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ORIGINAL INVESTIGATION

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Genetic influence on human lifespan and longevity

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Abstract There is an intense search for longevity genes in both animal models and humans. Human family studies have indicated that a modest amount of the overall variation in adult lifespan (approximately 20–30%) is accounted for by genetic factors. But it is not known if genetic factors become increasingly important for survival at the oldest ages. We study the genetic influence on human lifespan and how it varies with age using the almost extinct cohorts of Danish, Finnish and Swedish twins born between 1870 and 1910 comprising 20,502 individuals followed until 2003–2004. We first estimate mean lifespan of twins by lifespan of co-twin and then turn to the relative recurrence risk of surviving to a given age. Mean lifespan for male monozygotic (MZ) twins increases 0.39 [95% CI (0.28, 0.50)] years for every year his co-twin survives past age 60 years. This rate is significantly greater than the rate of 0.21 (0.11, 0.30) for dizygotic (DZ) males. Females and males have similar rates and these are negligible before age 60 for both MZ and DZ pairs. We moreover find that having a co-twin surviving to old ages substantially and significantly increases the chance of reaching the same old age and this chance is higher for MZ than for DZ twins. The relative recurrence risk of reaching age 92 is 4.8 (2.2, 7.5) for MZ males, which is significantly greater than the 1.8 (0.10, 3.4) for DZ males. The patterns for females and males are very similar, but with a shift of the female pattern with age that corresponds to the better female survival. Similar results arise when considering only those Nordic twins that survived past 75 years of age. The present large population based study shows genetic influence on human lifespan. While the estimated overall strength of genetic influence is compatible with previous studies, we find that genetic influences on lifespan are minimal prior to age 60 but increase thereafter. These findings provide a support for the search for genes affecting longevity in humans, especially at advanced ages.

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Introduction

The success in identifying longevity genes in Nematoda, *Drosophila* and mouse has facilitated large scale efforts to identify longevity genes in humans (Johnson 2005). Human studies have suggested a moderate clustering of extreme longevity within families (Kerber et al. 2001; Perls and Dellara 2003), and twin studies have indicated that approximately 20–30% of the overall variation in lifespan can be attributed to genetic factors (Herskind et al. 1996; Ljungquist et al. 1998; Skytthe et al. 2003). Our aim is to study the *age dependence* of genetic influence on lifespan. Survival to older ages, e.g. 75, could change the genetic influence on lifespan in either direction. That is, removing early deaths could induce a reduction in genetic influence because the influence of genetic diseases with high early mortality such as familial hypercholesterolemia and cystic fibrosis (genetic diseases with increased mortality at younger ages) are removed. On the other hand, many violent deaths are excluded when considering only those who survived to

older ages and this could increase genetic influences, relatively speaking, by removing early 'random' deaths. Knowing whether total genetic influences increase, remain constant or decrease with age may contribute to efforts of identifying genes that affect human lifespan, and provide information about the feasibility of (environmental) interventions.

Surviving to a given age is probably a multifactorial phenotype involving multiple biological processes, environmental influences and randomness. In particular, it has been speculated whether and how genes are involved in reaching the highest ages, which are central questions to the many ongoing efforts aimed at identifying longevity-genes (Tan et al. 2004). As mentioned family studies have shown a moderate clustering of extreme longevity within families. However, traditional family studies cannot, unlike twin studies, disentangle the effect of genes and environment. Furthermore, even if genetic in origin this finding may be due to a clustering of genetic effects in certain families only and thus may not be detectable in populations.

To study longevity in a population a large study base is required. To this end, we were able to consider data from the Swedish, Finnish and Danish national twin registries as part of the GenomEUtwin project. These jointly comprise the largest population-based sample of twins with almost complete lifespans ever studied. These cohorts of Nordic twins show good representativeness for longevity studies. After infancy, twin cohorts have mortality rates similar to the background population (Christensen et al. 1995; Vagero and Leon 1994). This persists when comparing mortality rates of various major causes of death (Christensen et al. 2001), indicating that twin survival is a good model for studying longevity.

Materials and methods

Participants in the study consist of MZ and same sex DZ pairs from the Danish, Finnish and Swedish twin cohorts born between 1870 and 1910.

The Danish twin registry

This registry was established in 1954 as the first nationwide twin registry in the world (Hauge 1981; Hauge et al. 1968). The birth registers from all 2,200 parishes of the relevant calendar years were manually scrutinized to identify all twin births. Through regional population registers (in operation since 1924) and other public sources, a search was made for the twins, or whenever needed, their closest relatives. As soon as a twin was traced, a questionnaire was sent to him or her. If neither of the partners was alive, a questionnaire was sent to the closest relative. Specific questions about the degree of similarity between the partners of a pair were included in the questionnaire to assess zygosity in like-sexed twins. For twins dying or emigrating at an early age it was impossible to obtain reliable data to be used in zygosity classification. Consequently, pairs were not followed-up if one or both partners died or emigrated before age 6. The validity of zygosity classification based on answers to mailed questionnaires has been evaluated by comparison with the results of later blood group determinants and the misclassification rate has been found to be less than 5% (Hauge 1981; Christiansen et al. 2003).

