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Family-centered prevention ameliorates the longitudinal association between risky family processes and epigenetic aging

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Background: Research has suggested that 'risky' family processes have unforeseen negative consequences for health later in life. The purpose of this study was to further understanding of risky family environments and development of health vulnerabilities by (a) examining the likelihood that elevated levels of parental depressive symptoms when children are age 11 forecast accelerated epigenetic aging 9 years later at age 20; (b) determining whether participation in an efficacious family-centered prevention program focused on enhancing supportive parenting and strengthening family relationships will ameliorate this association; and (c) testing a moderation-mediation hypothesis that prevention-induced reductions in harsh parenting across adolescence will account for prevention effects in reducing accelerated epigenetic aging. **Methods:** In the rural southeastern United States, parents and 11-year-old children from 399 families participated in the Strong African American Families (SAAF) program or a control condition. Parents reported their own depressive symptoms when their children were 11, and both youths and parents reported youth exposure to harsh parenting at ages 11 and 16. Blood was drawn from youths at age 20 to measure accelerated epigenetic aging using a marker derived from the DNA methylation of cells. **Results:** Elevated parental depressive symptoms forecast accelerated epigenetic aging among youths in the control condition, but not among SAAF participants. Moderated-mediation analyses confirmed that reductions in harsh parenting accounted for SAAF's protective effects on epigenetic aging. Subsequent exploratory analyses indicated that accelerated epigenetic aging forecast emotional distress among young adults in the control condition but not among those who participated in SAAF. **Conclusions:** This study is unique in using a randomized prevention trial to test hypotheses about the ways risky family processes contribute to accelerated epigenetic aging. The results suggest that developmentally appropriate family-centered interventions designed to enhance parenting and strengthen families can buffer the biological residue of life in a risky family. **Keywords:** Depression; epigenetics; epigenetic clock; health; intervention; parenting; prevention.

Introduction

A growing body of research hints at the possibility that emotionally cold and harsh interactions with one's parents during childhood contribute not only to psychosocial and psychiatric problems but also to vulnerability to chronic diseases later in life (Shonkoff, Boyce, & McEwen, 2009). For example, retrospective studies reveal that adults reared in harsh home environments evince higher blood pressure, poorer metabolic profiles, and higher levels of depressive symptoms than do adults reared in less harsh households (Miller, Chen, & Parker, 2011). The risky family model was designed to offer a theoretical account of the ways in which living in such family environments during childhood and adolescence contribute to health vulnerabilities that carry forward across the life span (Repetti, Taylor, & Seeman, 2002). It posits that home environments headed by a primary caregiver with elevated levels of depressive symptoms who uses harsh parenting practices trigger a cascade of psychological and biological vulnerabilities, including deficits in the regulation of negative emotions and increased reactivity of the hypothalamic-pituitary-adrenal (HPA) axis and the sympathetic nervous system (SNS). Activation of the end products of the HPA axis and the SNS, the hormones cortisol, epinephrine, and norepinephrine, are adaptive in the short run; however, sustained exposure to these hormones can weather physiological systems, resulting in premature aging of cells and a shortened life expectancy (Geronimus, Hicken, Keene, & Bound, 2006).

This study was designed to advance understanding of the association between risky family processes and subsequent health status by testing hypotheses involving prospective associations among elevated parental depressive symptoms, harsh parenting, and premature epigenetic aging. The hypotheses were tested with a sample of rural African American youths who took part in a randomized prevention trial and who were followed from preadolescence (age 11) to young adulthood (age 20). The preventive intervention, the Strong African American Families (SAAF) program, was designed to enhance nurturant-involved parenting, reduce harsh parenting, and strengthen family relationships (Brody et al., 2004); it is described in more detail later. To

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measure premature aging, we used a recently developed epigenetic biomarker derived from the DNA methylation of cells. This marker has been validated in cells from more than a dozen tissues and reflects the disparity between an individual's biological and chronological ages (Horvath, 2013; Horvath et al., 2014). Accelerated epigenetic aging also forecasts heightened risks for all-cause mortality (Marioni et al., 2015). In the following sections, we present the study hypotheses and the rationale for them.

Among women living in low-SES circumstances, depression is two to three times more prevalent in both community and postpartum samples (Hobfoll, Ritter, Lavin, Hulsizer, & Cameron, 1995; Mayberry, Horowitz, & Declercq, 2007). Depressive symptomatology has been associated with diminished maternal sensitivity to infant needs and with heightened levels of harsh and disengaged behavior with children and adolescents (Essex et al., 2011; Halligan, Herbert, Goodyer, & Murray, 2007). Research suggests that growing up with a large dose of harsh parenting does not lead to positive physical and mental health outcomes (Miller et al., 2011). Offspring of mothers with high levels of depressive symptoms experience elevated HPA and SNS activation in childhood and adolescence (Field, 1994). Its most prominent legacy concerns problems with emotion regulation; youths who receive harsh parenting respond more intensely to new stressors even if they represent independent life events outside the home (Cicchetti & Rogosch, 2009). When they reach adulthood, these offspring continue to exhibit hypervigilance, impaired emotion regulation, dysregulation of the HPA axis, and heightened SNS activation (Weissman et al., 2006). Thus, we predicted that exposure to elevated levels of parental depressive symptoms and the harsh parenting practices they sponsor will forecast accelerated epigenetic aging.

