Psychoneuroendocrinology 102 (2019) 121–127

Contents lists available at ScienceDirect

Psychoneuroendocrinology

journal homepage: www.elsevier.com/locate/psyneuen

Unraveling the association between depression and telomere length using genomics

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ARTICLE INFO

Keywords:
Major depressive disorder
Depression
Telomere length
Cellular aging
Polygenic analyses
Genetic correlation

ABSTRACT

Objective: While there is robust evidence for a cross-sectional association between depression and shorter telomere length, suggestive of advanced biological aging, the nature of this association remains unclear. Here, we tested whether both traits share a common genetic liability with novel methods using genomics.

Methods: Data were from 2032 participants of the Netherlands Study of Depression and Anxiety (NESDA) with genome-wide genetic information and multiple waves of data on DSM-IV lifetime depression diagnosis, depression severity, neuroticism and telomere length. Polygenic risk scores (PRS) for both traits were built using summary results from the largest genome-wide association studies (GWAS) on depression (59,851 cases and 113,154 controls) and telomere length (37,684 samples). Additionally, a PRS for neuroticism was built (337,000 samples). Genetic overlap between the traits was tested using PRS for same- and cross-trait associations. Furthermore, GWAS summary statistics were used to estimate the genome-wide genetic correlation between traits.

Results: In NESDA data, the PRS for depression was associated with lifetime depression (odds ratio = 1.36; p = 6.49e-7) and depression severity level (β = 0.13; p = 1.24e-8), but not with telomere length. Similar results were found for the PRS for neuroticism. Conversely, the PRS for telomere length was associated with telomere length (β = 0.07; p = 8.42e-4) and 6-year telomere length attrition rate (β = 0.04; p = 2.15e-2), but not with depression variables. In summary-level analyses, the genetic correlation between the traits was small and not significant (rg = -0.08; p = .300).

Conclusion: The use of genetic methods in this paper indicated that the established phenotypic association between telomere length and depression is unlikely due to shared underlying genetic vulnerability. Our findings suggest that short telomeres in depressed patients may simply represent a generic marker of disease or may originate from non-genetic environmental factors.

1. Introduction

Research in the past decade provided consistent evidence for an inverse cross-sectional association between Major Depressive Disorder (MDD) and telomere length, suggestive of advanced biological aging in the depressed (Wolkowitz et al., 2010). Meta-analyses with > 34,000 subjects found average shorter telomere length in depressed patients versus controls, with a small to medium effect sizes (Cohen's d range = 0.21-0.55) (Darrow et al., 2016; Ridout et al., 2016; Schutte and Malouff, 2015). The meta-analyses included data from the large observational Netherlands Study of Depression and Anxiety (NESDA) cohort, in which we previously showed shorter telomeres ~800 persons with remitted MDD and ~1100 persons with current MDD, as compared to ~500 healthy controls (Cohen's d = 0.12) (Verhoeven et al., 2014). While the cross-sectional relationship is established by robust evidence, much is still unknown about the nature of this association.

Longitudinal research has so far failed to provide conclusive evidence regarding the direction of the relationship. Two prospective studies found depression status at baseline to be associated with greater telomere shortening over time (Shalev et al., 2014; Vance et al., 2018), while several others found no association between diagnosis and subsequent telomere length change (Chang et al., 2018a; Hoen et al., 2011; Schutte and Malouff, 2015). Two large longitudinal studies, including the NESDA cohort, showed a non-dynamic association between depression and short telomeres, with limited within-person variation over 6 and 10 year follow-up (Verhoeven et al., 2017 2016), suggestive of a rather stable relationship without dynamic interactions over time. The

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https://doi.org/10.1016/j.psyneuen.2018.11.029
Received 3 September 2018; Received in revised form 19 November 2018; Accepted 21 November 2018

presence of such constant non-dynamic associations could be interpreted as the result of shared etiological mechanisms that equally affect both depression and telomere length. As a first step to help unravel this relationship, we tested whether both traits have a common genetic base using data and novel methodologies recently developed in genomic epidemiology.

