The Biological Consequences of Biological Father Loss: Comparing the Effect of Father Incarceration, Union Dissolution, and Death for Child and Mother Telomere Length

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Abstract

Recent research has shown that telomere length has an important association with indicators of aging and lifetime stress. Telomere length is thought to be an early indicator of poor health prior to major health symptoms manifesting—especially for children and healthy adults. One important stressor for many children is the loss of their biological father. Similarly the loss of a spouse or partner can be difficult for the mother. To date limited research has examined the effect of father/partner loss on telomere length, and none have explicitly compared different types of exits nor compared their influence on the child and the mother. This paper utilizes the Fragile Families and Child Wellbeing Study telomere length (mothers- n=2678, children- n=2843) when the child is 9 years old. We find significant effects of biological father incarceration and death for children and significant effects of incarceration, union dissolution, and death of the biological father for the mother.

Extended Abstract

A large body of research has documented chronic stress associated with the loss of a family member is thought to hasten the degeneration of physiological functioning (1-6). Moreover, a recent review suggests that the resulting cellular effects of chronic stress are often indistinguishable from aging, suggesting that chronic stress can disproportionately age individuals (2-4,7). One recent line of research suggests that DNA telomere length (TL), the TTAGGG sequence repeat at the end of the chromosome, has an important association with indicators of aging and lifetime stress (1-6) and thus may provide leverage for studying the association between family member loss and poor health (8). Using TL as a marker for weathering provides a more time-sensitive and gradated measure of long-term health and wellbeing compared to mortality or disease status—especially for children.

Over the past 15 years, research has shown that telomere length, the TTAGGG sequence repeat at the end of the chromosome, has an important association with indicators of aging and lifetime stress (9-16). Due to the mechanism of DNA replication, chromosomes shorten with each cycle of chromosomal replication and cellular division. The repetitive telomere sequence is sacrificed to protect genes or other essential information near the ends of chromosomes. In addition, the presence of the telomere prevents fusion of adjoining chromosomal ends. Over time, due to each cell division, the telomere ends become shorter, and so the telomere has been referred to as a "mitotic clock" (11,16). In some cell lineages, such as stem cells, germ cells and in many cancer cells, the expression of telomerase renews telomere length. In most differentiated cell lineages, however, telomerase is not expressed (11). This provides a limit to the number of times a cell line can replicate. When telomeres are sufficiently short the cell enters a state of replicative senescence and stops dividing. In one model, as more cells enter senescence, organismal senescence begins, giving a specific biological reason for Gompertz-Makeham law of mortality (that mortality rises rapidly with age) (11, 17). TL is also associated with a wide range of disease morbidities (18-21). Thus it is no surprise that research has focused on TL as a measure of 'biological age,' and that TL is quickly becoming a popular biomarker for aging research. Several risk factors appear to predict TL, including smoking (22), mental illness (23-25) (particularly depression), stress (12,25-27), obesity (22,28,29) and caregiving for someone with a major illness (30). This work generally shows that having a risk factor is negatively associated with TL in adulthood.

Research on the link between social factors and TL is in its infancy. Nevertheless, multiple studies support the hypothesis that several social factors are associated with TL (7, 22-31). For example, in a recent review of the literature 5 studies were found to examine some aspect of SES (education or income) and all of them found that those with lower SES had shorter telomeres (31). Other studies have shown that parenting styles, family structure and even adult cargiving all appear to influence telomere length for

adults. Far less is known about children. One recent publication showed that having a transition in early life was associated with a shorter telomere length—however, the type of transition (e.g. marriage or divorce) was not specified and it was confounded with other variables in the model. However, multiple papers have shown that early negative childhood events such as maltreatment, poverty, and maternal depression are associated with shorter telomere lengths.

One particularly devastating loss for a family is the loss of one of its members. A robust literature has shown strong effects of the loss of a biological father on children. Although many recover emotionally, the economic and resource loss is often not easily overcome even after many years. Importantly, the type of loss, whether through union dissolution, death or incarceration also appears to be important. Similarly, the loss of a partner can be extremely difficult for the mother as well. The increased demand on her time, emotional resources, and abilities can significant erode her health. Also interesting to note is that often the type of loss of a partner comes with differential support—suggesting possibly different effects on her health.

This paper uses recently assayed telomere length data from the Fragile Families and Child Wellbeing study to examine the role of a biological father loss on both mother and child health. We use telomere length as an early indicator of stress-related biological aging. Using these data we can compare how loss of a father and partner influences both mother and child and how the type of loss (incarceration, separation (e.g. divorce), or death). We also examine how the timing of the loss may or may not be important by using the longitudinal prospective data collection between birth and age 9. Finally, we inspect how some mediators such as changes in resources and social support may explain these effects.