Prevention researchers have demonstrated a specific form of moderation in which program effects are stronger for individuals who are at highest risk at program entry (Brody, Kogan, & Grange, 2012). This type of moderated program effect can be viewed from a risk reduction perspective. The program reduces the naturally occurring association of risk with outcomes that emerges in the control condition. Conceptually, this pattern is identical to a protective-stabilizing effect described in the resilience literature, in which a resilience resource reduces the negative impact of risk factors over time (Rutter, 2005). From this perspective, a prevention program can be viewed as a 'constructed resilience resource' because it is designed to support processes that promote resilience. SAAF was conceptualized as a resilience resource. It was designed to mitigate the negative impact of life stress on rural African American youths by increasing family-based protective parenting processes and decreasing harsh parenting (Brody et al., 2004). SAAF has demonstrated stress-buffering capabilities for rural African American youths experiencing even extreme levels of stress (Brody et al., 2012). For this reason, in this quasi-experimental analysis, we tested the moderated-mediation hypothesis that heightened parental depressive symptoms would forecast accelerated epigenetic aging for youths assigned randomly to the control condition but not for youths assigned to the SAAF condition, and that SAAF-induced reductions in harsh parenting would account for this protective-stabilizing effect.

Summary of this study

In this study, rural African American 11-year olds and their primary caregivers took part in the SAAF randomized prevention trial. Caregivers reported their own depressive symptoms when their children were 11, and both youths and parents reported youth exposure to harsh parenting at ages 11 and 16. Blood was drawn at age 20 to measure epigenetic aging using a marker derived from the DNA methylation of cells.

Methods

Participants

Details of the original SAAF prevention trial are provided elsewhere (Brody et al., 2004). Briefly, participants included 667 African American families who resided in rural Georgia. A targeted youth from each family (mean age at pretest = 11.2, *SD* = 0.34) and the parent who had primary responsibility for the youth's care took part in data collections. At pretest, although the primary caregivers in the sample worked an average of 39.4 hr per week, 46.3% lived below federal poverty standards; the proportion was 53.7% at the age 20 assessment. Table 1 provides demographic data on the sample. At age 20, 500 emerging adults were selected randomly to have blood drawn. The selection of a random subsample was made necessary by financial constraints associated with the costs of assaying genome-wide methylation. Of this subsample, 399 agreed to participate; they constituted the sample in this study. At age 11, 242 of these participants had been assigned randomly to the SAAF condition and 157 had been assigned randomly to the control condition. The original random assignment oversampled participants into the SAAF condition; this accounts for the greater number of 20-year olds in the SAAF group. Comparisons of pretest data collected at age 11 and data collected at age 18, using independent *t*-tests and chi-square tests, of the youths who provided blood samples at age 20 with those who did not provide them did not reveal any differences on any demographic or study variables (see Table 2). Written informed consent was provided. Each family was paid $100 after the assessment. All procedures were approved by the Institutional Review Board at the University of Georgia.

Intervention implementation

The SAAF prevention program consisted of seven consecutive, 2-hr weekly meetings held at community facilities, with separate parent and youth skill-building curricula and a family curriculum (see Brody et al., 2012, for a complete description, including a summary of efficacy findings). Parents in the prevention condition were taught the consistent provision of instrumental and emotional support, high levels of

Table 1 Sample characteristics at study entry: The Strong African American Families program in rural Georgia ($N = 399$)

	Control				Intervention			
Characteristics	*n*	*M*	%	SD	*n*	*M*	%	*SD*
Youth age (in years)		11.73		0.32		11.61		0.37
Parent age (in years)		37.56		7.28		37.57		8.03
Family poverty status								
Above 150% poverty guideline	63		40.1		86		35.5	
Below 150% poverty guideline	34		21.7		61		25.5	
Below 100% poverty guideline	60		38.2		95		39.3	
Youth gender								
Male	77		49.0		104		43.0	
Female	80		51.0		138		57.0	
Parental education								
<High school	36		22.9		47		19.5	
High school degree or GED	40		25.5		86		35.3	
Some college or trade school	75		47.8		96		39.8	
≥College graduate	6		3.8		13		5.4	
Parental employment								
Unemployed	33		21.0		54		22.3	
Employed	124		79.0		188		77.7	
Parental marital status								
Married or partnered	68		43.3		98		40.5	
Single	89		56.7		144		59.5	

Table 2 Pretest equivalence of experimental condition for participants who did or did not provide methylation data

	With methylation data				Without methylation data					
	Intervention ($n = 242$)		Control ($n = 157$)		Intervention ($n = 127$)		Control ($n = 141$)			
Variables at Pretest	*M*	*SD*	*M*	*SD*	*M*	*SD*	*M*	*SD*	$F(1, 663)$	*p*
Gender (male)	0.43	0.50	0.49	0.50	0.52	0.50	0.48	0.50	1.515	.219
Family socioeconomic risk	2.43	1.46	2.29	1.55	2.42	1.43	1.99	1.48	1.562	.212
Parental depression group	0.27	0.44	0.29	0.45	0.19	0.40	0.23	0.42	0.078	.781
Harsh parenting	7.12	1.38	6.98	1.41	7.24	1.62	7.08	1.49	0.003	.955

monitoring and control, adaptive racial socialization strategies, and methods for communicating about sex and alcohol use. Youths learned adaptive behaviors to use when encountering racism, the importance of forming goals for the future and making plans to attain them, and resistance efficacy skills.

