Title

Telomere length and health outcomes: A two-sample genetic instrumental variables analysis

Permalink

https://escholarship.org/uc/item/4s16g92m

Authors

Hamad, Rita
Walter, Stefan
Rehkopf, David H

Publication Date

2016-09-01

DOI

10.1016/j.exger.2016.06.005

Peer reviewed

Telomere Length and Health Outcomes: A Two-Sample Genetic Instrumental Variables Analysis

Rita Hamad[a,*], Stefan Walter[b], David H. Rehkopf[a]

[a]Stanford University, Department of Medicine, 1070 Arastradero Road, Palo Alto, California, 94304, USA

[b]University of California San Francisco, Department of Epidemiology & Biostatistics, 550 16th Street, San Francisco, California, 94158, USA

Abstract

Objective—Previous studies linking telomere length (TL) and health have been largely associational. We apply genetic instrumental variables (IV) analysis, also known as Mendelian randomization, to test the hypothesis that shorter TL leads to poorer health. This method reduces bias from reverse causation or confounding.

Methods—We used two approaches in this study that rely on two separate data sources: (1) individual-level data from the Health and Retirement Study (HRS) (N=3,734), and (2) coefficients from genome-wide association studies (GWAS). We employed two-sample genetic IV analyses, constructing a polygenic risk score (PRS) of TL-associated single nucleotide polymorphisms. The first approach examined the association of the PRS with nine individual health outcomes in HRS. The second approach took advantage of estimates available in GWAS databases to estimate the impact of TL on five health outcomes using an inverse variance-weighted meta-analytic technique.

Results—Using individual-level data, shorter TL was marginally statistically significantly associated with decreased risk of stroke and increased risk of heart disease. Using the meta-analytic approach, shorter TL was associated with increased risk of coronary artery disease (OR 1.02 per 100 base pairs, 95%CI: 1.00, 1.03).

Discussion—With the exception of a small contribution to heart disease, our findings suggest that TL may be a marker of disease rather than a cause. They also demonstrate the utility of the inverse variance-weighted meta-analytic approach when examining small effect sizes.

1. INTRODUCTION

Telomeres are DNA-protein structures that include a repeated nucleotide sequence at the ends of eukaryotic chromosomes, acting to protect the degradation of functional DNA

[*]Corresponding author: rhamad@stanford.edu.

sequences during cellular replication (Olovnikov 1973). Shortening of telomeres in human cells *in vitro* has been shown to lead to cellular dysfunction, senescence, and death (Allsopp and Harley 1995; Blackburn 2000). Shorter mean telomere length (TL) is associated with increased risk of mortality (Cawthon and others 2003; Kim and others 2012b; Rehkopf and others 2013), and a growing number of observational and longitudinal studies have also demonstrated links between shorter TL and human illnesses. For example, researchers have found associations between shorter TL and higher prevalence of coronary artery disease, Type 2 diabetes, Alzheimer's, and Parkinson's disease, and an association between shorter TL and vulnerability to acute infections (Brouilette and others 2007; Cohen and others 2013; Zhu and others 2011). Almost without exception, however, these studies have been conducted on small samples using observational methods, leaving researchers unable to determine whether TL is in fact a cause of illness or simply a marker of the disease process.

There are a number of reasons why observational studies of TL and chronic disease may not have accurately estimated the relationship. A growing literature suggests that lower socioeconomic status is associated with shorter telomeres (Cherkas and others 2006; Needham and others 2013; Robertson and others 2013). While this may suggest that TL is a pathway through which adverse social conditions get "under the skin," translating into vulnerability to disease and "host susceptibility" (Cassel 1976), it may also be an indication that the relationship between TL and health is confounded by environmental and social exposures. For example, many lifestyle factors – including smoking, exercise, and diet – have been associated with TL (Lin and others 2012; Mirabello and others 2009; Shammas 2011; Valdes and others). It may be the case that these factors or other physiological attributes of disease itself actually cause shortened TL rather than the reverse. An additional source of potential bias in observational studies of TL obtained from blood and buccal cells is that these samples are composed of different cell types, each of which have different mean TL (Lin and others 2010). Therefore differences in cell type composition due to differences in immune function or infection could confound findings in observational studies.

Given the limitations of observational studies, one promising avenue is genetic instrumental variables (IV) analysis, also known as Mendelian randomization. Genetic IV analyses are increasingly used in the field of epidemiology, enabling researchers to estimate causal relationships when randomization of the exposure is not feasible (Lawlor and others 2008). In the case of telomeres, prior research has identified numerous genetic markers – or single nucleotide polymorphisms (SNPs) – that are associated with longer telomeres. These SNPs undergo random assortment from parents to offspring, creating a "randomization" of the exposure, i.e., TL. Thus, while the effect of TL on health outcomes cannot be directly measured due to an inability on the part of researchers to randomly assign TL, investigators use a quasi-randomly assigned third variable that influences health through its effect on TL – i.e., the presence of SNPs that predict TL – to estimate the relationship between TL and health. This method has become increasingly applied to estimate the effects on health outcomes of risk factors that otherwise are not amenable to randomization (Klerk and others 2002; Theodoratou and others 2012; Wehby and others 2011).

In this study, we construct a polygenic risk score that incorporates seven TL-associated SNPs identified in a recent meta-analysis of genome-wide association studies (GWAS). We

then employ a two-sample genetic IV analysis to determine the causal effect of TL on a variety of health outcomes previously associated with TL. We first use data on individual-level outcomes, testing the hypothesis that differences in TL contribute to the development of human disease. This is a typical genetic IV approach that is often underpowered when effect sizes are small. Consequently, we also take advantage of coefficients from several GWAS databases, highlighting the utility of publicly available GWAS data to reach sufficient sample sizes to query phenotype-outcome relationships of potentially small magnitude in a meaningful way.

2. METHODS

2.1. Individual-Level Data Set

We used data from the U.S. Health and Retirement Study (HRS), a longitudinal panel study that has collected data biennially since 1992 among a representative sample of over 26,000 men and women over 50 years of age, with an over-sampling of older individuals. The survey also includes data on respondents' spouses, which includes individuals under 50 years of age. A description of the HRS, including survey design, can be found in previous studies (Juster and Suzman 1995). We restricted our analyses to individuals for whom we have data on both genotype and TL (N = 5,225). Because the samples in which the TL-associated SNPs were identified include only those of European descent, as described below, we further restricted our sample to include only HRS participants who self-reported as non-Hispanic white (N = 3,734).