Genetic research now provides unique opportunities to investigate shared risks between traits by using new statistical tools and results from genome-wide association studies (GWAS) (Maier et al., 2018). GWAS data can be used to uncover shared genetic control between traits (i.e. genetic correlation) and to create individual predictions of genetic risk for certain phenotypic traits with a so-called polygenic risk score (PRS), a cumulative measure of the genetic burden for one trait carried by an individual (see (Maier et al., 2018) for an elaborate explanation). These methods provide the possibility to examine whether depression and short telomere length emerge from a common genetic liability.

Both telomere length and MDD have shown to be heritable traits (TL: 44–80% (Broer et al., 2013); MDD: 38% (Flint and Kendler, 2014)) giving rise to the possibility that shared genetic effects might impact both. First, a GWAS including 37,684 samples identified seven Single Nucleotide Polymorphisms (SNPs) that were independently associated with telomere length with relatively large effect sizes, of which the majority was located on genes that are known to be involved in telomere biology (e.g., TERC, TERT, NAF1, OBFC1, RTEL1) (Codd et al., 2013). Furthermore, an overarching meta-analysis (Wray et al., 2018) of all available GWAS datasets with depression, including 135,458 cases and 344,901 controls was recently performed by the Psychiatric Genomics Consortium (PGC). This GWAS revealed a highly polygenic architecture with many genetic variants with small effects, among which 44 loci were associated at genome-wide significant level.

Only very few studies have examined the possible shared genetic basis of the depression-telomere length relationship, and so far only reported on associations between telomere length genotype and depression phenotypes (Chang et al., 2018b; Wei et al., 2016; Wium-Andersen et al., 2017), while no study had the opportunity to examine the association between genetic risk variants for depression and telomere length phenotypes. One study did not find an association between a PRS for telomere length built with nine SNPs and self-reported lifetime depression in 17,693 female individuals of European ancestry (Chang et al., 2018b). On the SNP level, Wei and colleagues found that in a Swedish population-based study with 236 persons with depression, based on self-reported questionnaire data, and 426 controls, genetic variation on the human TERT gene was associated with depression, but only in those not exposed to childhood adversity (Wei et al., 2016). Overall, no study to date had the opportunity to thoroughly investigate the possibility of genetic overlap from multiple angles in one cohort.

For the first time, the current study examined same- and cross-trait associations genetic associations between MDD and telomere length, including a wide array of phenotypes determined over a 6-year period, namely lifetime depression diagnosis, depression severity, neuroticism, telomere length and 6-year telomere attrition rate. We estimated the degree of genetic overlap between the two traits with 1) individual-level data in a large well-characterized cohort enriched for psychiatric phenotypic information (N = 2032) using PRSs and 2) summary-level data analyses using genetic correlation analyses (see Fig. 1). This was done using results from the largest GWAS on telomere length (Codd et al., 2013) and the recent largest GWAS for MDD that for resulted in genome-wide hits (Wray et al., 2018). To our knowledge, moreover, this study is the first to examine associations between the PRS of telomere length and telomere attrition rate. Additionally, a PRS for neuroticism was built (based on GWAS in the UK Biobank), since this is a rather stable personality trait that is considered a strong risk factor for the development of depression (Hakulinen et al., 2015) with strong genetic correlation with depression (Luciano et al., 2018) and was previously associated to telomere length (Schoormans et al., 2018).

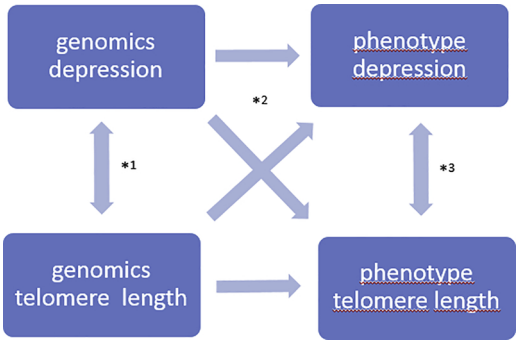

Fig. 1. Use of genomics in study design.
*1 = genome-wide genetic correlation using summary statistics in summary-level data
*2 = same and cross-trait associations using polygenic risk scores in individual-level data
*3 = phenotypic association from earlier investigations, summarized in meta-analyses (e.g. Ridout et al., 2016).