Data collection procedures

All data were collected in participants' homes using a standardized protocol at pretest and long-term follow-up. Two African American field researchers worked separately with the primary caregiver and the target child. Certified phlebotomists went to each participant's home to draw a blood sample when the participants were 20 years of age.

Measures

Parental depression status. When youths were 11 years of age, parents reported their depressive symptoms on the Center for Epidemiologic Studies Depression scale (CES–D; Radloff, 1977), which is widely used with community samples. Primary caregivers, 91.5% of whom were mothers, rated each of 20 symptoms on a scale of 0 (rarely or none of the time), 1 (some or little of the time), 2 (occasionally or a moderate amount of the time), or 3 (most or all of the time). Alpha was .85. Consistent with psychometric studies of the CES–D, we used a score of 16 as a cutoff for identifying parents with lower ($n = 288$; 115 in the control group and 173 in the SAAF group) versus higher levels of depressive symptoms ($n = 111$; 42 in the control group and 69 in the SAAF group). To simplify language, we hereafter refer to these groups of families as the euthymic group and the high depressive symptoms group.

Harsh parenting. At age 11 (pretest) and age 16 (long-term follow-up), both parents and youths reported the frequency of harsh parenting using a questionnaire developed for research with rural African American families (Brody et al., 2001). Four items assessed parents' use of striking and shouting at their children to discipline them; the items were rated on a scale of 1 (*never*) to 4 (*always*). Harsh parenting was operationalized as the average of the parents' and youths' ratings for both pre- and post trial administrations (Cronbach's alphas were .61 and .65, respectively). Low internal consistency for measures of harsh parenting is common in the literature due to low base rates of these disciplinary practices (Brody et al., 2001; Simons & Burt, 2011). The correlation between the parent and offspring reports of harsh parenting was $r = .30$, $p < .001$, for both the pretest and age 16 assessments.

Youths' epigenetic aging. When each participant was age 20 years, a certified phlebotomist went to the participant's home to draw a blood sample. The sample was frozen and delivered to the Psychiatric Genetics Lab at the University of Iowa, where peripheral blood mononuclear cells (PBMC) were

isolated by density-gradient centrifugation (Ficoll-Paque Media PM 400; GE Healthcare). Genomic DNA was extracted with Qiagen DNA Mini prep Kits, and quality verified on an Agilent 2100 Bioanalyzer. Following bisulfite conversion, genome-wide DNA methylation profiling was conducted with the Illumina (San Diego, CA, USA) HumanMethylation450 Beadchip at the University of Minnesota Genome Center, using the protocol specified by the manufacturer. The chip contains 485,577 probes recognizing at least 20,216 transcripts, potential transcripts, or CpG islands. Participants were randomly assigned to 12 sample slides, with groups of eight slides representing the samples from a single 96-well plate undergoing bisulfite conversion in a single batch. Eight replicates of the same DNA were also included to monitor slide-to-slide and batch-related bisulfite conversion variability. The resulting microarray data were inspected for complete bisulfite conversion; average beta value (i.e., average methylation) for each CpG residue was determined using the Illumina Genome Studio Methylation Module, Version 3.2 (Illumina). The beta value at a CpG locus is the ratio between the intensity of the methylated probe to the sum of intensities of the methylated and unmethylated probes, also known as the total probe intensities. The resulting data were then cleaned using a Perl-based algorithm to remove those beta values with detection p values, an index of the likelihood that the observed sequence represents random noise, greater than 0.05.

Epigenetic age was calculated from the DNA methylation data based on scripts developed by Horvath (2013). It reflects the disparity between an individual's biological and chronological ages. This procedure uses a mixed effect model implemented in the BMIQ package to conduct normalization of the relevant CpG methylation levels. Epigenetic age is predicted on the basis of the DNA methylation levels in regression coefficients obtained from numerous training sets. Discrepancies between epigenetic age and chronological age, in the form of residuals, were detected using a regression procedure in which epigenetic age was the outcome and chronological age was the independent variable. The residual served as the measure of epigenetic aging, which has a mean of 0 and represents both positive and negative deviations from chronological age in years.

Intervention status and gender. Intervention status and gender were dummy coded. SAAF participants were coded 1 and control participants were coded 0; male participants were coded 1 and female participants were coded 0.

Socioeconomic risk index. Six dichotomous variables formed a socioeconomic risk index that was used as a control in the data analyses. A score of 1 was assigned to each of the following: family poverty based on federal guidelines, primary caregiver unemployment, receipt of Temporary Assistance for Needy Families, primary caregiver single parenthood, primary caregiver education level less than high school graduation, and caregiver-reported inadequacy of family income. The scores were summed to form the index.