Our primary predictor variable was mean TL, which was obtained in 2008 from HRS participants who consented to provide a salivary sample. Samples were analyzed by Telome Health using a standard quantitative polymerase chain reaction assay. TL was measured in standard fashion, using the telomere-to-single copy gene (T/S) ratio. This ratio is determined by comparing the telomere sequence copy number (T) with a single-copy gene copy number (S). The equation for conversion of the T/S ratio to TL varies by lab, and for this study was: base pairs = (T/S) * 2,400.

Genetic data were also collected from respondents during the 2008 study wave. Subjects provided DNA samples using a mouthwash technique. Genotyping was conducted by the National Institutes of Health Center for Inherited Disease Research using the Illumina Human Omni-2.5 Quad beadchip, which includes roughly 2.5 million SNPs.

Outcome variables included measures of multiple health outcomes. In each survey wave, respondents self-reported whether they had ever been diagnosed with the following health conditions: diabetes, hypertension, heart disease, lung disease, stroke, arthritis, psychiatric disease, and cancer. For each of these conditions, we constructed a binary variable indicating whether the individual ever reported being diagnosed in any survey wave (1992–2010). Respondents also self-reported their height and weight, allowing us to calculate their body mass index (BMI) for each survey wave. We created a binary variable indicating whether an individual ever met criteria for obesity (BMI greater than 30).

Covariates included age, gender, and educational attainment. We measured age at collection of the telomere assay in 2008. Educational attainment was constructed as a categorical variable with four levels: less than high school education (reference group), high school or GED completed, some college, and college completed. We also account for residual population stratification by including the first four genetic principal components-derived eigenvectors, a technique used in prior work (Chang and others 2014; Walter and others 2015).

2.2. GWAS Databases

The polygenic risk score we constructed, described below, was derived from a genome-wide meta-analysis that identified SNPs associated with TL (Codd and others 2013). It included 37,684 individuals, with replication of selected variants in 10,739 controls. Additionally, we used coefficients drawn from five GWAS databases that examine health outcomes similar to those included in HRS (Locke and others 2015; Ripke and others 2013; Schunkert and others 2011; Soranzo and others 2010; Stahl and others 2010). These are summarized in Table 1. Of note, several of these outcomes represent different constructs than those in HRS. For example, in HRS, respondents self-report a diagnosis of "heart disease," while actual cases of coronary artery disease have been identified in the CARDIoGRAM study. These should therefore not be interpreted as identical to the outcomes from the HRS analyses, but rather complementary.

2.3. Polygenic Risk Score

We constructed a polygenic risk score (PRS) based on seven SNPs found to be significantly associated with TL in the largest genome-wide meta-analysis to date (Codd and others 2013). We used information on these seven SNPs from each individual in HRS to create the PRS. Weights were assigned for each SNP using the number of base pairs of decrease in TL associated with each allele, as reported in the meta-analysis (Codd and others 2013). Importantly, a higher value of the PRS means that an individual is genetically predisposed to shorter telomeres. Prior research has suggested that analyses using a PRS may be more successful in predicting disease risk than use of individual genetic markers (Dudbridge 2013).

2.4. Data Analysis

2.4.1. Individual-level Data—We first examined the association between the PRS and the health outcomes of interest using individual-level HRS data, employing a two-sample instrumental variables approach. This technique is valuable in cases where there are concerns of weak instrument bias and insufficient power in the first stage of a typical IV analysis (Evans and Davey Smith 2015; Pierce and Burgess 2013). We conducted the first stage of the IV analysis in the HRS sample, regressing TL on the PRS and the covariates above, to evaluate the validity of the PRS in this sample. As expected, this F-statistic was small (Table 3), confirming the importance of the two-sample IV approach, although the coefficient was in the expected direction, with higher levels of the PRS associated with shorter TL.

Given that the first stage using individual-level data was likely underpowered, we next regressed each of the individual-level health outcomes on the PRS itself in separate logistic regressions, controlling for the covariates described above, and no longer relying on the first-stage estimates of predicted TL. In this case, the coefficient on the PRS is interpreted as the increased odds of each of the health outcomes of interest for each 100 base pair decrease in mean TL. Standard errors were clustered at the household level and robust to an unknown form of heteroscedasticity. This approach rests on the assumption that the separate samples employed in the first and second stages are drawn from the same population, leading us to restrict the HRS sample to individuals of self-identified non-Hispanic white race to match the samples of European ancestry included in the genome-wide meta-analysis from which the PRS coefficients were obtained. Another important assumption is that the first and second stage samples are independent. This assumption is met, as HRS data were not included in the GWAS samples.

2.4.2. GWAS Disease Data—Given the small effect sizes, using individual-level data is likely to be underpowered even with a two-sample IV approach. We therefore next employed an inverse-variance weighted meta-analytic approach to estimate the effect of TL on a subset of these health outcomes that were available from the GWAS databases described above in order to leverage the greater statistical power available in these larger samples. This approach has been described previously (Burgess and others 2013), and is increasingly employed in two-sample genetic IV analyses in the absence of individual-level data (Mukherjee and others ; Østergaard and others 2015). In this case, it allowed for the estimation of the effect of TL on the odds of a given binary health outcome given a difference of 100 base pairs in TL; or for continuous outcomes (hemoglobin A1c and BMI), this method gives the change in the outcome measure per 100 base pairs. As shown previously (Burgess and others 2015), the coefficient and standard error can be calculated as follows:

$$\hat{\beta} = \frac{\sum_{k=1}^{K} X_k Y_k \sigma_{Y_k}^{-2}}{\sum_{k=1}^{K} X_k^2 \sigma_{Y_k}^{-2}}$$

$$se(\hat{\beta}) = \sqrt{\frac{1}{\sum_{k=1}^{K} X_k^2 \sigma_{Y_k}^{-2}}}$$

Here, X_k is the GWAS-derived estimate of the association between each SNP k with TL, while Y_k is the GWAS-derived estimate of the association between each SNP and the outcome of interest with standard error σ_{Y_k}.