The Danish sample used in the present study consists of 4,890 pairs of like-sex twins for which both members survived 6 years of age. Twins born between 1870 and 1910 have been followed up through January 8th 2005, at which time their mortality status and hence lifespan was determined. The cohort is almost extinct (in 98% of pairs both twins are dead) and the youngest surviving twin was at least 94 years of age at last follow up.

The Swedish twin registry

The Swedish twin registry, which covers all twin births in Sweden between 1886 and 2002, was started in 1959, by contacting all parish offices in Sweden to obtain information on all twin births between 1886 and 1925. Through contacts with the parishes, twins' residences were followed through 1947, when the Swedish personal registration number (PNR) was initiated. Through parish records in 1947, PNR could be recorded and subsequently matched to the national population registry. All like-sexed pairs living in Sweden in 1960 were contacted by a postal questionnaire. Information on intrapair similarity (for subsequent assignment of zygosity) and smoking status was collected at this time. Only the information from pairs in which both responded was kept and recorded. Zygosity determination has been validated in several studies, some including DNA markers, and is 98% accurate (Lichtenstein et al. 2002). The register is updated for changes in vital status and addresses on a monthly basis. (Cederlöf 1966; Lichtenstein et al. 2002).

The sample of Swedish twins used in this study consisted of 4,694 pairs of like-sexed twins born during 1886–1910, where both members were alive and living in Sweden as of the 1st of March 1961. Hence twins from the youngest and oldest birthyears had to survive until the age of 51 and 75, respectively, and the sample thus does not contain early deaths. For this study, the cohort has been followed up through 14th of November 2003. Hence the minimum age for a twin who is still alive at the end of follow up is 92 years of age.

The Finnish Twin Cohort Study was started in 1974. All sets of persons born on the same day, in the same local community, of the same-sex and with the same surname at birth were identified to form the basis of the older Twin Cohort of twins born before 1958. A questionnaire mailed in 1975 to all pairs with both twins alive in 1974 was used to establish twinship and zygosity

(Kaprio et al. 1978), while further enquiries to local parish records and population registers was used to clarify whether subjects fulfilling the selection criteria were biological twins. This particular cohort of twins was subsequently contacted again in 1981 and 1990. Zygosity has been determined by a very accurate and validated deterministic algorithm (Sarna et al. 1978). Vital status and causes of death are updated by computer linkage using the unique personal identification numbers assigned in the 1960s and consequently at birth to all Finnish citizens and residents. Linkage is done with the Population Register Centre of Finland (http://www.vaestorekisterikeskus.fi) and Statistics Finland (Kaprio and Koskenvuo 2002).

The Finnish sample used in this study consists of 667 like-sexed pairs of twins born during 1880–1910, where both members were alive and living in Finland as of 1st of January 1974. Hence twins from the youngest and oldest birthyears had to survive until the age of 64 and 94, respectively in order to be included, and the sample thus does not contain early deaths.

For this study, the cohort is followed up through 1st of July 2003, so the minimum age for a twin who is still alive at the end of follow up is 92 years of age.

Analyses of twin similarity

The concept of age-specific genetic influence on the human lifespan is somewhat difficult to formalize, when compared to the question of age-specific genetic influences on a truly age-dependent phenotype, e.g. body mass index (BMI). In the latter case, a distinct phenotype is defined at any age (e.g. BMI at age 60), it can be measured and studied using the traditional methods of genetic analysis, yielding an age-trajectory of heritability of BMI. However, in the case of human longevity, there is only one phenotype for each individual: age at death, and it is therefore not as straightforward to define an age-specific measure of genetic influence on longevity, as for BMI. We remark that one may consider 'years of life after a certain age' as the phenotype and applying traditional methods to the sample truncated by a given age, i.e. by taking all pairs where both members have survived past that age. This procedure may be repeated for each chosen age cut-off. However, the estimation of heritability in remaining lifespan at each age of death (year) by successive application of the polygenic model to each stratum defined by truncation of lifespan may cause serious bias as pointed out in Martin and Wilson (1982) and in Iachine (2004).