Hereby, we were able to validate associations with the PRS for depression and further test whether neuroticism can be considered a psychological endophenotype. To summarize, here we tested whether the phenotypic correlations between depression, depression-related traits and short telomere length are rooted in common genetic base potentially indicating shared etiology.

2. Methods

2.1. Study sample

Data were from the Netherlands Study of Depression and Anxiety (NESDA), an ongoing longitudinal cohort study examining the course and consequences of depressive and anxiety disorders. Study sample and methods have been described in detail elsewhere (Penninx et al., 2008). In short, the NESDA baseline sample consisted of 2981 persons between 18 and 65 years, including persons with a current or remitted depressive and/or anxiety disorder (74%) and healthy controls (26%). There were two exclusion criteria: 1) insufficient command of the Dutch language, and 2) a primary other clinical diagnosis of e.g. bipolar disorder, obsessive-compulsive disorder, PTSD, severe substance use disorder or a psychotic disorder. The study was approved by the Ethical Review Board of participating centres and all participants signed informed consent. Participants were assessed during a 4-hour clinic visit. Every 2 years after the baseline assessment, face-to-face follow-up assessments were conducted. The present study included psychiatric data from baseline, 2-year, 4-year and the 6-year follow-up. Telomere length was assessed both at baseline and at 6-year follow-up. A sample of 2032 unrelated individuals of European ancestry (based on self-report information further extended with principal components analyses of genotype data) with genetic, psychiatric and telomere length data at baseline were selected. Among these subjects, telomere length was available at 6-year follow-up for 1335 participants. Within the selected participants we calculated 10 ancestry-informative principal components accounting for potential populations substructures within European. These principal components were used to adjust analyses as appropriate for genetic associations in order to avoid the risk of identifying spurious associations driven by potentially different allele frequencies across sub-populations.

2.2. Phenotype measurements

2.2.1. Psychiatric characteristics

Persons were classified as control subjects or persons with a lifetime diagnosis of depression. Presence of a DSM-IV lifetime diagnosis of

depression across 6-year follow-up was assessed using the Composite Interview Diagnostic Instrument (CIDI, version 2.1) administered by specially trained research staff at baseline or one of the biannual follow-up assessments. Control subjects were defined as having no lifetime history of depressive or anxiety disorders. Severity of depressive symptoms in the past week was assessed in all subjects with the 30-item Inventory of Depressive Symptoms - Self Report (IDS-SR) (Rush et al., 1996) at baseline, 2-year, 4-year and 6-year follow-up. An average score was calculated across the assessments. Neuroticism was measured with NEO Five-Factor Inventory (NEO-FFI) (Costa and McCrae, 1995) at baseline, 2-year and 4-year follow-up and an average score was calculated. Age and sex were assessed during the baseline interview.

2.2.2. Leukocyte telomere length

Telomere length was assessed at baseline and 6-year follow-up. Fasting blood was drawn from participants in the morning and stored in a −20 °C freezer. Baseline and 6-year telomere length were determined at the laboratories of Telomere Diagnostics, Inc. (Menlo Park, CA) and University of California, San Francisco in 2012 and 2014, respectively, using quantitative polymerase chain reaction (qPCR) adapted from the published original method by Cawthon (Cawthon, 2002). Telomere sequence copy number in each patient's sample (T) was compared to a single-copy gene copy number (S), relative to a reference sample. The resulting T/S ratio is proportional to mean telomere length. The detailed method is described elsewhere (Verhoeven et al., 2014). As previously described, T/S ratios were converted into base pairs (bp) with the following formula: bp = 3274 + 2413x((T/S-0.0545)/1.16). The 6-year follow-up T/S ratios were adjusted relative to the baseline samples for systematic differences caused by different reference samples, by rerunning 226 baseline samples together with the 6-year follow-up samples. On average the T/S ratios of the 6-year follow-up runs were at 76% of the T/S ratios of baseline; consequently, the follow-up T/S ratios were divided by 0.76 (Verhoeven et al., 2016). The reliability of the assays was adequate; the inter-assay coefficient of variation was sufficiently low. A telomere length change score was calculated by subtracting baseline values from 6-year values, to allow testing for associations with baseline telomere length and 6-year telomere attrition rate.