As above, this approach assumes that the first and second stage samples are independent. In this case, while it is possible that a given individual was recruited for multiple GWAS samples, this is unlikely. At worst, this would be limited to a handful of individuals, and would not present a serious violation of this assumption.

Data analysis was conducted using Stata 14 (College Station, Texas) and R 3.2.1. Analytic code to conduct the inverse-variance weighted analysis has been provided in prior publications (Pierce and Burgess 2013).

2.5. Ethics Approval

Ethics approval for the HRS was provided by the University of Michigan Health Services Institutional Review Board. Approval for this study was provided by Stanford University Institutional Review Board (protocol 25818).

3. RESULTS

3.1. Characteristics of HRS Sample

Almost 60% of study subjects were female, with an average age of 69.5 in 2008 (Table 2). Participants were diverse with respect to educational attainment. The prevalence for the health conditions under investigation ranged from 10.6% for stroke to 67.8% for arthritis.

3.2. Two-Sample IV Analysis: Individual-level Data

Using individual-level data in HRS, the first stage of the IV analysis demonstrated that a higher PRS is associated with a decrease in TL, as expected, although this was not statistically significant likely due to the smaller size of this sample relative to the GWAS (β = −43.0 base pairs per unit of PRS, 95%CI: −96.9, 10.9) (Table 3). As described above, this supports our use of the two-sample IV analysis. In the second stage, the PRS was not statistically significantly associated with any of the health outcomes under study (Table 4). The majority of the odds ratios were close to one. This suggests that TL does not have a causal effect on these measures of disease. An increase in TL of 100 base pairs was marginally associated with a decreased odds of stroke (OR 0.92, 95%CI: 0.83, 1.01) and an increased odds of heart disease (OR 1.06, 95%CI: 0.99, 1.13).

3.3. Two-Sample IV Analysis: GWAS Data

Using the inverse variance-weighted approach with coefficients from GWAS databases (Table 5), we found that a decrease in TL of 100 base pairs was associated with an increased odds of heart disease (OR 1.02, 95%CI: 1.003, 1.03). There were no other associations of a meaningful magnitude or that attained traditional statistical significance levels for the other health outcomes we examined.

4. DISCUSSION

Our study employs two-sample genetic IV analyses to examine the effect of TL on health using both individual-level data and meta-analytic approaches. Using a weighted polygenic risk score of TL-associated genetic markers as an instrument for TL, we find that shorter telomeres lead to a marginally significantly decreased odds or stroke and increased odds of heart disease in individual-level data. Given the small sample size of HRS, however, and the low F-statistic on the first stage of the IV analysis in this sample, it is likely that this type of analysis is underpowered in the setting of small effect sizes. This increases the chances of Type II error, i.e., failing to reject the null when it is not true. We therefore carried out

parallel analyses using an inverse variance-weighted meta-analytic approach with GWAS data, in which the finding for heart disease was confirmed. This speaks to the potential significance of TL as an actual determinant for heart disease, rather than only as a marker of disease process. Our novel methodological approach to the question provides a critical piece of evidence for the literature on understanding the potential impacts of TL on health. Our findings for heart disease were of a small magnitude, and our null results (with narrow confidence intervals) for other outcomes suggest that TL should primarily be viewed as a marker of disease processes, rather than a cause in itself.

Our findings on heart disease are consistent with a large body of observational studies in the extant literature, in which shortened telomeres have been extensively implicated in the development of heart disease (Brouilette and others 2007; Farzaneh-Far and others 2010; Fitzpatrick and others 2007; Fyhrquist and others 2013; Nilsson and others 2013; Samani and others 2001; Starr and others 2007). This finding has been replicated in studies using a variety of designs – including cross-sectional, longitudinal, and case-control – and in diverse populations, although prior studies employ largely correlational methodologies. A recent systematic review of observational studies also found a consistent relationship between shorter TL and higher risk of coronary heart disease (Haycock and others 2014). One prior study also documented an association between TL-associated SNPs and coronary artery disease (Codd and others 2013), although the authors did not conduct a two-stage analysis as we do here, to determine the effect of the SNPs that acts through TL itself. The two-sample IV technique we employ here suggests that this relationship may in fact be causal, although the meta-analysis approach precludes the ability to test the assumptions behind the IV model. It is not straightforward to compare this effect size with those of prior studies given the differences in analytic strategies and measurement of outcomes, but the magnitude of the association in the present study is among the smallest of those previously documented. Of note, we report odds ratios per 100 base pairs. To put this difference in TL in context, population-based studies have shown that TL differs by approximately 14 base pairs with each year of age (Needham and others 2015). This suggests that prior associations are biased in part by unobserved confounding or reverse causation, and may indicate that TL should be viewed as a marker rather than a determinant of disease.

While other studies have found that shorter telomeres are associated with other health outcomes that we examine here – including obesity, diabetes, hypertension, cancer, and arthritis – we are not able to confirm that there exists a causal relationship for these conditions (Codd and others 2013; Demissie and others 2006; Nai-chieh and others 2012; Pellatt and others 2013; Valdes and others ; Zhai and others 2006). Prior observational studies may suffer from confounding and reverse causation (i.e., the impacts of disease on TL), and thus a true causal link may not exist. Alternately, our null findings for these conditions may be a result of measurement error, in that HRS participants self-reported whether they had been diagnosed with each disease, or insufficient power in the individual-level analysis. Nevertheless, we replicated several of the null results using the meta-analytic approach, which is ideal for detecting small effect sizes. Unfortunately, GWAS databases are not available for the full range of health outcomes in HRS, so we are not able to confirm all of the findings using the meta-analytic approach.

Our results also suggest that there is no association between TL and psychiatric disease using individual-level data, with no association for depression specifically using the meta-analytic approach. Preliminary randomized studies have shown that meditation and stress reduction techniques may lead to increased telomerase activity – which is thought to promote telomere maintenance – and longer telomeres (Lavretsky and others 2013; Ornish and others 2013). This would suggest that improved mental health may bring about longer TL, while our study suggests that the reverse may not be true. Nevertheless, the primary limitation of the IV approach is that results represent a "local average treatment effect." In other words, our findings are that differences in TL *brought about by differences in genetic markers* do not lead to large differences in health outcomes. Our results therefore do not rule out the possibility that changes in TL brought about by changes in behaviors, social conditions, or other non-genetic factors may affect disease.