In our analysis, we consider two approaches. The first is based on studying the life expectancy of MZ and DZ twins while conditioning on the co-twin lifespan in order to narrow the focus to certain age groups. In the second approach, we use the lifespan variable to define an age-dependent dichotomous phenotype (survival until a given age) and analyse it using the traditional methods of analysis for binary traits.

Conditional lifespan

We consider the analysis of regression of twin on co-twin lifespan: DeFries and Fulker (1985) have shown how twin similarity for a quantitative trait can be assessed by regressing the score of one twin on another. In general, the resulting regression coefficient is an estimate of the corresponding twin correlation under the assumption of symmetry between twin and co-twin (Rodgers and McGue 1994). We use a regression approach to assess MZ and DZ twin similarity for lifespan in the overall sample as well as in subsets of the sample defined by co-twin age at death. Pearson and Lawley (Aitken 1934) have shown that the regression coefficient is unaffected by selection on the independent variable if the overall regression is linear. Consequently, by conditioning on co-twin age at death we are able to determine whether twin similarity for age at death is constant or varies across the lifespan. Difference between regression slopes of MZ and DZ twins indicates the presence of genetic effects. Because of the sex difference in lifespan, all analyses are conducted separately in the male and female samples.

For this analysis, we use the almost extinct cohorts of Swedish, Danish and Finnish twins born between 1870 and 1910. To handle the differences in sampling procedures, we stratify the data into the two samples; the Danish twins surviving past age 6 and the combined cohort of Swedish and Finnish twins surviving respective dates of entrance. Expected lifespan conditional on co-twin lifespan is estimated for each sample. Estimates are obtained in two ways from pairs in which both members were dead at last follow-up (9,956 out of 10,251 pairs): as mean lifespan of twins over 5-year intervals of co-twin lifespan and as the smoothed expected lifespan of twins given lifespan of co-twin by the lowess-method with bandwidth parameter 0.8 (Chambers et al. 1983; Stata Statistical Software College Station 2005). Hence, we obtain graphs of expected lifespan of twin by lifespan of co-twin for MZ and DZ twins. To further quantify twin similarity conditional on co-twin age at death, we regress twin lifespan on co-twin lifespan within specified intervals of co-twin age at death (e.g. after age 15, before 60, 60+, 75+ and age 85+). Data were double-entered since every twin is also a co-twin. Due to within pair dependence, however, the robust variance estimator was used to take pair clustering into account (Rogers 1993). Inferences were carried out using standard methods (95% CI, two-sided t test).

Survival to given age

To investigate the presence of genetic influences on human lifespan, we also consider the dichotomous phenotype of surviving to a given age. We compare MZ and DZ twins with respect to relative recurrence risk and tetrachoric correlation in reaching a certain age. From estimates of such measures for each year

from 6 to 92 years of age for MZ and DZ twins, we obtain an indication of possible genetic influence at various stages in life, including survival at the oldest ages. Greater MZ than DZ similarity in the above measures suggests a genetic influence under the equal environment assumption of MZ and DZ pairs (Neale and Cardon 1992).

The prevalence and the relative recurrence risk, λ, of surviving up to a certain age defined as the ratio of the chance a twin reached a certain age given that the co-twin reached that age to the prevalence of reaching that age in the whole (sex-specific) twin population, are estimated using the maximum likelihood estimators given in for instance (Witte et al. 1999). We obtain an estimator of λ as the ratio of probandwise concordance rate to the prevalence estimates. Standard bootstrap methods (Efron and Tibshirani 1986) are used for the comparison of prevalence and probandwise concordance rate for MZ and DZ pairs for each age and for constructing 95% confidence intervals.

Differences in favour of MZ to DZ twins in above relative recurrence risks suggest the presence of genetic influences. For high prevalences, a small difference between MZ and DZ concordance rates does not necessarily imply little genetic influence. This motivates a supplemental measure of similarity. A measure of similarity that relates to the polygenic model of quantitative genetics is the tetrachoric correlation, i.e. the correlation in the bivariate normal distribution of (continuous) liability to survive to a given age (Neale and Cardon 1992). Briefly, the twin survives if the liability to survive to a given age exceeds a certain threshold. The tetrachoric correlation in surviving to a given age is then the correlation in twin liability. We estimate tetrachoric correlations for MZ and DZ pairs by standard maximum likelihood methods (Kohler and Rodgers 1999).

The trait of survival to a given age is estimated for two samples; First, the Danish cohort of twins born between 1870 and 1910 consisting of twins surviving 6 years of age and secondly for the Nordic cohort of Danish, Swedish and Finnish cohorts born during 1900–1910 in which both twins survived past 75 years of age ensuring identical and complete ascertainment in all three countries. Hence for the latter group who have already reached age 75, we study the presence of genetic influences on remaining survival to a given age from 75 to 92 years.