METHODS

Sample

Our data are taken from the *Fragile Families and Child Wellbeing Study* (FFCWS). FFCWS is based on a stratified, multi-stage, probability sample of children born in large U.S. cities between September 1998 and September 2000, with an oversample of children born to unmarried parents (three-quarters unwed, one-quarter wed) (36). Because of the large oversample of non-marital births and the urban nature of the sample, the families in this sample have a particularly wide range in quality of environments. This feature of the data affords us greater power to detect effects of social environment than an equally sized sample of all births. Baseline interviews with mothers and fathers were conducted within 48-hours of the child's birth, and subsequent interviews were conducted when the focal child was 1, 3, 5 and 9 years old. Saliva DNA samples were taken at the age 9 follow-up, using the Oragene®DNA sample collection kit (DNA Genotek Inc, Ontario).

Telomere Measurement

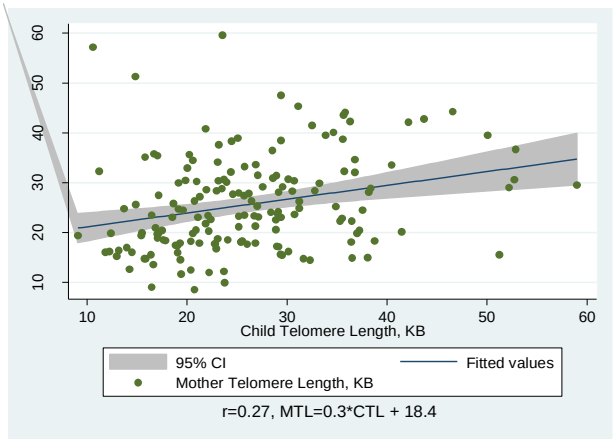
Telomere length will be measured using a quantitative real-time PCR assay that incorporates an oligomer standard to permit measurement of absolute (in Kbase per chromosome) rather than relative telomere length (37)**.** Briefly, this method adapts the approach of Cawthon (38) in which relative telomere length is determined by quantitative PCR by determining the ratio of telomere copy repeats to a single copy reference gene (the gene is *36B4*). To determine absolute telomere length, an 84mer oligomer standard TTAGGG is used to construct a standard curve. A separate standard curve for the single copy gene incorporates a 79mer containing the reference gene 36B4. This enables calculation of total telomere length per diploid genome, while the 36B4 product gives the number of diploid genomes. Length per chromosome is given by dividing telomere length per genome by 92, the number of telomeres per diploid genome. Samples are measured in triplicate and the results averaged—the coefficient of variation for these triplicate measurements was < 11%--which is well below the standard 15-20% suggesting a relatively high level of accuracy. Distribution of samples in the 96 well plates is randomized and each plate will contain at least 10 repeats from prior runs in order to detect and limit potential batch effects. To

further identify batch effects, reference DNA from a *tel*- cell line, and the same line after stable integration of *tel* were included in each plate. Reference DNA was harvested at a single time, aliquoted, and frozen. In preliminary experiments, normalizing the telomere length by this standard did not significantly change results, but the reference was included in ensuing runs for quality control.

Also, since much of the current work on TL has been done on blood leukocytes and not on saliva, we conducted a small laboratory study comparing the two (23). The results of the healthy adult sample generally found telomere length was significantly longer in saliva than blood leukocyte derived DNA; however saliva and leukocyte DNA lengths were highly and significantly correlated (R=0.8, p=0.001). The difference in absolute TL is not surprising, since difference cell types (saliva, mainly epithelial cells vs blood leukocytes) have different rates of division and thus difference rates of telomere shortening and different micro-environmental exposures. However, the fact that they are all highly correlated in adults suggests that there is no *a priori* reason to prefer one tissue source over another. Therefore, it may be feasible to compare studies that evaluate telomere lengths across different DNA sources. Unless researchers are interested in one particular cell type (e.g. cancer) there is little reason to believe that one tissue TL is better index of biological aging than another

Initial Results

Telomeres In a pilot study of 174 African American mothers from the FFCWS, several important results were found. First, there is a moderate and significant (R=.27, p=.002; see figure) correlation between mother's and child's TL. Some of this correlation is probably due to biological inheritance, but some of it may also be due to similar recent life experiences. We found that death, relationship history, and incarceration) is strongly related to TL; stronger than mental health, BMI and smoking status. Namely, being a single parent is clearly a difficult and stressful circumstance for mothers. Even when parents are good and are able to protect children from the difficulties, the mother's health (in this case) may be strongly negatively influenced by remaining single. There is also some limited evidence that having a biological father exit the family through separation or divorce matters for TL, but the timing of that event appears to be important. Finally, we found a strong negative association with TL and having a father/partner incarcerated.



We are now examining the full Fragile Families telomere data. And, in general the results from the initial pilot project are replicating in the larger study. In particular the strong negative effect of father/partner incarceration is striking. Even controlling for several possible moderators does not appear to account for the effect of incarceration for both mothers and children. Although much more smaller in number father/partner death also appears to have a negative relation with TL. Finally, it appears that the timing of the father union dissolution is key—with early exits having larger negative associations. We are currently examining these effects by key demographic groups (i.e. race and gender). And we are beginning our examination of potential moderators.