There are a few studies that have examined the relationship between TL-associated SNPs and health, rather than the association of telomeres with health. One study found no association between a TL-associated SNP – SIRT1 – and a variety of health outcomes, although this study was limited to a few hundred participants and only included a single SNP (Kim and others 2012a). Another study examined seven SNPs associated with TL, finding that several were associated with higher rates of cancer and autoimmune disease (Codd and others 2013). This analysis did not include other potential covariates to rule out confounding by genetic ancestry or socioeconomic position. For example, some sub-populations have different allele frequencies due to population stratification or disparities in social and environmental exposures, which would result in confounding because of different rates of disease (Hamad and others under review). Another study found that a single TL-associated SNP was associated with higher risks of heart disease and diabetes, but also failed to control for confounding by race or SEP (Maubaret and others 2013). None of these studies included a measure of TL, thus representing a reduced form estimate of the impact of the observed SNPs rather than a two-staged least-squares analysis of the impact of TL itself; in other words, these estimates provide only the association between the SNPs and the health outcomes, rather than the causal effect that acts through TL itself. Moreover, the effect size cannot be put into context with observational evidence obtained from studies relating TL to health. A fourth study involved a genetic IV analysis using nine TL-associated SNPs, and found no significant association with risk of diabetes (Nai-chieh and others 2012). This study did not consider other disease outcomes.

We were unable to conduct analyses to explore the relationship between TL and health among non-white participants, given that the overwhelming majority of GWAS databases are conducted among individuals of European ancestry. This highlights the need for further research to identify TL-associated SNPs among minority groups, and speaks to the larger gap in the literature on genetic studies among non-white populations (Bustamante and others 2011).

There are several avenues for future research to elaborate upon these findings. As additional TL-associated SNPs are identified and replicated in large samples, a more comprehensive PRS could be constructed that may result in a stronger genetic instrument. Further studies are also needed on the mechanisms through which social conditions may impact TL, and on

Author Manuscript

Author Manuscript

Author Manuscript

Author Manuscript

the biological mechanisms through which shortened telomeres may play a role in coronary artery disease.

This study has several limitations. First, the individual-level analyses may suffer from measurement error, in that health outcomes were self-reported by participants. It is unlikely to bias the results, but may contribute to the null findings in the HRS sample. Also, while HRS includes a representative and diverse sample of older adults, it is likely limited by survivorship bias and may not reflect the experiences of younger individuals. In other words, those with longer telomeres and a genetic propensity for longevity are over-represented. This form of censorship may result in collider bias, as participation in this study is conditional upon survival to age 50 (Boef and others 2015). Importantly, the meta-analytic approach does not suffer from survivorship bias to the same degree, as the GWAS samples are not limited to older individuals. Nevertheless, future individual-level studies should attempt to replicate these results in younger samples. The limited sample size in HRS also limits our ability to detect small effect sizes in this sample, increasing the chances of Type II error, although a primary strength of this study is the use of the meta-analytic approach that overcomes these limitations of the individual-level data.

Our study employs a two-sample genetic IV analysis to examine whether TL is causally related to human health. It improves upon findings from prior observational studies in its implementation of an IV approach that limits confounding by unobserved factors and precludes reverse causation. Our results are among the first to suggest that TL makes a small causal contribution to heart disease, but not other chronic disease outcomes. This represents a major advance in understanding the role of TL in human health.

Acknowledgments

ABBREVIATIONS

BMI	body mass index
GWAS	genome-wide association studies
HRS	Health and Retirement Study
IV	instrumental variable
PRS	polygenic risk score
SNP	single nucleotide polymorphism
TL	telomere length

REFERENCES

Allsopp RC, Harley CB. Evidence for a critical telomere length in senescent human fibroblasts. Exp Cell Res. 1995; 219: 130–136. [PubMed: 7628529]

Blackburn EH. Telomere states and cell fates. Nature. 2000; 408: 53–56. [PubMed: 11081503]

Boef AGC, le Cessie S, Dekkers OM. Mendelian Randomization Studies in the Elderly. Epidemiology. 2015; 26: e15–e16. [PubMed: 25643110]

Brouilette SW, Moore JS, McMahon AD, Thompson JR, Ford I, Shepherd J, Packard CJ, Samani NJ. Telomere length, risk of coronary heart disease, and statin treatment in the West of Scotland Primary Prevention Study: a nested case-control study. The Lancet. 2007; 369: 107–114.

Burgess S, Butterworth A, Thompson SG. Mendelian randomization analysis with multiple genetic variants using summarized data. Genet Epidemiol. 2013; 37: 658–665. [PubMed: 24114802]

Burgess S, Scott RA, Timpson NJ, Davey Smith G, Thompson SG. Using published data in Mendelian randomization: a blueprint for efficient identification of causal risk factors. Eur J Epidemiol. 2015; 30: 543–552. [PubMed: 25773750]

Bustamante CD, De La Vega FM, Burchard EG. Genomics for the world. Nature. 2011; 475: 163–165. [PubMed: 21753830]

Cassel J. The contribution of the social environment to host resistance. Am J Epidemiol. 1976; 104: 107–123. [PubMed: 782233]

Cawthon RM, Smith KR, O'Brien E, Sivatchenko A, Kerber RA. Association between telomere length in blood and mortality in people aged 60 years or older. The Lancet. 2003; 361: 393–395.