Analyses are carried out using the Stata statistical software (Efron and Tibshirani 1986; Stata Statistical Software College Station 2005). A significance level of 5% is used.

Results

The Swedish, Finnish and Danish twin cohorts born before 1910 altogether comprise 9,272 male twins and 11,230 female twins. Numbers of twin pairs by zygosity and gender are listed in Table 1 and ordered by pairs in which both survive, only one survived and neither survived up to a given age. Furthermore, numbers are listed for those who survived past age 6 (Danish cohort born 1870–1910) and for those who survived past age 75 (Swedish, Finnish and Danish cohort born 1900–1910).

Conditional lifespan

A graphical presentation of twin similarity for lifespan in the Danish sample is given in Fig. 1 and for the combined Swedish and Finnish sample in Fig. 2. The individual data points are the mean twin lifespans over 5-year intervals of co-twin age at death and the smoothed curves are obtained by lowess smoothing. The mean lifespan of twins increases with co-twin lifespan and this trend occurs more rapidly for MZ than for DZ twins when the co-twin exceeds approximately age 60 for both males and females. Before age 60 no systematic change in mean lifespan is observed for both MZ and DZ pairs, males and females (Fig. 1 and Table 2). Regressing twin lifespan on co-twin age at death for the Danish sample gives an overall regression coefficient of 0.15 in MZ males with 95% CI (0.08, 0.22) and 0.10 (0.04, 0.15) in DZ males; the comparable values in the

Table 1 Number of twin pairs by age (in years) survived past

Age	MZ male	DZ male	MZ female	DZ female
Danish pairs—both survived past age 6 years				
25+	813/42/4	1,386/119/2	823/52/5	1,512/127/5
75+	263/318/278	349/681/477	364/346/170	561/736/347
85+	54/197/608	67/357/1,083	116/287/477	167/580/897
Swedish, Danish, Finnish pairs—both survived past age 75 years				
75+	397/25/2	581/62/1	618/27/2	1,120/79/0
80+	219/151/54	290/279/75	449/156/42	754/382/63
85+	106/163/155	108/287/249	234/271/142	374/558/267
90+	30/109/285	28/169/447	93/218/336	103/439/657

Danish cohort born during 1870–1910, where both survived past age 6 years in the upper-half of the table. The combined Swedish, Danish and Finnish cohort born 1900–1910 and where both survived past age 75 years in the lower-half of the table. Numbers are listed in the order 'both survived/one survived/neither survived'

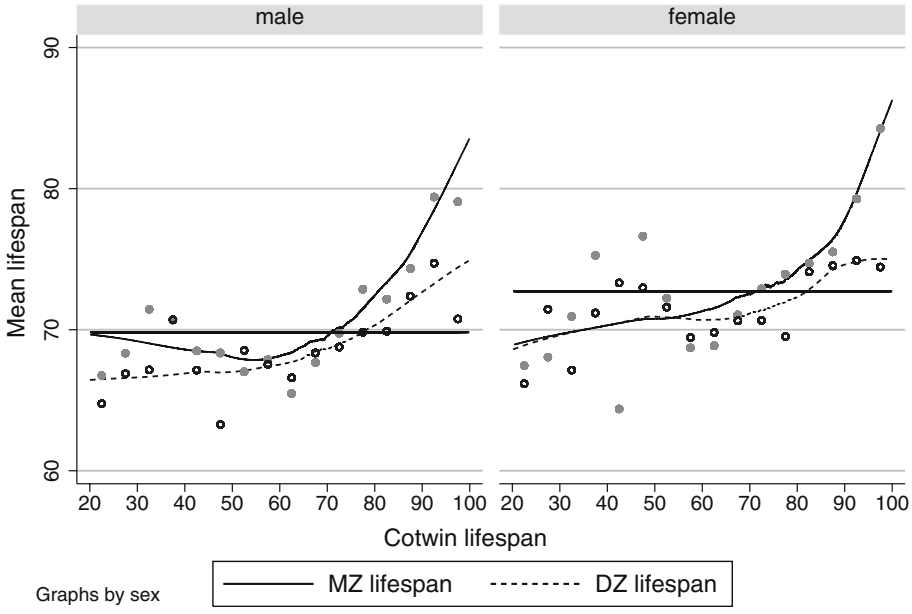

Fig. 1 Mean lifespan of twin given lifespan of co-twin of Danish twin cohort 1870–1910. *Dots* denote mean lifespan of twin given 5 year lifespan interval of co-twin (MZ twins by *solid circles*, DZ twins by *hollow circles*). The curves arise from lowess smoothing (bandwidth 0.8). *Horizontal lines* are mean lifespan for the whole sample (males and females, respectively)