2.3. Genetic measurements

2.3.1. Genotyping of NESDA sample

Genotyping (95% of the samples on Affymetrix 6.0 Human SNP array and the remaining on Perlegen-Affymetrix 5.0 array), quality control steps and imputation were previously described in detail (Mbarek et al., 2017). In short, after platform-specific QC the missing SNP genotypes between each platform were imputed using the GONL (Genome of the Netherlands) reference panel and then merged. Following more stringent QC, the SNPs from the cross-platform imputed dataset (~1.2 M) were used for a second round of imputations to the 1000Genomes Phase 3 all ancestries reference panel.

2.3.2 GWAS summary statistics. Summary statistics were obtained from the two largest GWAS on telomere length and depression. Codd et al. (Codd et al., 2013) performed a GWAS on telomere length in 37,684 samples. The Psychiatric Genomics Consortium (PGC) performed an overarching meta-analysis (Wray et al., 2018) of all available GWAS datasets with MDD, including 135,458 cases and 344,901 controls. For the present analyses we used GWAS summary statistics publicly released by PGC, obtained for a subset of 59,851 cases and 113,154 controls after the exclusion of the dataset from 23andME. Additionally, summary statistics for neuroticism were obtained from a large GWAS meta-analyses released by UK Biobank including 337,000 samples (Allen et al., 2012; Churchhouse and Neale, 2017).

2.3.2. Polygenic risk scores (PRS)

The PRS for telomere length level was based on the seven independent genome-wide significant SNPs in the discovery GWAS: ACYP2, TERC, NAF1, TERT, OBFC1, ZNF208 and RTEL1 (Codd et al., 2013). The seven SNPs were extracted from the 1000Genomes imputed dataset and all had an extremely high imputation quality (all $R^2 > 0.98$). The effects of the seven SNPs were looked up in the MDD GWAS (Wray et al., 2018) and presented in a supplementary table. The PRS was calculated as the number of risk alleles weighted by their effect sizes from the discovery statistics. The PRS for MDD was built based on the full polygenic signal from the MDD GWAS (Wray et al., 2018). Since NESDA data were part of the GWAS, we re-ran the MDD meta-analyses after removal of overlapping datasets (~3 K samples). Summary statistics of the discovery were filtered by removing In/Del and strand ambiguous variants, SNPs with INFO < 0.9, MAF < 0.01. Overlapping SNPs between the ~1.2 M cross-platform GONL imputed and those retained from the discovery summary statistics were carried forward. PRS were built according to LDpred method (Vilhjálmsson et al., 2015) using the dedicated software. A 1000 unrelated individuals were selected to calculate LD for reference. The fraction of causal SNPs was set at 5% consistently with the estimate for schizophrenia by Palla and Dudbdrige (2015). Last, the PRS for neuroticism used the same procedure applied for PRS for MDD using the LDpred method.

2.4. Data analyses

Same- and cross-trait associations of the PRSs for depression, neuroticism and telomere length with phenotypes were estimated using regression models (binary logistic for lifetime diagnosis and linear for depression severity, neuroticism, telomere length and telomere attrition rate), adjusted for baseline age, sex and 10 ancestry-informative genetic principal components (see Fig. 1). No other covariates were included since it has been demonstrated that adjusting for heritable covariates can bias effect estimates (Aschard et al., 2015). In linear regression analyses, the proportion of variance explained by the PRS was estimated based in the difference in R^2 between a linear model including only covariates and a model additionally including the PRS. In analyses with lifetime diagnosis as outcome, the change in Nagelkerke's pseudo-R^2 was re-scaled to the liability scale as suggested by Lee et al. (Lee et al., 2012) obtaining a value directly comparable with heritability and robust against ascertainment bias. Linear transformation on the liability scale was based on lifetime risk (K) for MDD of 0.15. Effect modification of lifetime diagnosis, age and sex was tested for the association between the PRS for telomere length and telomere length phenotypes by creating telomere length-by-effect modifier interaction terms and including them in the regression analyses predicting phenotype.