Chang SC, Glymour MM, Walter S, Liang L, Koenen KC, Tchetgen EJ, Cornelis MC, Kawachi I, Rimm E, Kubzansky LD. Genome-wide polygenic scoring for a 14-year long-term average depression phenotype. Brain and behavior. 2014; 4: 298–311. [PubMed: 24683521]

Cherkas LF, Aviv A, Valdes AM, Hunkin JL, Gardner JP, Surdulescu GL, Kimura M, Spector TD. The effects of social status on biological aging as measured by white-blood-cell telomere length. Aging cell. 2006; 5: 361–365. [PubMed: 16856882]

Codd V, Nelson CP, Albrecht E, Mangino M, Deelen J, Buxton JL, Hottenga JJ, Fischer K, Esko T, Surakka I. Identification of seven loci affecting mean telomere length and their association with disease. Nat Genet. 2013; 45: 422–427. [PubMed: 23535734]

Cohen S, Janicki-Deverts D, Turner RB, Casselbrant ML, Li-Korotky H-S, Epel ES, Doyle WJ. Association Between Telomere Length and Experimentally Induced Upper Respiratory Viral Infection in Healthy AdultsTelomere Length and Respiratory Viral Infection. JAMA. 2013; 309: 699–705. [PubMed: 23423415]

Demissie S, Levy D, Benjamin EJ, Cupples LA, Gardner JP, Herbert A, Kimura M, Larson MG, Meigs JB, Keaney JF, Aviv A. Insulin resistance, oxidative stress, hypertension, and leukocyte telomere length in men from the Framingham Heart Study. Aging Cell. 2006; 5: 325–330. [PubMed: 16913878]

Dudbridge F. Power and Predictive Accuracy of Polygenic Risk Scores. PLoS Genet. 2013; 9: e1003348. [PubMed: 23555274]

Evans DM, Davey Smith G. Mendelian Randomization: New Applications in the Coming Age of Hypothesis-Free Causality. Annual Review of Genomics and Human Genetics. 2015; 16

Farzaneh-Far R, Lin J, Epel E, Lapham K, Blackburn E, Whooley MA. Telomere Length Trajectory and Its Determinants in Persons with Coronary Artery Disease: Longitudinal Findings from the Heart and Soul Study. PLoS One. 2010; 5: e8612. [PubMed: 20072607]

Fitzpatrick AL, Kronmal RA, Gardner JP, Psaty BM, Jenny NS, Tracy RP, Walston J, Kimura M, Aviv A. Leukocyte Telomere Length and Cardiovascular Disease in the Cardiovascular Health Study. Am J Epidemiol. 2007; 165: 14–21. [PubMed: 17043079]

Fyhrquist F, Saijonmaa O, Strandberg T. The roles of senescence and telomere shortening in cardiovascular disease. Nature Reviews Cardiology. 2013; 10: 274–283. [PubMed: 23478256]

Hamad R, Tuljapurkar S, Rehkopf D. Racial and Socioeconomic Variation in Genetic Markers of Telomere Length.

Haycock PC, Heydon EE, Kaptoge S, Butterworth AS, Thompson A, Willeit P. Leucocyte telomere length and risk of cardiovascular disease: systematic review and meta-analysis. BMJ. 2014; 349: g4227. [PubMed: 25006006]

Juster FT, Suzman R. An Overview of the Health and Retirement Study. The Journal of Human Resources. 1995; 30: S7–S56.

Kim S, Bi X, Czarny-Ratajczak M, Dai J, Welsh DA, Myers L, Welsch MA, Cherry KE, Arnold J, Poon LW. Telomere maintenance genes SIRT1 and XRCC6 impact age-related decline in telomere length but only SIRT1 is associated with human longevity. Biogerontology. 2012a; 13: 119–131. [PubMed: 21972126]

Kim S, Bi X, Czarny-Ratajczak M, Dai J, Welsh DA, Myers L, Welsch MA, Cherry KE, Arnold J, Poon LW, Jazwinski SM. Telomere maintenance genes SIRT1 and XRCC6 impact age-related decline in telomere length but only SIRT1 is associated with human longevity. Biogerontology. 2012b; 13: 119–131. [PubMed: 21972126]

Klerk M, Verhoef P, Clarke R, Blom HJ, Kok FJ, Schouten EG. MTHFR Studies Collaboration Group. MTHFR 677C→T polymorphism and risk of coronary heart disease: a meta-analysis. JAMA. 2002; 288: 2023–2031. [PubMed: 12387655]

Lavretsky H, Epel E, Siddarth P, Nazarian N, Cyr NS, Khalsa D, Lin J, Blackburn E, Irwin M. A pilot study of yogic meditation for family dementia caregivers with depressive symptoms: effects on mental health, cognition, and telomerase activity. International journal of geriatric psychiatry. 2013; 28: 57–65. [PubMed: 22407663]

Lawlor DA, Harbord RM, Sterne JA, Timpson N, Davey Smith G. Mendelian randomization: using genes as instruments for making causal inferences in epidemiology. Stat Med. 2008; 27: 1133–1163. [PubMed: 17886233]

Lin J, Epel E, Blackburn E. Telomeres and lifestyle factors: Roles in cellular aging. Mutation Research/Fundamental and Molecular Mechanisms of Mutagenesis. 2012; 730: 85–89. [PubMed: 21878343]

Lin J, Epel E, Cheon J, Kroenke C, Sinclair E, Bigos M, Wolkowitz O, Mellon S, Blackburn E. Analyses and comparisons of telomerase activity and telomere length in human T and B cells: insights for epidemiology of telomere maintenance. J Immunol Methods. 2010; 352: 71–80. [PubMed: 19837074]

Locke AE, Kahali B, Berndt SI, Justice AE, Pers TH, Day FR, Powell C, Vedantam S, Buchkovich ML, Yang J. Genetic studies of body mass index yield new insights for obesity biology. Nature. 2015; 518: 197–206. [PubMed: 25673413]

Maubaret CG, Salpea KD, Romanoski CE, Folkersen L, Cooper JA, Stephanou C, Wah Li K, Palmen J, Hamsten A, Neil A, Stephens JW, Lusis AJ, Eriksson P, Talmud PJ, Humphries SE. Association of TERC and OBFC1 Haplotypes with Mean Leukocyte Telomere Length and Risk for Coronary Heart Disease. PLoS One. 2013; 8: e83122. [PubMed: 24349443]

Mirabello L, Huang W-Y, Wong JYY, Chatterjee N, Reding D, David Crawford E, De Vivo I, Hayes RB, Savage SA. The association between leukocyte telomere length and cigarette smoking, dietary and physical variables, and risk of prostate cancer. Aging Cell. 2009; 8: 405–413. [PubMed: 19493248]

Mukherjee S, Walter S, Kauwe JSK, Saykin AJ, Bennett DA, Larson EB, Crane PK, Glymour MM. Genetically predicted body mass index and Alzheimer's disease–related phenotypes in three large samples: Mendelian randomization analyses. Alzheimer's & Dementia.