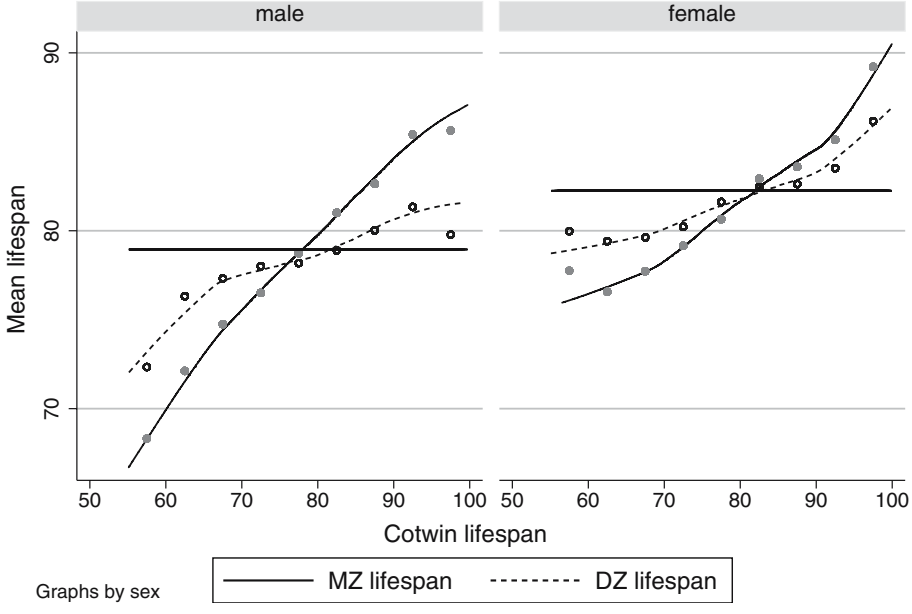

Fig. 2 Mean lifespan of twin given lifespan of co-twin of Swedish and Finnish joint twin cohort 1886–1910 excluding early deaths. *Dots* denote mean lifespan of twin given 5 year lifespan interval of co-twin (MZ twins by *solid circles*, DZ twins by *hollow circles*). The curves arise from lowess smoothing (bandwidth 0.8). *Horizontal lines* are mean lifespan for the whole sample (males and females, respectively)

Table 2 Regression slope of expected lifespan of twins conditional on lifespan interval of co-twin

Age	All (>15)	≤60	>60	>75	>85
Danish cohort 1870–1910					
MZM	0.15 (0.04) (851)	0.00 (0.10) (285)	0.39 (0.06) (817)	0.39 (0.11) (575)	0.71 (0.31) (245)
DZM	0.10 (0.03) (1,500)	−0.01 (0.05) (581)	0.21 (0.05) (1,418)	0.23 (0.09) (1,023)	0.00 (0.25) (417)
MZF	0.18 (0.04) (862)	0.00 (0.10) (248)	0.30 (0.06) (829)	0.36 (0.10) (692)	0.64 (0.25) (385)
DZF	0.08 (0.03) (1,607)	0.02 (0.06) (588)	0.19 (0.05) (1,526)	0.25 (0.08) (1,260)	0.04 (0.18) (710)
Swedish and Finnish joint cohort 1886–1910					
MZM	–	–	0.43 (0.03) (829)	0.41 (0.05) (716)	0.42 (0.11) (355)
DZM	–	–	0.15 (0.03) (1,380)	0.20 (0.04) (1,213)	0.20 (0.11) (565)
MZF	–	–	0.32 (0.03) (987)	0.34 (0.04) (918)	0.43 (0.08) (604)
DZF	–	–	0.17 (0.02) (1,930)	0.17 (0.03) (1,808)	0.30 (0.06) (1,228)

Danish cohort born between 1870 and 1910 in the upper-half and the combined Swedish and Finnish cohort born between 1886 and 1910 in the lower-half of the table (see the text for inclusion criteria). Numbers in entries are slope, standard error and number of pairs, respectively

female sample are 0.18 (0.10, 0.26) and 0.08 (0.03–0.13), respectively (see Table 2). These values are clearly consistent with a modest heritability as estimated in previous research. The relationship depicted in the figures, however, suggests that the magnitude of twin similarity for lifespan varies with age.