LD-score regression (LDSC) (Bulik-Sullivan et al., 2015) method was applied to summary statistics from GWAS to estimate the genetic correlation between telomere length, neuroticism and depression. In univariate LDSC, GWAS test statistics are regressed on LD-scores with the resulting slope providing an estimate of a trait SNP-heritability (h^2_{SNP}, the total variance in liability explained by the joint effect of all genotyped SNPs). In The bivariate extension of the method, the genetic covariance between two traits captured by all genotyped SNPs is estimated based on the slope obtained by regressing the products of the test statistics of the two traits on the LD-score. This allows the estimation of genetic correlation (*rg*, determined by the number of SNPs, and their level of concordance, shared between two traits). Please see (Bulik-Sullivan et al., 2015) for an extended description.

3. Results

3.1. Sample characteristics

Characteristics are presented in Table 1. The mean age of the study sample at baseline (N = 2032) was 42.5 years (SD = 12.8; range 18-65) and 67.3% was female. Lifetime depression diagnosis was present for 83.1% (N = 1688) of the sample. The average severity score was 19.0,

Table 1
Sample characteristics (N = 2032).

Demographics	
Age at baseline (mean ± s.d., range)	42.5 (12.8) [18-65]
Sex (% female, N)	67.3, 1367
European ancestry (%, N)	100, 2032
Psychiatric characteristics	
Lifetime depression diagnosis (% yes, N)	83.1, 1688
Depression severity (average 6-year IDS score) (mean ± s.d.)	19.0 (12.0)
Neuroticism (average 4-year NEO-FFI score) (mean ± s.d.)	35.3 (8.6)
Cellular aging	
Telomere length (base pairs) (mean ± s.d.)	5468.8 (624.7)

Abbreviations. IDSInventory of Depressive Symptoms; NEO-FFINEO Five-Factor Inventory.

indicative of mild depression severity. The average neuroticism score was 35.3 [range = 12–60].

3.2. Genetic analyses

3.2.1. PRS analyses

In analyses based on NESDA data, the PRS for MDD was associated with lifetime MDD diagnosis (per SD increase: OR = 1.36, p = 6.49e-7, explaining 2.6% of MDD liability variance), depressive symptom severity (per SD increase: β = 0.13, p = 1.24e-8, explaining 1.6% of the variance) and neuroticism (β = 0.13, p = 2.48e-9, explaining 1.7% of the variance), see Table 2. The PRS for MDD was, however, not associated with telomere length or telomere length attrition rate phenotypes, see Table 2. Similarly, the PRS for neuroticism was associated to lifetime MDD diagnosis (0.9% of variance explained), depression severity (1.3% of the variance explained) and neuroticism (1.7% of the variance explained), but not to telomere length phenotypes, see Table 2.

The PRS of telomere length was strongly associated to phenotype telomere length (per SD increase: β = 0.07; p = 8.42e-4, explaining 0.5% of the variance), and to 6-year telomere length attrition rate (per SD increase: β = 0.04; p = 2.15E-2, explaining 0.1% of the variance). Yet, no associations were found between PRS for telomere length and depression phenotypes, see Table 2. The effects of the seven genome-wide significant hits from the telomere length GWAS in the depression GWAS are presented in Supplementary Table 1 and showed no significant associations, confirming an absence of shared genetic etiology.