Nai-chieh Y, Chen BH, Song Y, Lu X, Chen Y, Manson JE, Kang M, Howard BV, Margolis KL, Curb JD. A prospective study of leukocyte telomere length and risk of type 2 diabetes in postmenopausal women. Diabetes. 2012; 61: 2998–3004. [PubMed: 22829448]

Needham BL, Adler N, Gregorich S, Rehkopf D, Lin J, Blackburn EH, Epel ES. Socioeconomic status, health behavior, and leukocyte telomere length in the National Health and Nutrition Examination Survey, 1999–2002. Social Science & Medicine. 2013; 85: 1–8. [PubMed: 23540359]

Needham BL, Rehkopf D, Adler N, Gregorich S, Lin J, Blackburn EH, Epel ES. Leukocyte Telomere Length and Mortality in the National Health and Nutrition Examination Survey, 1999–2002. Epidemiology. 2015. 30. [PubMed: 25286049]

Nilsson PM, Tufvesson H, Leosdottir M, Melander O. Telomeres and cardiovascular disease risk: an update 2013. Translational Research. 2013; 162: 371–380. [PubMed: 23748031]

Olovnikov AM. A theory of marginotomy: the incomplete copying of template margin in enzymic synthesis of polynucleotides and biological significance of the phenomenon. Journal of theoretical biology. 1973; 41: 181–190. [PubMed: 4754905]

Ornish D, Lin J, Chan JM, Epel E, Kemp C, Weidner G, Marlin R, Frenda SJ, Magbanua MJM, Daubenmier J. Effect of comprehensive lifestyle changes on telomerase activity and telomere length in men with biopsy-proven low-risk prostate cancer: 5-year follow-up of a descriptive pilot study. The lancet oncology. 2013; 14: 1112–1120. [PubMed: 24051140]

Østergaard SD, Mukherjee S, Sharp SJ, Proitsi P, Lotta LA, Day F, Perry JR, Boehme KL, Walter S, Kauwe JS. Associations between Potentially Modifiable Risk Factors and Alzheimer Disease: A Mendelian Randomization Study. PLoS Med. 2015; 12: e1001841. [PubMed: 26079503]

Pellatt AJ, Wolff RK, Torres-Mejia G, John EM, Herrick JS, Lundgreen A, Baumgartner KB, Giuliano AR, Hines LM, Fejerman L, Cawthon R, Slattery ML. Telomere length, telomere-related genes, and breast cancer risk: the breast cancer health disparities study. Genes Chromosomes Cancer. 2013; 52: 595–609. [PubMed: 23629941]

Pierce BL, Burgess S. Efficient Design for Mendelian Randomization Studies: Subsample and 2-Sample Instrumental Variable Estimators. Am J Epidemiol. 2013.

Rehkopf DH, Dow WH, Rosero-Bixby L, Lin J, Epel ES, Blackburn EH. Longer leukocyte telomere length in Costa Rica's Nicoya Peninsula: A population-based study. Exp Gerontol. 2013; 48: 1266–1273. [PubMed: 23988653]

Ripke S, Wray NR, Lewis CM, Hamilton SP, Weissman MM, Breen G, Byrne EM, Blackwood DH, Boomsma DI, Cichon S, Heath AC, Holsboer F, Lucae S, Madden PA, Martin NG, McGuffin P, Muglia P, Noethen MM, Penninx BP, Pergadia ML, Potash JB, Rietschel M, Lin D, Muller-Myhsok B, Shi J, Steinberg S, Grabe HJ, Lichtenstein P, Magnusson P, Perlis RH, Preisig M, Smoller JW, Stefansson K, Uher R, Kutalik Z, Tansey KE, Teumer A, Viktorin A, Barnes MR, Bettecken T, Binder EB, Breuer R, Castro VM, Churchill SE, Coryell WH, Craddock N, Craig IW, Czamara D, De Geus EJ, Degenhardt F, Farmer AE, Fava M, Frank J, Gainer VS, Gallagher PJ, Gordon SD, Goryachev S, Gross M, Guipponi M, Henders AK, Herms S, Hickie IB, Hoefels S, Hoogendijk W, Hottenga JJ, Iosifescu DV, Ising M, Jones I, Jones L, Jung-Ying T, Knowles JA, Kohane IS, Kohli MA, Korszun A, Landen M, Lawson WB, Lewis G, Macintyre D, Maier W, Mattheisen M, McGrath PJ, McIntosh A, McLean A, Middeldorp CM, Middleton L, Montgomery GM, Murphy SN, Nauck M, Nolen WA, Nyholt DR, O'Donovan M, Oskarsson H, Pedersen N, Scheftner WA, Schulz A, Schulze TG, Shyn SI, Sigurdsson E, Slager SL, Smit JH, Stefansson H, Steffens M, Thorgeirsson T, Tozzi F, Treutlein J, Uhr M, van den Oord EJ, Van Grootheest G, Volzke H, Weilburg JB, Willemsen G, Zitman FG, Neale B, Daly M, Levinson DF, Sullivan PF. A mega-analysis of genome-wide association studies for major depressive disorder. Mol Psychiatry. 2013; 18: 497–511. [PubMed: 22472876]

Robertson T, Batty GD, Der G, Fenton C, Shiels PG, Benzeval M. Is socioeconomic status associated with biological aging as measured by telomere length? Epidemiologic reviews. 2013; 35: 98–111. [PubMed: 23258416]

Samani NJ, Boultby R, Butler R, Thompson JR, Goodall AH. Telomere shortening in atherosclerosis. The Lancet. 2001; 358: 472–473.