As Figs. 1 and 2 indicate the mean lifespan of twins tends to be proportional to co-twin lifespan when co-twin lifespan exceeds 60 years (to at least 90 years). By linear regression, the increase in mean lifespan for Danish male MZ pairs is estimated at 0.39 year per co-twin lifespan year with 95% CI (0.28, 0.50) when co-twin lifespan exceeds 60 years (see Table 2). For male DZ pairs, the rate is 0.21 (0.11, 0.30) and the difference between MZ and DZ rates is significant ($P < 0.01$). For Danish females, the increase in mean lifespan is 0.30 (0.19, 0.42) for MZ pairs and 0.19 (0.10, 0.29) year per co-twin lifespan year for DZ pairs when co-twin lifespan exceeds 60 years; however, the difference is not significant ($P = 0.14$) in this case. These differences remain or increase when co-twin lifespan exceeds older ages (75 and 85 years) as given in Table 2. The rates from the Danish sample with complete follow-up (i.e. survival past age 6) agree very well with those estimated in the combined Swedish and Finnish sample presented in Table 2, except possibly for the highest age group. The Swedish and Finnish sample shows a pattern of increase in mean lifespan with co-twin lifespan (after age 60) for MZ and DZ pairs that is consistent with a constant genetic influence on lifespan with age. For Swedish and Finnish male MZ pairs the rates are 0.43, 0.41 and 0.42 year per co-twin lifespan when co-twin lifespan exceeds age 60, 75 and 85, respectively, while for male DZ pairs the corresponding rates are 0.15, 0.20 and 0.20. Swedish and Finnish females show similar estimates (0.32, 0.34 and 0.43 for MZ and 0.17, 0.17 and 0.30 for DZ pairs when co-twin lifespan exceeds age 60, 75 and 85, respectively). Hence the difference between MZ and DZ slopes shown in Table 2 remains almost constant with increasing age and the differences are significant ($P < 0.001$) except when co-twin lifespan exceed 85 years (where *P*-values are 0.16 and 0.19 for males and females, respectively).

To sum up, Table 2 gives estimates of the regression coefficients that show (1) prior to age 60 (and after age 6) there is no indication of similarity in twin age at death; (2) From age 60 co-twin age at death is significantly predictive of twin lifespan. For MZ twins lifespan is increasing approximately 0.40 years in males and 0.35 years in females for every additional year of co-twin life from age 60 to at least age 85. For DZ twins, the increase in lifespan is approximately 0.20 years in males and 0.25 years in females for every additional year of co-twin life from age 60 to at least age 85. These data thus suggest minimal genetic effects on lifespans less than age 60 and moderate genetic effects on lifespans greater than age 60.

The above estimates, obtained from pairs born before 1910 in which both members were dead at last follow-up (9,956 out of 10,251 pairs), were practically unchanged when considering earlier, but extinct birth cohorts indicating that the results are less sensitive to the applied mild censoring concerning relatively few pairs. Furthermore, restricting the combined Swedish and Finnish sample to the 1900–1910 birthcohort did not affect results. The Danish sample with complete follow-up (i.e. survival past age 6) showed virtually unchanged estimates of regression slopes when the Swedish inclusion criteria were implemented on the data, except from age 85 (results not shown). This indicates relative robustness of slopes towards the truncation by date. From age 85, estimates of slope were very similar to those obtained from the combined Swedish and Finnish sample.

Survival to given age

Prevalence and relative recurrence risk for reaching a given age from age 6 to age 92 was estimated for the Danish cohort 1870–1910. The prevalence decreases with age as expected. There is no indication of systematic difference in prevalence for MZ and DZ pairs (results not shown). Estimates of the relative recurrence risk of surviving to a given age are presented in Table 3 (including inference results). The relative recurrence risk of surviving up to some age from age 6 to 60 is close to 1 for both

Table 3 Relative survival probabilities of twins given survived age of co-twin compared with the prevalence of survived age for twins, i.e. relative recurrence risks by age

Age	MZ male	DZ male	MZ female	DZ female
25+	1.00 (1.00, 1.01)	1.00 (1.00, 1.00)	1.00 (1.00, 1.01)	1.00 (1.00, 1.00)
50+	1.00 (0.99, 1.01)	1.01 (1.00, 1.02)	1.01 (1.00, 1.02)	1.00 (0.99, 1.01)
60+	1.01 (0.99, 1.03)	1.01 (0.99, 1.02)	1.02 (1.00, 1.03)	1.01 (1.00, 1.02)
70+	1.11 (1.07, 1.15)	1.05 (1.02, 1.09)	1.05 (1.02, 1.07)	1.02 (1.00, 1.05)
75+	1.27 (1.20, 1.34)	1.11 (1.05, 1.17)	1.11 (1.07, 1.15)	1.07 (1.03, 1.11)
80+	1.43 (1.27, 1.58)	1.23 (1.11, 1.35)	1.20 (1.12, 1.27)	1.18 (1.12, 1.24)
85+	1.99 (1.60, 2.39)	1.68 (1.37, 1.98)	1.52 (1.34, 1.69)	1.31 (1.18, 1.45)
90+	3.58 (2.12, 5.03)	1.88 (0.82, 2.95)	2.18 (1.68, 2.68)	1.43 (1.08, 1.78)
92+	4.83 (2.17, 7.49)	1.76 (0.07, 3.45)	2.50 (1.71, 3.29)	1.57 (1.03, 2.11)