Effect modification analyses predicting phenotype telomere length revealed no interaction effects for age-by-PRS telomere length (β = -.050; p = .502) or lifetime depression-by-PRS TL (β = -0.59; p = .220). Effect modification for sex was present (sex-by-PRS telomere length β = -.213; p = .006). Split file analyses showed that the PRS telomere length was more strongly associated to phenotype telomere length in males (β = .163; p = 1.0e-5) than in females (β = .029; p = .268). Similar results were found in split file analyses for telomere length change with a stronger effect of PRS telomere length in males (β = .091; p = 4.52e-3) than in females (β = .022; p = .319), although the overall sex-by-PRS telomere length interaction effect for telomere attrition rate was not statistically significant (p = .102).

3.2.2. Genome-wide genetic correlation using summary statistics

Univariate LDSC analyses provided significant h^2_{SNP} estimates for MDD (8.7%, s.e. = 0.4%), neuroticism (12%, s.e. = 0.6%) and telomere length (7.0%, s.e. = 1.6%). In bivariate LDSC, the genetic correlations between telomere length and depression (rg = -0.077; s.e. = 0.075; p = .300) and telomere length and neuroticism (rg = -0.037; s.e. = 0.056; p = .511) were small and not significant, further suggesting the absence of pleiotropy. The genetic correlation between depression and neuroticism, however, was strong and highly significant (rg = 0.699; s.e. = 0.028; p = 2.96e-141).

4. Discussion

In the present study, we examined the nature of the association between depression and telomere length using data and statistical tools from genomics. We found strong significant associations of the PRS for depression with lifetime depression diagnosis and depression severity and similarly for the PRS of telomere length with telomere length and, for the first time, telomere attrition rate. However, no cross-trait associations were found, indicating that the polygenic load of one trait did not overlap with the polygenic load of the other. Likewise, we found strong associations of the PRS for neuroticism with depression phenotypes, but not with telomere length phenotypes. Further, using summary statistics, we found a strong genetic correlation between depression and neuroticism but no genetic correlation between the either of those traits with telomere length. Altogether, these results suggest that it is unlikely that the phenotypic association between telomere length and depression is a consequence of shared underlying genetics.

Previous research has provided robust evidence for a phenotypic association between short telomere length and depression status (Ridout et al., 2016), and - to a lesser extent - neuroticism (Schoormans et al., 2018). However, many questions regarding the nature of the associations are still unanswered. Most studies up until now have been crippled by their observational and often cross-sectional designs, leaving room for multiple interpretations. Up until now, researchers have been unable to determine whether depression and short telomeres are causally associated, whether they share a common liability or whether the association may be the result of unmeasured or residual confounding. In the present study, by applying novel methods from genetic epidemiology, we found no evidence for a shared genetic predisposition for the risk of short telomere length and the risk of depression or neuroticism. Interestingly, and as expected, the genetic risks

Table 2
Associations of polygenic risk scores (PRS) with phenotypes (N = 2032).

	Depression PRS				Neuroticism PRS				Telomere length PRS			
	B	SE	OR / β	p-value	B	SE	OR / β	p-value	B	SE	OR / β	p-value
Depression phenotypes												
Lifetime major depressive disorder [no = reference][a]	0.31	0.06	1.36	**6.49e-7**	0.17	0.06	1.19	**4.55e-3**	0.10	0.06	1.11	.091
Depression severity (average 6-year IDS score)[a]	1.54	0.27	0.13	**1.24e-8**	1.35	0.27	0.11	**4.17e-7**	0.20	0.27	0.02	.466
Neuroticism (average 4-year NEO-FFI score)[a]	1.13	0.19	0.13	**2.48e-9**	1.10	0.19	0.13	**4.33e-9**	−0.01	0.19	−0.001	.973
Telomere length phenotypes												
Telomere length at baseline[a]	−10.8	13.4	−0.02	.417	−0.77	13.2	−0.001	.954	44.1	13.2	0.07	**8.42e-4**
Telomere length attrition (6-year change score; N = 1335)[b]	−5.3	10.4	−0.01	.607	−12.58	10.0	−0.02	.208	23.2	10.1	0.04	**2.15e-2**

Note. Significant p-values are highlighted as bold.
Abbreviations. IDS = Inventory of Depressive Symptoms; NEO-FFI = NEO Five-Factor Inventory.
[a] Adjusted for age, sex and 10 Principal Components.
[b] Additionally adjusted for telomere length at baseline.

for neuroticism and depression were strongly related, indicating a clear genetic connection between the two traits and suggesting that person with a high genetic risk for neuroticism also have a high genetic risk for depression. Neuroticism is considered a stable trait it might thus indeed serve as an endophenotype for depression (and other mental health disorders).