Lifestyle variables. To account for variables that could provide plausible rival explanations, all analyses controlled for adiposity, smoking, and unhealthful behaviors. Adiposity was operationalized as BMI assessed at the age 20 home visits. At the same visit, frequency of cigarette smoking was measured via youth self-report, on a seven-point scale ranging from *not at all* to *about two packs a day*. Unhealthful behavior was indexed using items from the Youth Risk Behavior Survey (Youth Risk Behavior Surveillance System, 2009). This scale has been used in several national, ethnically diverse surveys and has shown good validity and reliability. Participants reported how often during the past 7 days they consumed fruit, vegetables, 100% fruit juices, and milk. Exercise was measured with a single item: 'During the past 7 days, on how many days were you physically active for a total of at least 60 min per day?' The nutrition and exercise items were reverse coded so that higher numbers indicated less healthful behaviors.

Young adults' emotional distress. At 21 years of age, young adults reported their emotional distress using the well-validated Emotion Well-being subscale from the RAND 36-Item Short-Form Health Survey (Hays, Sherbourne, & Mazel, 1993). The subscale included five items assessing young adults' ratings of their emotional distress, on a response set ranging from 1 (none of the time) to 5 (most of the time); e.g., 'You have felt so down in the dumps that nothing could cheer you up,' and 'You have felt downhearted and blue.' Higher scores indicated more negative emotions and poorer mental health. After reverse scoring, all items were averaged to yield an emotional distress score with a range of 0 to 100 (α = .70).

Results

Does parental depression status when youths are age 11 forecast epigenetic aging at age 20?

Our initial analysis was designed to determine whether parental depression status forecast a higher epigenetic age among youths in the control condition. Presumably, youths in the control condition display normative associations between parental depression and epigenetic aging. Adjusting for gender; age 11 SES risk and harsh parenting; age 20 BMI, smoking, and unhealthful behaviors; and methylation batch assignment; a linear regression equation revealed a main effect of parental depression on epigenetic aging. Consistent with the first study hypothesis, youths in the high parental depressive symptoms group at age 11 evinced higher epigenetic ages at age 20 [Table 3, Model 1, b = 1.815, SE = 0.819, p = .028, 95% CI (0.196, 3.433)]. The results also were significant when depression report scores were treated as a continuous variable, b = 0.108, SE = 0.045, p = .017.

Can participation in prevention programming ameliorate risk for epigenetic aging?

Next, we tested the hypothesis that participation in SAAF would ameliorate the association between parental depressive symptoms and accelerated epigenetic aging. To do this, Model 2 in Table 3 added the multiplicative interaction term involving parental depressive symptoms and assignment to SAAF or the control condition. The analysis again revealed a main effect for depression symptom level and a significant interaction between depressive symptom level and SAAF participation, b = −2.046, SE = 1.027, p = .047, 95% CI [−4.066, −0.027]. When parental depression was treated as a continuous variable, the results were the same as when it was treated as a categorical variable, b = –.118, SE = .056, p < .05. To interpret this interaction, we conducted planned group comparisons to

Table 3 Parent depression status and intervention status as predictors of epigenetic aging

	PBMC Epigenetic Aging (age 20)					
	Model 1 (control group, $n = 157$)			Model 2 (whole sample, $N = 399$)		
Predictors	*B*	*SE*	β	*B*	*SE*	β
1. Gender, male	2.253	.780	.244**	2.102	.478	.225***
2. Family socioeconomic risk (age 11)	−.014	.246	−.005	.095	.160	.031
3. Harsh parenting (age 11)	−.186	.188	−.078	−.214	.114	−.091
4. Body mass index (age 20)	.031	.038	.064	.043	.027	.080
5. Smoking frequency (age 20)	−1.334	1.653	−.065	−.538	1.080	−.025
6. Health behaviors (age 20)	.062	.100	.048	−.044	.068	−.031
7. Parental depression group (age 11)	1.815	.819	.174*	1.896	.820	.183*
8. Intervention (SAAF)	–	–	–	.123	.563	.013
9. Parental depression × Intervention	–	–	–	−2.046	1.027	−.166*

PBMC, peripheral blood mononuclear cells; SAAF, Strong African American Families intervention program.
Analyses also controlled for batch assignment.
*$p < .05$, two-tailed.
**$p < .01$, two-tailed.
***$p < .001$, two-tailed.

test the hypothesis that youths whose parents reported high levels of depressive symptoms who had been assigned randomly to the control condition would show higher mean levels of epigenetic aging than would (a) similar youths assigned randomly to the SAAF condition, and (b) euthymia group youths assigned to either intervention condition. Figure 1A depicts the results of this analysis. The assumption of homogeneity of variance was met: $F(3, 394) = 0.365$, $p = .78$. The patterning of means conformed to the study prediction: Youths whose parents reported high levels of depressive symptoms who were assigned to the control condition showed accelerated epigenetic aging, with higher predicted epigenetic ages than youths in the other three groups, who did not differ from one another. To gauge the magnitude of this difference, we computed the effect size indicator Cohen's *d*. The *d* for the comparison was −0.41 for the mean difference between epigenetic aging for youths with a parent reporting high levels of depressive symptoms assigned to the control or SAAF condition, respectively. This *d* value indicated that, among SAAF participants, epigenetic aging was almost half a *SD* lower than that of controls.