Parentheses: 95% confidence intervals. Danish twin cohort born during 1870–1910 and survived age 6 years

MZ and DZ twins. For this age interval of relatively high prevalence genetic effects in surviving to these ages still may be present as noted above but not reflected in the relative recurrence risk measure. Starting from around age 60, the relative recurrence risk increases with increasing survived age for both male and female twins and is higher for MZ twins than DZ twins. Also, power for detecting this difference in recurrence risk increases at this age range as individuals die. Furthermore, the difference in relative recurrence risk between MZ twins and DZ twins in Table 3 increases with age from age 60 and onwards indicating survival advantage with age due to genetic effects in both males and females.

The differences between MZ and DZ pairs are significant ($P < 0.01$) at each age. It is noteworthy that the estimates of relative recurrence risk similar to those for males are found for females at a 5–10 year higher age range as seen from Table 3. For the highest age group, the relative risk of a female MZ twin reaching age 92 given that her co-twin reached this age is 2.5 (1.7–3.3) compared to the risk of reaching age 92 for female twins generally. For DZ female twins, the relative recurrence risk of reaching age 92 is 1.6 (1.03–2.1). For males, the relative recurrence risk for reaching age 92 it is 4.8 (2.2,7.5) for MZ and 1.8 (0.1,3.4) for DZ pairs.

Figure 3 presents the relative recurrence rate by age for the restricted cohort of those Nordic twins born during 1900–1910, where both survived past 75 years of age. The relative recurrence risk is 1 for both MZ and DZ pairs at age 75 and then the same pattern evolves as above of increasing differences in relative recurrence risk between MZ twins than DZ twins with increasing age for both males and females. This clearly indicates the presence of genetic influence at old ages.

For the Nordic elderly who already survived past 75 years of age presented in Fig. 3, MZs are significantly more correlated than DZs and the difference remains relatively constant with age for both males and females. This indicates a constant and significant heritability in liability to reaching a given age due to genetic effects of the total variance, i.e., the heritability, from 75 to 92 years of age for this selected group who already reached age 75. Moreover, this is compatible with the observed almost constant increase in difference between MZ and DZ relative recurrence rates shown in Fig. 3.

The results presented above persist when stratifying by country, i.e. the same patterns of change in the similarity measures for MZ and DZ pairs were observed for the Swedish, Finnish and Danish cohorts separately although differences in mortality rates exist among the three countries. Similarly, changing the birthcohort either by narrowing or widening the defining birthyear interval had no observed influence on the results.

Discussion

Based on a large population-based and almost exhaustive sample of twins more than 90 years of follow up, we find evidence of familial clustering of longevity. The present study is the first to demonstrate that at population level genetic variants for survival may exist with a pattern compatible with a significant and constant to increasing influence of genetic factors with age. That is, we find among elderly that the difference in mean lifespan of twins by co-twin lifespan is in favour of MZ twins relative to DZ twins and increases with age. Furthermore, our findings indicate a proportional change in lifespan by co-twin lifespan for both MZ and DZ pairs

Fig. 3 Twin pairs surviving past age 75 years only. Tetrachoric correlation (*left vertical axis*) and relative recurrence rate, RRR (*right vertical axis*) of twin pairs by survived age. Swedish, Finnish and Danish cohort born during 1900–1910 and survived past age 75 years

and that the rate of change is significantly higher for MZ than DZ pairs. These findings suggest minimal genetic effects on lifespans less than age 60 and moderate genetic effects on lifespans greater than age 60. Also, when considering the chance of surviving to a certain age, we observe a significantly stronger dependence on co-twin survival for MZ pairs than for DZ pairs among those aged 60 years or over.

During the last decade, a series of twin studies have shown that approximately 20–30% of the overall variation in lifespan is caused by genetic differences. This seems to be a rather consistent finding in various Nordic countries in different time periods and even so among other species not living in the wild (Finch and Tanzi 1997; Herskind et al. 1996; Ljungquist et al. 1998; Mousseau and Roff 1987). Although we do not estimate heritability as a function of age, our findings support the existence of such an influence at various stages in human lifespan, in particular among elderly (as seen from the significant difference in MZ and DZ dependence measures).