Schunkert H, König IR, Kathiresan S, Reilly MP, Assimes TL, Holm H, Preuss M, Stewart AF, Barbalic M, Gieger C. Large-scale association analysis identifies 13 new susceptibility loci for coronary artery disease. Nat Genet. 2011; 43: 333–338. [PubMed: 21378990]

Shammas MA. Telomeres, lifestyle, cancer, and aging. Curr Opin Clin Nutr Metab Care. 2011; 14: 28–34. [PubMed: 21102320]

Soranzo N, Sanna S, Wheeler E, Gieger C, Radke D, Dupuis J, Bouatia-Naji N, Langenberg C, Prokopenko I, Stolerman E, Sandhu MS, Heeney MM, Devaney JM, Reilly MP, Ricketts SL, Stewart AFR, Voight BF, Willenborg C, Wright B, Altshuler D, Arking D, Balkau B, Barnes D, Boerwinkle E, Böhm B, Bonnefond A, Bonnycastle LL, Boomsma DI, Bornstein SR, Böttcher Y, Bumpstead S, Burnett-Miller MS, Campbell H, Cao A, Chambers J, Clark R, Collins FS, Coresh J, de Geus EJC, Dei M, Deloukas P, Döring A, Egan JM, Elosua R, Ferrucci L, Forouhi N, Fox CS, Franklin C, Franzosi MG, Gallina S, Goel A, Graessler J, Grallert H, Greinacher A, Hadley

D, Hall A, Hamsten A, Hayward C, Heath S, Herder C, Homuth G, Hottenga J-J, Hunter-Merrill R, Illig T, Jackson AU, Jula A, Kleber M, Knouff CW, Kong A, Kooner J, Köttgen A, Kovacs P, Krohn K, Kühnel B, Kuusisto J, Laakso M, Lathrop M, Lecoeur C, Li M, Li M, Loos RJF, Luan Ja, Lyssenko V, Mägi R, Magnusson PKE, Mälarstig A, Mangino M, Martínez-Larrad MT, März W, McArdle WL, McPherson R, Meisinger C, Meitinger T, Melander O, Mohlke KL, Mooser VE, Morken MA, Narisu N, Nathan DM, Nauck M, O'Donnell C, Oexle K, Olla N, Pankow JS, Payne F, Peden JF, Pedersen NL, Peltonen L, Perola M, Polasek O, Porcu E, Rader DJ, Rathmann W, Ripatti S, Rocheleau G, Roden M, Rudan I, Salomaa V, Saxena R, Schlessinger D, Schunkert H, Schwarz P, Seedorf U, Selvin E, Serrano-Ríos M, Shrader P, Silveira A, Siscovick D, Song K, Spector TD, Stefansson K, Steinthorsdottir V, Strachan DP, Strawbridge R, Stumvoll M, Surakka I, Swift AJ, Tanaka T, Teumer A, Thorleifsson G, Thorsteinsdottir U, Tönjes A, Usala G, Vitart V, Völzke H, Wallaschofski H, Waterworth DM, Watkins H, Wichmann HE, Wild SH, Willemsen G, Williams GH, Wilson JF, Winkelmann J, Wright AF, Wtccc, Zabena C, Zhao JH, Epstein SE, Erdmann J, Hakonarson HH, Kathiresan S, Khaw K-T, Roberts R, Samani NJ, Fleming MD, Sladek R, Abecasis G, Boehnke M, Froguel P, Groop L, McCarthy MI, Kao WHL, Florez JC, Uda M, Wareham NJ, Barroso I, Meigs JB. Common variants at 10 genomic loci influence hemoglobin $A_1(C)$ levels via glycemic and nonglycemic pathways. Diabetes. 2010; 59: 3229–3239. [PubMed: 20858683]

Stahl EA, Raychaudhuri S, Remmers EF, Xie G, Eyre S, Thomson BP, Li Y, Kurreeman FA, Zhernakova A, Hinks A. Genome-wide association study meta-analysis identifies seven new rheumatoid arthritis risk loci. Nat Genet. 2010; 42: 508–514. [PubMed: 20453842]

Starr JM, McGurn B, Harris SE, Whalley LJ, Deary IJ, Shiels PG. Association between telomere length and heart disease in a narrow age cohort of older people. Exp Gerontol. 2007; 42: 571–573. [PubMed: 17267157]

Theodoratou E, Palmer T, Zgaga L, Farrington SM, McKeigue P, Din FV, Tenesa A, Davey-Smith G, Dunlop MG, Campbell H. Instrumental variable estimation of the causal effect of plasma 25-hydroxy-vitamin D on colorectal cancer risk: a mendelian randomization analysis. PLoS One. 2012; 7: e37662. [PubMed: 22701574]

Valdes AM, Andrew T, Gardner JP, Kimura M, Oelsner E, Cherkas LF, Aviv A, Spector TD. Obesity, cigarette smoking, and telomere length in women. The Lancet. 366: 662–664.

Walter S, Glymour M, Koenen K, Liang L, Tchetgen Tchetgen E, Cornelis M, Chang S-C, Rewak M, Rimm E, Kawachi I. Do genetic risk scores for body mass index predict risk of phobic anxiety? Evidence for a shared genetic risk factor. Psychol Med. 2015; 45: 181–191. [PubMed: 25065638]

Wehby GL, Fletcher JM, Lehrer SF, Moreno LM, Murray JC, Wilcox A, Lie RT. A genetic instrumental variables analysis of the effects of prenatal smoking on birth weight: evidence from two samples. Biodemography Soc Biol. 2011; 57: 3–32. [PubMed: 21845925]

Zhai G, Aviv A, Hunter DJ, Hart DJ, Gardner JP, Kimura M, Lu X, Valdes AM, Spector TD. Reduction of leucocyte telomere length in radiographic hand osteoarthritis: a population-based study. Annals of the Rheumatic Diseases. 2006; 65: 1444–1448. [PubMed: 17038452]

Zhu H, Belcher M, van der HARST P. Healthy aging and disease: role for telomere biology? Clinical Science. 2011; 120: 427–440. [PubMed: 21271986]

HIGHLIGHTS

- Telomere length is associated with illness and mortality in observational studies.

- We apply Mendelian randomization to address confounding and reverse causation.

- Shorter telomeres raise the risk of heart disease with no effect for other diseases.

- Telomere length may be a marker of disease rather than a cause.

