carriers found among the 93-year-old Danes in the present study is, as expected, lower than 22%.

The fact that our result contradicts previous finding for the relationship between *ApoE* status and age may call for caution in the conclusions drawn. The *ApoE* carriers from 1905 included in the follow-up from 1998 to 2009 in our study can be expected to be among the most robust *ApoE ε4* carriers born in 1905. First of all, the carriers having had to survive to age 92–93 despite being *ApoE ε4* carrier is associated with an increased mortality at younger ages (Gerdes *et al*., 2000). Secondly, among those alive in 1998, the nonparticipants will include a large fraction of terminal individuals, among these many severely demented, which is also associated with *ApoE ε4* carrier status. Despite this selection toward including the healthiest *ApoE* carriers from the 1905 cohort, we can document that over the follow-up period, the mortality risk of *ApoE ε4* carriers increases to a level that is borderline significant ($P = 0.14$) higher than the mortality level for the overall Danish 1905 cohort (complete data through Statistics Denmark). However, replications of our finding in future studies will be needed to increase our understanding of the relationship between *ApoE* status, age, and mortality among the oldest old as differences may exist among ethnically different populations.

In this study, we addressed the question of the genetic influence of the *ApoE* gene on total mortality with age in the Danish 1905 cohort and found that the influence of *ApoE* status on survival increases in the age interval 92–103 in healthy long-lived individuals. This is opposite to the predictions of the heterogeneity hypothesis and points toward other models such as the multi-factorial threshold model.

References

Aalen OO. A linear regression model for the analysis of life times. Stat. Med 1989;8:907–925. [PubMed: 2678347]

Andersen K, Nybo H, Gaist D, et al. Cognitive impairment and mortality among nonagenarians: the Danish 1905 cohort survey. Dement. Geriatr. Cogn. Disord 2002;13:156–163. [PubMed: 11893837]

Andersen-Ranberg K, Schroll M, Jeune B. Healthy centenarians do not exist, but autonomous centenarians do: a population-based study of morbidity among Danish centenarians. J. Am. Geriatr. Soc 2001;49:900–908. [PubMed: 11527481]

Bathum L, Christiansen L, Jeune B, Vaupel J, McGue M, Christensen K. Apolipoprotein e genotypes: relationship to cognitive functioning, cognitive decline, and survival in nonagenarians. J. Am. Geriatr. Soc 2006;54:654–658. [PubMed: 16686878]

Christensen K, Johnson TE, Vaupel JW. The quest for genetic determinants of human longevity: challenges and insights. Nat. Rev. Genet 2006;7:436–448. [PubMed: 16708071]

Engberg H, Christensen K, Andersen-Ranberg K, Vaupel JW, Jeune B. Improving activities of daily living in danish centenarians – but only in women: a comparative study of two birth cohorts born in 1895 and 1905. J. Gerontol. A Biol. Sci. Med. Sci 2008;63:1186–1192. [PubMed: 19038833]

Ewbank DC. Mortality differences by APOE genotype estimated from demographic synthesis. Genet. Epidemiol 2002;22:146–155. [PubMed: 11788960]

Ewbank DC. The APOE gene and differences in life expectancy in Europe. J. Gerontol. A Biol. Sci. Med. Sci 2004;59:16–20. [PubMed: 14718482]

Ewbank DC. Differences in the association between apolipoprotein E genotype and mortality across populations. J. Gerontol. A Biol. Sci. Med. Sci 2007;62:899–907. [PubMed: 17702883]

Fried LP, Tangen CM, Walston J, et al. Frailty in older adults: evidence for a phenotype. J. Gerontol. A Biol. Sci. Med. Sci 2001;56:M146–M156. [PubMed: 11253156]

Gerdes LU, Jeune B, Ranberg KA, Nybo H, Vaupel JW. Estimation of apolipoprotein E genotype-specific relative mortality risks from the distribution of genotypes in centenarians and middle-aged men: apolipoprotein E gene is a “frailty gene,” not a “longevity gene”. Genet. Epidemiol 2000;19:202–210. [PubMed: 11015124]