Two alternative views in gerontological research have led to opposite predictions regarding the importance of genetic factors with age. First, is the expectation that the accumulation of unique environmental exposures during a long life is the major determinant of lifespan and health at older ages (Harris et al. 1992), which predicts decreased heritability at older ages. Alternatively, evolutionary biologists have argued that the reduced selective pressure against deleterious genetic mutations expressed only late in life predicts an increase in genetic variance among the oldest (Partridge and Gems 2002).

Our findings suggest that genetic effects are important for survival at older ages. These effects may arise either through central pathways ('longevity enabling genes') or indirectly through genes associated with disease or states of certain mortality risk (Johnson 2005).

The validity of these findings assumes that environmental factors of importance for the trait act the same for MZ and DZ twins (the 'equal environments assumption'). Many such factors may be suspected (like social support, closeness in residence, etc.) that would inflate our estimates of genetic influence. However, the observed patterns in similarity of MZ and DZ pairs with age indicate a relatively constant advantage in survival from moment to moment and the shift of this pattern with age between sexes merely suggests a genetic origin of influences.

The interpretation of our findings depends on the fact that the genetic influence measures we have considered are cumulative in nature: The dichotomous phenotype of survival until a given age and the conditional expected longevity by definition are not completely restricted to genetic effects in a specific age group. The genetic influence demonstrated may not necessarily be of local age-specific nature, e.g. in the sense of increased expression of some deleterious or protective longevity genes at certain ages, but might also be due to genes reducing mortality in the younger age groups. This issue may be studied using models accounting for time-dependent gene expression patterns that would predict genetic influence at various ages. For developing such models of quantitative genetics, our estimates of (difference in) relative recurrence risk and correlation for MZ and DZ pairs may serve as a useful guideline, since results would need to be replicated or predicted by such a unified model.

In our study, we did not observe substantial differences in prevalence of surviving to a given age between zygosity groups for all ages. The pattern of increasing relative recurrence risks with age is very similar for males and females although with later onset for females, which corresponds to the better female survival. The Nordic cohorts of twins are homogeneous on many genetical and environmental factors and good agreement in the similarity measures was observed across cohorts (at ages where comparison is possible).

Clustering of late deaths in a few families with many extreme long-living individuals has provided support for a familial component. Perls and co-workers found that the risk ratio of survival for siblings of centenarians versus siblings of 73-year-old was about 4 for ages 80–94 (Perls et al. 1998; Perls and Dellara 2003). Kerber et al. (2001) also found, based on Mormon genealogies, an increased recurrence risk for siblings for surviving to extreme ages, although the estimate was somewhat lower than found by Perls. Gudmundsson et al. (2000) using the population-based genealogy in Iceland, found that the first-degree relatives of the probands who live to an extreme old age (the 95% percentile) are twice as likely as the controls to survive to the same age. It is noteworthy that our results on relative survival risk by age for DZ siblings (as seen in Table 3) are in good agreement with these findings regarding pattern of estimates for both genders [see for instance, Perls and Dellara (2003)]. Furthermore, since based on twin cohorts, which allows for better disentanglement of environmental and genetical origin of effects, our findings strongly support that genetic effects are among the mechanisms in surviving to higher ages.

There is probably considerable genetic heterogeneity for lifespan, and it is possible that in some families there is a considerable genetic component, but which accounts for only a fraction of the variance in the population at large. A similar scenario is found in breast cancer where twin studies (Lichtenstein et al. 2000) suggest a small overall genetic contribution, but where the genes *BRCA1* and *BRCA2* within some families confer a major risk (Narod and Foulkes 1994). Some evidence for decreasing genetic susceptibility to death from coronary heart disease with age is given in (Marenberg et al. 1994). It is, however, suggested in (Zdravkovic et al. 2002) that genetic factors to coronary heart disease mortality are in operation throughout the entire lifespan. We note that genetic factors associated with one single high risk disease may be less influential at older age groups.

There are few genetic association and linkage studies of extreme and healthy survival. Only one common polymorphism, namely Apo-E, has consistently been shown to be associated with survival until extreme ages (Gerdes et al. 2000). Puca et al. performed a genome-wide scan for candidate gene polymorphisms using 308 individuals belonging to 137 sibships demonstrating exceptional longevity. By using nonparametric analysis, borderline significant evidence for linkage was noted for chromosome 4 at D4S1564 (Puca et al. 2001). However, this finding could not be replicated in an independent French sample (Geesaman et al. 2003) and no association between the candidate gene microsomal triglyceride transfer protein (*MTP*) haplotype and longevity in humans is seen (Bathum et al. 2005; Nebel et al. 2005). Our results based on twin cohorts suggests that genetic variants may also be found in population-wide samples and not only in cohorts of extreme longlivers (Perls et al. 1998). Linkage studies in large samples of extreme long-lived siblings may be among the best approaches to identify such genes (Wright et al. 2003).

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