SAAF-induced reductions in harsh parenting accounted for the program's effects on epigenetic aging among youths in the high depressive symptoms group

Next, we addressed the moderation-mediation hypothesis. First, we examined with planned comparisons whether *decreases* in harsh parenting from the pretest, when the participants were 11 years of age, to the long-term follow-up, when they were age 16, were lowest for youths in the control condition whose parents reported higher levels of depressive symptoms. The results of this analysis are presented in Figure 1B. The assumption of homogeneity of variance was met: $F(3, 377) = 0.747$, $p = .53$. This analysis showed that, although the use of harsh parenting declined for the sample as a whole, the parents with depression in the control condition evinced less of a decline than did parents in the other SAAF × parental depression combinations. The same results emerged when parental depression was treated as a continuous variable, $b = -.029$, $SE = .014$, $p < .05$.

The aforementioned results provided a basis for tests of the moderation-mediation hypothesis that SAAF-induced reductions in harsh parenting would account for the program's protective effects for youths whose parents reported high levels of depressive symptoms. This hypothesis was tested using a multigroup structural equation model with latent difference scores. The latent difference score reflected the degree to which harsh parenting changed from the time prior to SAAF implementation to the long-term follow-up at age 16. Regression coefficients were then calculated reflecting the associations between SAAF status and change in harsh parenting (Path A) and changes in harsh parenting and epigenetic aging (Path B) for parents in the euthymic group and in the high depressive symptoms group. The indirect or mediating effect of changes in harsh parenting was quantified as the product of these two regression coefficients (A × B). Nonparametric bootstrapping was used to obtain the bias corrected and accelerated confidence intervals of the indirect effect. Gender, family SES risk, BMI, smoking, unhealthful behaviors, and methylation batch were controlled in the model.

The results of this analysis were consistent with the hypothesis suggesting that SAAF deterred youths' epigenetic aging by reducing their exposure to harsh caregiving practices from high depressive symptoms group parents. Figure 2 depicts these findings. The negative coefficient for Path A (−0.516, $p < .05$ for high depressive symptoms

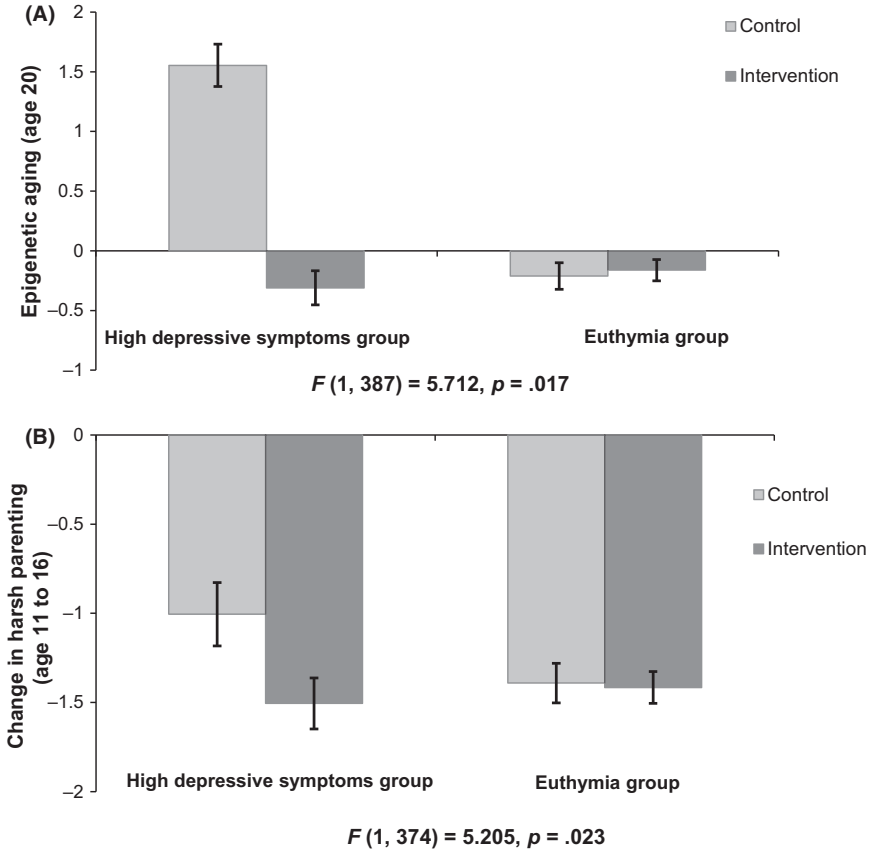

Figure 1 Means of (A) epigenetic aging at age 20 and (B) decrease in harsh parenting from age 11 to age 16 for the control and intervention groups by parent-reported depression status. High depressive symptoms group, n = 111: control group = 42, SAAF group = 69. Euthymia group, n = 288: control group = 115, SAAF group = 173. Error bars = ±1 standard error