Our results are in line with earlier studies that examined associations between telomere length genetics and depression phenotype (no studies to date examined the opposite cross-trait associations). Chang et al. (2018b) found an association between the PRS for telomere length (combined from nine alleles) and telomere length phenotype, but not with lifetime depression or depressive symptom trajectories in 17,693 women of European ancestry. Similarly, a study linking the PRS of telomere length to nine different health outcomes in 3734 participants of the Health and Retirement Study, found no association with any of the health outcomes, including depression and general psychiatric health (Hamad et al., 2016). Wei and colleagues (Wei et al., 2016), on the other hand, found that homozygosity of the risk allele A of SNP rs2736100 (TERT) was associated to depression but only in a subgroup of subjects not exposed to childhood adversity from 1436 participants of a Swedish cohort study. Considering the potential pitfalls of candidate GxE studies this result should be considered with extreme caution (Duncan and Keller, 2011).

This results of the present study may help to shed light on previous studies using other genomic methods that attempted to identify a potential direct causal connection between telomere length and depression, such as Mendelian Randomization (MR). MR is a method developed from instrumental variable analyses which uses genetic variants as instruments to estimate the causal effect of an exposure on an outcome (see (Burgess et al., 2012) for an elaborate explanation of this research method). Wium-Andersen et al. (2017) examined telomere length's causal effect on depression using MR and found no indication for a causal relation between TERT, TERC and OBFC1 and depression (register-based attendance at hospital for depression) in 67,306 individuals from the Danish general population. Since MR leverages on the pleiotropic effect of certain SNPs on different traits and in the present study we found no indication of pleiotropic effect of telomere length SNPs on depression (both in PRS analyses in our NESDA data and by looking up the effect of telomere length related SNPs in the large MDD GWAS), this lack of pleiotropic effect may explain the negative results of the study by Wium-Andersen et al. (2017). Furthermore, we showed a significant association between the PRS of telomere length and 6-year telomere length attrition rate. This is in accordance with large twin studies which yielded heritability estimates for telomere attrition between 24% and 32% (Hjelmborg et al., 2015). Here, we showed associations with a PRS built from seven SNPs, showing that the same SNPs that are genetically related to longer telomere length, are also positively associated to telomere length attrition, indicating "slower" attrition. This might be related to pathways that involve upregulation of the telomere-maintaining enzyme telomerase, since e.g. TERT and TERC encode for the catalytic subunit and the telomerase RNA component, respectively (Codd et al., 2013). Last, we found preliminary evidence for a sex difference in the strength of the association between PRS and telomere length phenotypes, with a stronger link in males than in females. This remains to be replicated by larger cohorts.