Herskind AM, McGue M, Holm NV, Sorensen TI, Harvald B, Vaupel JW. The heritability of human longevity: a population-based study of 2872 Danish twin pairs born 1870–1900. Hum. Genet 1996;97:319–323. [PubMed: 8786073]

Hjelmborg JV, Iachine I, Skytthe A, et al. Genetic influence on human lifespan and longevity. Hum. Genet 2006;119:312–321. [PubMed: 16463022]

Katz S, Ford AB, Moskowitz RW, Jackson BA, Jaffe MW. Studies of illness in the aged. The index of ADL: a standardized measure of biological and psychosocial function. JAMA 1963;185:914–919. [PubMed: 14044222]

Lin DY, Wei LJ, Ying Z. Checking the Cox model with cumulative sums of martingale-based residuals. Biometrika 1993;80:557–572.

Nielsen GG, Gill RD, Andersen PK, Sørensen TIAS. A counting process approach to maximum likelihood estimation in frailty models. Scand J. Stat 1992;19:25–44.

Nybo H, Gaist D, Jeune B, et al. The Danish 1905 cohort: a genetic-epidemiological nationwide survey. J. Aging Health 2001;13:32–46. [PubMed: 11503846]

Nybo H, Petersen HC, Gaist D, et al. Predictors of mortality in 2,249 nonagenarians – the Danish 1905-Cohort Survey. J. Am. Geriatr. Soc 2003;51:1365–1373. [PubMed: 14511155]

Pedersen CB, Gotzsche H, Moller JO, Mortensen PB. The Danish Civil Registration System. A cohort of eight million persons. Dan. Med. Bull 2006;53:441–449. [PubMed: 17150149]

Preston, SH.; Heuveline, P.; Guillot, M. Demography: Measuring and Modeling Population Processes. Blackwell Publishing; Oxford, UK: 2001.

Skytthe A, Pedersen NL, Kaprio J, Stazi MA, Hjelmborg JV, Iachine I, Vaupel JW, Christensen K. Longevity studies in GenomEU-twin. Twin Res 2003;6:448–454. [PubMed: 14624729]

Vaupel JW. Inherited frailty and longevity. Demography 1988;25:277–287. [PubMed: 3396752]

Vaupel JW, Manton KG, Stallard E. The impact of heterogeneity in individual frailty on the dynamics of mortality. Demography 1979;16:439–454. [PubMed: 510638]

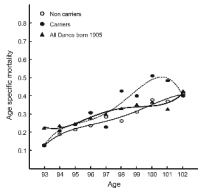

Fig. 1.

Data from the Danish 1905 cohort, The Longitudinal Study of Centenarians and for all Danes born in 1905 showing age-specific mortality by APOE4 status. Above age 99, data from The Longitudinal Study of Centenarians were added to the 1905 cohort data to improve power.

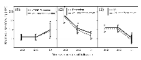

Fig. 2.

The graphs illustrate the relative risk of dying for intervals of years since start of follow up for (1) *ApoE ε4* carriers vs. noncarriers, (2) dependent vs. independent, and (3) men vs. women. *ApoE*, dependency status, and sex are adjusted for each other, and vertical lines represent 95% confidence intervals. Solid lines represent a model where selection is incorporated and adjusted for.

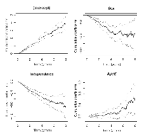

Fig. 3.
Aalen additive hazards model fit. Estimated cumulative regression coefficients along with 95% pointwise confidence bands (see text for explanation).

Table 1

Basic characteristics of the study population

	Number/deaths (% of total deaths)	
Variable	**Men**	**Women**
1905 cohort		
Dependency status		
Independent	210/207 (47)	609/595 (51)
Dependent	241/228 (53)	546/516 (49)
ApoE ε4 status		
Carriers	98/96 (22)	252/247 (22)
Noncarriers	353/339 (78)	903/864 (78)
Longitudinal Study of Danish Centenarians (1895 cohort)		
Dependency status		
Independent	15/15 (33)	35/35 (22)
Dependent	30/30 (67)	127/127 (78)
ApoE ε4 status		
Carriers	7/7 (21)	19/19 (16)
Noncarriers	26/26 (79)	89/89 (84)