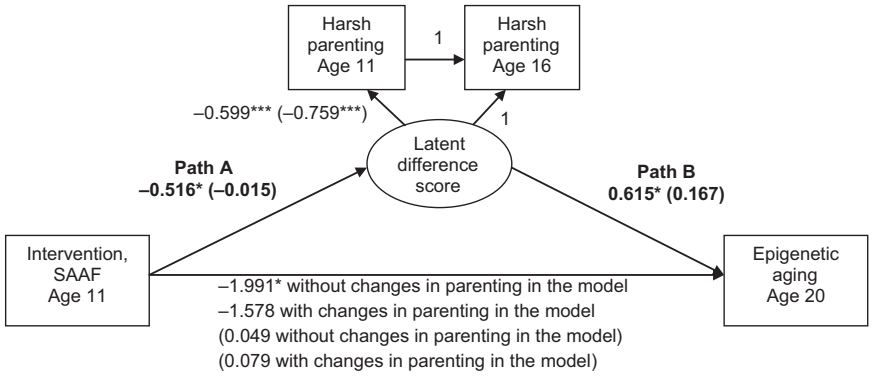

Figure 2 A moderated-mediation model of intervention status, changes in harsh parenting from age 11 to age 16, and epigenetic aging at age 20 for the high depressive symptoms group versus the euthymic group. Family socioeconomic-related risk, gender, BMI, smoking, unhealthful behaviors, and batch assignment were controlled (not shown). Unstandardized coefficients are presented. Numbers in parentheses refer to coefficients for the euthymic group. *p < .05, two-tailed. ***p < .001, two-tailed

group parents) indicates that participation in SAAF was associated with statistically significant declines in the use of harsh parenting. The path coefficient shown in parentheses for euthymic parents, as expected, was not significant. The positive coefficient for Path B (0.615, p < .05 for the high depressive symptoms group) indicates that, the more a parent reporting high depressive symptoms used harsh parenting from the pretest to the long-term follow-up, the higher the youth's predicted epigenetic age at age 20. Multiplying these coefficients yielded an indirect 'mediated' effect of −0.318, bootstrapped 95% CI [−0.922, −0.032]. The results were the same when parental depression was treated as a continuous variable. Consistent with the moderated-mediation hypothesis, these values indicate that the indirect pathway from SAAF to reductions in harsh parenting to relatively lower epigenetic ages among depression group youths was statistically significant. Overall model fit was good, with $\chi^2(46) = 47.41$, p = .41, comparative fit index = .99, and root mean square error of approximation = .01 [95% CI 0, .05].

We also tested models in which SAAF worked through changes in parents' and youths' depression, youth self-control, and youth externalizing problems, regardless of changes in harsh parenting. No evidence emerged to support these models.

Exploratory analyses

The aforementioned findings highlighted the long-term negative effects of maternal depression on accelerated biological aging for those youths who did not participate in SAAF. The analyses presented here were designed to extend these findings by exploring the notions that (a) maternal depression status, when the study participants were 11 years of age, forecast offspring reports of emotional distress at age 22, (b) epigenetic aging at 20 years of age forecast self-reports of emotional distress one year later, at age 21, and (c) maternal depression is associated with offspring emotional distress in young adulthood, via its association with accelerated epigenetic aging for participants assigned randomly to the control condition but *not* the SAAF condition. These questions were explored using a multigroup structural equation model. Maternal depression at 11 years of age was an exogenous construct, not predicted by any prior variable in the model, and epigenetic aging at 20 years of age and emotional distress at 21 years of age were specified as endogenous constructs. The results of this analysis for the *control* group indicated that young adult emotional distress at age 21 was forecast by parental depression [$b = 6.013$, $SE = 3.10$, $p = .05$, 95% CI (0, 12.095)] and epigenetic aging at age 20 [$b = 2.204$, $SE = 0.827$, $p = .008$, 95% CI (0.574, 3.3791)]. Consistent with the reported results, parental depression forecast accelerated epigenetic aging, $b = 0.719$, $SE = 0.289$, $p = .013$, 95% CI (0.202, 1.366). Multiplying the latter two coefficients yielded an indirect 'mediated' effect of epigenetic aging on the link between parental depression status and young adult emotional distress. The indirect effect was calculated 1,000 times using random replacement to build a sampling distribution. The indirect effect was 1.585, bootstrapped 95% CI (0.317, 3.821) for participants in the control condition.

None of these path coefficients were significant for participants in the SAAF condition: parental depression status and young adult emotional distress, $b = -0.610$, $SE = 2.70$, $p = .821$, 95% CI [−5.903, 4.682]; parental depression status and epigenetic aging, $b = -0.200$, $SE = 0.623$, $p = .748$, 95% CI [−1.127, 1.357], epigenetic aging and emotional distress, $b = -0.081$, $SE = 0.265$, $p = .761$, 95% CI [−0.653, 0.392]. Together, the exploratory analyses suggest that accelerated epigenetic aging may be one risk pathway by which parental depression has consequences for offspring mental health. From a resilience perspective, these links were ameliorated for youths who participated in the SAAF prevention program.