Table 1

Genome-wide Association Studies Included in Two-Sample Instrumental Variables Analyses

Study	Cases (N)	Controls (N)	Outcome of Interest	Data Repository Website
MAGIC	N/A	46,368	Hemoglobin A1c	http://www.magicinvestigators.org/
Stahl et al. (2010)	5,500	20,000	Rheumatoid arthritis	http://www.broadinstitute.org/ftp/pub/rheumatoid_arthritis/Stahl_etal_2010NG/
CARDIoGRAMplusC4D	22,233	64,762	Coronary artery disease	http://www.cardiogramplusc4d.org/
Psychiatric Genomics Consortium	18,478	17,558	Depression	http://www.med.unc.edu/pgc/downloads
GIANT Consortium	N/A	339,224	Body mass index	http://www.broadinstitute.org/collaboration/giant/index.php/GIANT_consortium_data_files

Table 2

Sample Characteristics, Health and Retirement Study

Demographics	
Female (%)	58.2
Education (%)	
Less than high school	13.3
High school / GED	38.3
Some college	23.8
College or more	24.6
Age in 2008	
Mean ± SD	69.5 ± 10.2
Median (IQR)	70 (62, 77)

Disease Diagnoses (%)	
Diabetes	20.5
Hypertension	61.4
Stroke	10.6
Arthritis	67.8
Heart disease	31.9
Lung disease	13.7
Psychiatric Illness	19.0
Cancer	19.7
Obesity	39.2

Genetic Characteristics	
Weighted polygenic risk score	
Mean ± SD	4.61 ± 1.08
Median (IQR)	4.66 (3.92, 5.33)
Telomere length in base pairs	
Mean ± SD	3166 ± 1248
Median (IQR)	3020 (2658, 3482)

N = 3,734. Only non-Hispanic white study participants with genetic and telomere data are included. For obesity, an individual is counted as a case if she ever had a body mass index greater than 30 based on self-reported height and weight. For other disease outcomes, an individual is counted as a case if she ever self-reported being diagnosed with that disease by a doctor.

Table 3

Association between Polygenic Risk Score and Telomere Length, First Stage of Instrumental Variable Analysis.

	β Coefficient [95% CI]
Polygenic risk score	−43.00 [−96.94, 10.93]
Sex (ref Male)	
Female	23.79 [−71.02, 118.61]
Education (ref <HS)	
High school	125.34[*] [9.40, 241.28]
Some college	135.63[*] [31.39, 239.88]
College	106.13[*] [9.64, 202.62]
Age	−7.08[**] [−11.70, −2.47]
Constant	3,766[**] [3,338, 4,194]
F-statistic	0.48

Note:

[*] p < 0.05,

[**] p < 0.01.

N = 3,734. Analyses conducted using individual-level data from the Health and Retirement Study. Standard errors are clustered by household. Additional covariates include four principal components-derived measures of genetic ancestry.

Table 4

Two-Sample Instrumental Variables Analyses of Impact of Telomere Length on Health, with Individual-Level Data from Health and Retirement Study

	Odds Ratio [95% CI]								
	Diabetes	Arthritis	Stroke	Heart Disease	Lung Disease	HTN	Psychiatric Disease	Cancer	Obesity
TL (per 100bp difference)	1.00 [0.93, 1.08]	1.00 [0.93, 1.06]	0.92 [0.83, 1.01]	1.06 [0.99, 1.13]	0.95 [0.87, 1.04]	0.96 [0.89, 1.04]	1.05 [0.97, 1.13]	0.96 [0.90, 1.03]	1.01 [0.95, 1.07]
Sex (ref Male)									
Female	0.58** [0.49, 0.68]	1.71** [1.48, 1.96]	0.63** [0.51, 0.78]	0.58** [0.50, 0.67]	0.95 [0.78, 1.15]	0.83* [0.70, 0.98]	1.70** [1.43, 2.02]	0.94 [0.83, 1.08]	0.85* [0.74, 0.98]
Education (ref <HS)									
High school	0.74* [0.59, 0.94]	0.71** [0.56, 0.91]	0.98 [0.72, 1.34]	0.89 [0.71, 1.11]	0.63** [0.49, 0.82]	1.09 [0.84, 1.41]	0.58** [0.45, 0.74]	0.89 [0.72, 1.11]	0.86 [0.69, 1.07]
Some college	0.65** [0.50, 0.84]	0.72* [0.55, 0.93]	0.73 [0.52, 1.04]	0.83 [0.65, 1.05]	0.51** [0.38, 0.68]	1.04 [0.78, 1.38]	0.51** [0.39, 0.68]	0.87 [0.69, 1.10]	0.83 [0.65, 1.06]
College	0.46** [0.35, 0.60]	0.49** [0.38, 0.63]	0.68* [0.48, 0.97]	0.61** [0.48, 0.78]	0.29** [0.21, 0.41]	1.15 [0.86, 1.52]	0.50** [0.38, 0.67]	0.54** [0.42, 0.68]	0.65** [0.52, 0.83]
Age	1.01 [1.00, 1.01]	1.04** [1.03, 1.05]	1.06** [1.05, 1.07]	1.05** [1.04, 1.05]	1.02** [1.01, 1.03]	1.04** [1.04, 1.05]	0.97** [0.96, 0.98]	0.97** [0.96, 0.98]	1.03** [1.03, 1.04]
Constant	0.43* [0.21, 0.89]	0.15** [0.075, 0.31]	0.0025** [0.00082, 0.0074]	0.022** [0.011, 0.043]	0.092** [0.039, 0.21]	0.0087** [0.0022, 0.034]	1.61 [0.74, 3.50]	6.51** [3.24, 13.0]	0.20** [0.10, 0.40]

Note:

*
p < 0.05,

**
p < 0.01.

N = 3,734. TL = telomere length = (T/S) * 2,400. Analyses conducted using logistic regressions, with the weighted polygenic risk score used as an instrument for TL. Standard errors are clustered by household. Additional covariates include four principal components-derived measures of genetic ancestry.

Table 5

Two-Sample Instrumental Variables Analyses of Impact of Telomere Length on Health, using Inverse Variance-Weighted Approach

	Coefficient [95% CI]				
	Hemoglobin A1c	Rheumatoid Arthritis	Heart Disease	Depression	Body Mass Index
TL (per 100bp difference)	0.00 [0.00, 0.00]	0.98 [0.95, 1.01]	1.02** [1.00, 1.03]	1.00 [0.97, 1.03]	0.00 [0.00, 0.00]

Note:

*
p < 0.05.

TL = telomere length = (T/S) * 2,400. Analyses conducted using inverse variance-weighted meta-analytic technique. Coefficients for arthritis, heart disease, and depression represent odds ratios, while values for hemoglobin A1c and BMI represent beta coefficients.