The majority of studies, including the current study, found no evidence for a shared genetic basis for short telomere length and depression or neuroticism. So, how can we interpret these findings in the context of the large amount of evidence from observational studies? One explanation of the null results is that telomere length should primarily be viewed as a risk marker of disease processes (i.e., a marker that is associated to an outcome such as depression, or diseases often comorbid to depression, but does not meet the formal criteria for causality), rather than a causal risk factor (Hamad et al., 2016). Observational studies may have suffered from confounding and no true causal link may exist. Another explanation is that depression is a highly heterogeneous disorder, which leaves the possibility that, while the PRS for telomere length was not associated to depression overall, it might map more closely onto specific subtypes of depression that remain to be elucidated. A third explanation, however, is that while the genetic impact associated to both traits is limited on the other trait, there might be a causal relationship inflicted by non-genetic factors. On the one hand, changes in telomere length as a consequence of changes in behaviors, stress levels or other (non-genetic) psychosocial factors may still affect depression status. On the other hand, changes in depression levels that are not due to the genetic make-up but rather to, for instance, early life trauma or chronic life stress may affect telomere length. This may be through pathways of depression-related biological abnormalities (e.g. inflammation and oxidative stress) that are associated with, and may cause, telomere shortening (Lindqvist et al., 2015). In a previous investigation in the NESDA cohort we showed that such factors, including pro-inflammatory cytokines, metabolic alterations and cigarette smoking, were indeed significant mediators of the depression-telomere length association (Révész et al., 2016). However, a recent cross-sectional study found shorter telomere length in never-depressed daughters (aged 10–14) of depressed mothers (Gotlib et al., 2014), suggesting that short telomere length might be present in subjects at higher genetic risk for depression, even before depression onset. Nevertheless, in the results by Gotlib et al. it is not possible to completely disengage the effect of genetic risk from environmental life stress, since all daughters were living at home with their affected mothers. The question whether depression and short telomere length are truly causally related or not might only be answered by studies with multiple assessments that follow participants during different parts of the lifespan, including childhood, adolescence, adulthood and old age, that include genetic, physiological, psychological and lifestyle examinations.

Major strengths of this study are the large sample size and its well-characterized participants including persons with a lifetime depression diagnosis and healthy controls. Furthermore, for the first time, both genetic and phenotypic data from depression and telomere length were available, allowing same- and cross-trait associations in one cohort. Next to telomere length phenotype we also had data of telomere length attrition rate and for the first time showed an association between the PRS for telomere length and slower attrition rate. Some limitations should also be noted. While we had a large sample size available for individual-level data analyses, this is considered a limited sample size in genetic research. This may have limited the ability to detect small effect sizes in this sample, increasing the chances of Type II errors. However, our results in individual-level data were confirmed by genetic correlation analyses with large samples ($> 100{,}000$). Last, although we used the most recent GWAS, only a limited proportion of SNP-heritability was explained by the PRS. In line with this, it should be noted that at the current stage, PRSs have limited value as clinical prediction tools; this will increase when larger training GWAS will become available. Nevertheless, results of PRS analyses should be considered for their value in elucidating genetic architectures and testing whether different traits have overlapping genetic liabilities.

In conclusion, the use of genomic methods in this paper indicated that the established phenotypic association between telomere length and depression is unlikely due to shared underlying genetic etiology. Our findings suggest that short telomere length might represent a generic marker of disease rather than a causal risk factor, or that short telomeres in depressed patients may originate from different non-genetic factors, such as chronic life stress, unhealthy behaviors and associated physiological disturbances.

Grant support

Telomere length assaying was supported through a NWO-VICI grant (number 91811602).

Conflicts of interest

BP has received research funding (not related to the current paper) from Boehringer Ingelheim and Jansen Research. The other authors declare no relevant conflict of interest.

Declaration of interest

None of this material has been published in print or in electronic format or is under consideration for publication elsewhere. The co-authors have read and approved the manuscript in its present form.

Acknowledgements

Funding for the NESDA study (www.nesda.nl) was obtained from the Netherlands Organization for Scientific Research (Geestkracht program grant 10-000-1002) and is supported by participating universities and mental health care organizations including the Center for Medical Systems Biology (CSMB, NWO Genomics), Biobanking and Biomolecular Resources Research Infrastructure (BBMRI-NL), VU University's Institutes for Health and Care Research (EMGO+) and Neuroscience Campus Amsterdam, University Medical Center Groningen, Leiden University Medical Center, National Institutes of Health (NIH, R01D0042157-01A, MH081802, Grand Opportunity grants 1RC2 MH089951 and 1RC2 MH089995). Part of the genotyping and analyses were funded by the Genetic Association Information Network (GAIN) of the Foundation for the National Institutes of Health. Computing was supported by BiG Grid, the Dutch e-Science Grid, which is financially supported by NWO.

Appendix A. Supplementary data

Supplementary material related to this article can be found, in the online version, at doi:https://doi.org/10.1016/j.psyneuen.2018.11.029.

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