Discussion

Children who grow up with a parent who reports relatively high levels of depressive symptoms are at heightened risk for impaired emotion regulation, dysregulation of stress hormone systems, and weathering of their bodies. Although studies suggest that exposure to harsh parenting may be implicated in these processes (Miller et al., 2011; Repetti et al., 2002), they were not designed to permit inferences about causality or clinical utility. We tested this hypothesis by conducting a secondary analysis of data from a family-oriented preventive intervention to determine whether youths in the high caregiver depressive symptoms group assigned to the control condition would evince the highest levels of accelerated epigenetic aging relative to similar youths in the intervention group and to euthymic group youths. The results supported this hypothesis and, importantly, moderated-mediation analyses demonstrated that intervention effects on reductions in harsh parenting accounted for the association between parental depression levels and slower epigenetic aging among offspring. Embedding the assessment of epigenetic aging in a prevention trial increases confidence in the causal nature of the linkages between harsh parenting and accelerated epigenetic aging.

Future research should elucidate the pathways that link harsh parenting to accelerated aging. We considered the possibility that SAAF rendered youths less vulnerable to obesity, smoking and the use of other drugs, depressive symptoms, externalizing problems, decrements in self-control, and general life stress. None of these variables forecast accelerated epigenetic aging. Another approach for future studies would be to focus on neuroendocrine pathways. Exposure to harsh parenting can occasion persistent activation of the stress-response systems, in particular the sympathetic nervous system and the HPA axis. The hormonal products of these systems, glucocorticoids and catecholamines, are elevated in youths who experience high levels of harsh parenting (Brody, Yu, Chen, & Miller, 2014). These hormones can affect DNA methylation of cells by altering the enzymatic activity of DNA methyltransferases, as well as histone deacetylases and acetyltransferases (Barnes & Adcock, 2009).

The exploratory analyses extend previous research by showing that offsprings' experience relatively early in life with parental depression can contribute to accelerated aging and mental health outcomes. This is the first demonstration, to our knowledge, of a prospective association between accelerated epigenetic aging and an indicator of emotional distress. The exploratory results also indicate that a good deal of plasticity exists in epigenetic aging, as evidenced by the protective effects of participation in SAAF. These results suggest several missing pieces that should be addressed but are beyond the scope of this

study. First, what are the biological processes that are affected by parental depression and by SAAF that presage accelerated epigenetic aging? Analyses of genome-wide methylation in greater detail will begin to provide answers to this question. The relationship between accelerated aging and emotional distress doubtless is bidirectional. Future research should use repeated measures to examine accelerated aging and emotional distress. Such research will help to clarify the roles of parental depression and epigenetic aging in predicting later mental health distress.

A hospitable feature of SAAF was its delivery by nonstigmatizing African American community members who are familiar with and who, themselves, identify with life in challenging communities in the rural South (Brody et al., 2012). Whereas prior research has established the efficacy of standardized depression treatments among minority group women in poverty, the difficulty that researchers have had in engaging women in care led them to conclude that 'engaging them through trusted providers could prove easier' (Miranda et al., 2003, p. 64). Indeed, the present data illustrate how a conveniently delivered family skill-building program facilitated by nonstigmatizing rural African American community members created health benefits that were evident in the cells and tissues of at-risk adolescents.

Several limitations of the trial must be noted. The SAAF trial was not designed with epigenetic aging as an endpoint. As a result, we did not collect pretrial DNA that could be used to determine whether the prevention and control groups' epigenetic aging profiles changed differentially over time. Until pretest and posttest data are available, conclusions about SAAF's ability to bring about changes in epigenetic aging must be viewed as suggestive. We also must consider the possibility that the lack of an association between maternal depression and epigenetic age acceleration observed in the SAAF group could be a false positive. A replication, of course, will be required to address this possibility. Future research should also examine the contribution of maternal depression that offspring experience before 11 years of age to epigenetic aging. Family emotional climates are particularly important to the well-being of young children. The sample was composed of African Americans living in the rural South. The findings' generalizability must be documented with other groups, living in rural or urban settings. These limitations notwithstanding, to the extent the present findings are replicated, the results may provide a strategy for reducing the health vulnerabilities of youths who have a parent with elevated levels of depressive symptoms.

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Key points

- Hypotheses were tested to determine whether parents' elevated levels of depressive symptoms and exposure to harsh parenting forecast youth epigenetic aging.
- Data were obtained from 399 youths participating in a family-centered prevention trial.
- The youths were African American and lived in a rural region of the southeastern United States.
- Parental depressive symptoms and harsh parenting forecast accelerated epigenetic aging.
- Prevention program effects completely ameliorated the risks that parental depressive symptoms and harsh parenting conferred on accelerated epigenetic aging.

References

Barnes, P.J., & Adcock, I.M. (2009). Glucocorticoid resistance in inflammatory diseases. *Lancet*, *373*, 1905–1917.

Brody, G.H., Ge, X., Conger, R.D., Gibbons, F.X., Murry, V.M., Gerrard, M., & Simons, R.L. (2001). The influence of neighborhood disadvantage, collective socialization, and parenting on African American children's affiliation with deviant peers. *Child Development*, *72*, 1231–1246.

Brody, G.H., Kogan, S.M., & Grange, C.M. (2012). Translating longitudinal, developmental research with rural African American families into prevention programs for rural African American youth. In R.B. King & V. Maholmes (Eds.), *The Oxford handbook of poverty and child development* (pp. 553–570). New York, NY: Oxford University Press-USA.

Brody, G.H., Murry, V.M., Gerrard, M., Gibbons, F.X., Molgaard, V., McNair, L.D., … & Neubaum-Carlan, E. (2004). The Strong African American Families program:

Translating research into prevention programming. *Child Development*, *75*, 900–917.
Brody, G.H., Yu, T., Chen, E., & Miller, G.E. (2014). Prevention moderates associations between family risks and youth catecholamine levels. *Health Psychology*, *33*, 1435–1439.
Cicchetti, D., & Rogosch, F.A. (2009). Adaptive coping under conditions of extreme stress: Multilevel influences on the determinants of resilience in maltreated children. In E.A. Skinner & M.J. Zimmer-Gembeck (Eds.), *New directions for child and adolescent development: No. 124. Coping and the development of regulation* (pp. 47–59). San Francisco, CA: Jossey-Bass.
Essex, M.J., Shirtcliff, E.A., Burk, L.R., Ruttle, P.L., Klein, M.H., Slattery, M.J., ... & Armstrong, J.M. (2011). Influence of early life stress on later hypothalamic-pituitary-adrenal axis functioning and its covariation with mental health symptoms: A study of the allostatic process from childhood into adolescence. *Development and Psychopathology*, *23*, 1039–1058.
Field, T. (1994). The effects of mother's physical and emotional unavaiability on emotion regulation. *Monographs of the Society for Research in Child Development*, *59*, 208–227.
Geronimus, A.T., Hicken, M., Keene, D., & Bound, J. (2006). 'Weathering' and age patterns of allostatic load scores among Blacks and Whites in the United States. *American Journal of Public Health*, *96*, 826–833.
Halligan, S.L., Herbert, J., Goodyer, I.M., & Murray, L. (2007). Disturbances in morning cortisol secretion in association with maternal postnatal depression predict subsequent depressive symptomatology in adolescents. *Biological Psychiatry*, *62*, 40–46.
Hays, R.D., Sherbourne, C.D., & Mazel, R.M. (1993). The RAND 36-item health survey 1.0. *Health Economics*, *2*, 217–227.
Hobfoll, S.E., Ritter, C., Lavin, J., Hulsizer, M.R., & Cameron, R.P. (1995). Depression prevalence and incidence among inner-city pregnant and postpartum women. *Journal of Consulting and Clinical Psychology*, *63*, 445–453.
Horvath, S. (2013). DNA methylation age of human tissues and cell types. *Genome Biology*, *14*, R115.
Horvath, S., Erhart, W., Brosch, M., Ammerpohl, O., von Schönfels, W., Ahrens, M., ... & Hampe, J. (2014). Obesity accelerates epigenetic aging of human liver. *Proceedings of the National Academy of Sciences of the USA*, *111*, 15538–15543.
Marioni, R.E., Shah, S., McRae, A.F., Chen, B.H., Colicino, E., Harris, S.E., ... & Deary, I.J. (2015). DNA methylation age of blood predicts all-cause mortality in later life. *Genome Biology*, *16*, 25.
Mayberry, L.J., Horowitz, J.A., & Declercq, E. (2007). Depression symptom prevalence and demographic risk factors among U.S. women during the first 2 years postpartum. *Journal of Obstetric, Gynecologic, and Neonatal Nursing*, *36*, 542–549.
Miller, G.E., Chen, E., & Parker, K.J. (2011). Psychological stress in childhood and susceptibility to the chronic diseases of aging: Moving toward a model of behavioral and biological mechanisms. *Psychological Bulletin*, *137*, 959–997.
Miranda, J., Chung, J.Y., Green, B.L., Krupnick, J., Siddique, J., Revicki, D.A., & Belin, T. (2003). Treating depression in predominantly low-income young minority women: A randomized controlled trial. *Journal of the American Medical Association*, *290*, 57–65.
Radloff, L.S. (1977). The CES–D Scale: A self-report depression scale for research in the general population. *Applied Psychological Measurement*, *1*, 385–401.
Repetti, R.L., Taylor, S.E., & Seeman, T.E. (2002). Risky families: Family social environments and the mental and physical health of offspring. *Psychological Bulletin*, *128*, 330–336.
Rutter, M.L. (2005). Environmentally mediated risks for psychopathology: Research strategies and findings. *Journal of the American Academy of Child and Adolescent Psychiatry*, *44*, 3–18.
Shonkoff, J.P., Boyce, W.T., & McEwen, B.S. (2009). Neuroscience, molecular biology, and the childhood roots of health disparities: Building a new framework for health promotion and disease prevention. *Journal of the American Medical Association*, *301*, 2252–2259.
Simons, R.L., & Burt, C.H. (2011). Learning to be bad: Adverse social conditions, social schemas, and crime. *Criminology*, *49*, 553–598.
Weissman, M.M., Wickramaratne, P., Nomura, Y., Warner, V., Pilowsky, D., & Verdeli, H. (2006). Offspring of depressed parents: 20 years later. *American Journal of Psychiatry*, *163*, 1001–1008.
Youth Risk Behavior Surveillance System. (2009). *Youth Risk Behavior Survey*. Retrieved from http://www.cdc.gov/healthyyouth/yrbs/questionnaire_rationale.htm.

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