Effects of Maternal Negative Parenting, Peer Victimization, and FKBP5 Gene on Adolescent Depression

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Abstract

Both the cumulative stress hypothesis and the match-mismatch hypothesis can account for the effects of distal and proximal adversity on individual depression, yet few studies have examined the moderating role of genetic factors therein. Employing questionnaire methods and DNA genotyping technology, a three-year longitudinal investigation was conducted with 970 adolescents. Maternal negative parenting and peer victimization were utilized as distal and proximal stress indicators, respectively, while the FKBP5 gene multi-locus additive score served as the genetic index, to investigate the three-way interaction among these factors on adolescent depression and gender differences. The results indicated that among male adolescents, the E × E × G interaction was significant. When the additive score was high and peer victimization levels were elevated, maternal negative parenting significantly negatively predicted depression, aligning with the match-mismatch hypothesis; when the additive score was low, the E × E interaction was not significant but tended to function in accordance with the cumulative stress hypothesis. Among female adolescents, the E × E × G interaction was not significant. The findings suggest that among male adolescents, both the cumulative stress and match-mismatch hypotheses can elucidate the mechanisms underlying depression, being applicable to individuals carrying different FKBP5 gene multi-locus additive scores.

Full Text

The Influence of Maternal Negative Parenting, Peer Victimization, and FKBP5 Gene on Adolescent Depression

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Abstract

Both the cumulative stress hypothesis and the match-mismatch hypothesis can explain how distal and proximal adversities influence individual depression, yet few studies have examined the moderating role of genetic factors. Using questionnaire methods and DNA genotyping technology, this study conducted a three-year longitudinal investigation of 970 adolescents. With maternal negative parenting and peer victimization as distal and proximal stress indicators, respectively, and a multi-locus additive score of the FKBP5 gene as the genetic index, we examined the three-way interaction among these factors on adolescent depression and its gender differences. Results revealed a significant three-way interaction (E × E × G) among male adolescents. For individuals with higher additive scores and higher peer victimization, maternal negative parenting significantly negatively predicted depression, supporting the match-mismatch hypothesis. When additive scores were lower, the E × E interaction was not significant but tended to function in accordance with the cumulative stress hypothesis. Among female adolescents, the E × E × G interaction was not significant. These findings suggest that both the cumulative stress and match-mismatch hypotheses can elucidate depression mechanisms in male adolescents, applying to individuals carrying different FKBP5 multi-locus additive scores.

Keywords: maternal negative parenting; peer victimization; FKBP5 gene; adolescent depression; cumulative stress hypothesis; match-mismatch hypothesis
Classification Codes: B844; B845

Depression represents one of the most prevalent mental health problems during adolescence. Extensive research demonstrates that stressful life events constitute important predictors of adolescent depression, which can be categorized based on timing into distal stress (occurring early in life) and proximal stress (occurring recently). Distal and proximal stressors interact in complex ways to influence depression onset and development. Currently, two competing models explain these interaction patterns: the cumulative stress hypothesis rooted in traditional psychopathology (McEwen, 1998; Vinkers et al., 2014) and the match-mismatch hypothesis grounded in biological evolutionary theory (Nederhof, 2012; Nederhof et al., 2014).

The cumulative stress hypothesis posits that distal adverse experiences increase individuals' vulnerability to stress and, through accumulation with proximal stress, lead to increased allostatic load, such as wear and tear on stress systems like the HPA axis, ultimately elevating depression risk. Conversely, when individuals experience lower overall stress (both distal and proximal), impairments to physiological, psychosocial, and cognitive functions remain relatively minor, resulting in lower depression risk (Brown et al., 2008; McEwen, 1998; Myers et al., 2015). For instance, research has found that individuals experiencing high levels of both childhood adversity and adult stressful life events exhibit higher depression levels than others (McLaughlin et al., 2010; Power et al., 2013; Shapero et al., 2013).

However, distal and proximal stress do not inevitably lead to maladjustment. The match-mismatch hypothesis suggests that moderate distal stress can promote the formation of adaptive phenotypic plasticity, thereby preparing the organism to cope effectively with similar (matching) stress environments that reoccur during development. Consequently, depression levels are lower when individuals experience matching distal and proximal stress; under mismatched conditions, individuals cannot utilize coping strategies established based on distal environments to face subsequent different (mismatched) experiences, ultimately increasing depression risk (Frankenhuis & Del Giudice, 2012; Gluckman et al., 2007; Nederhof et al., 2014; Schmidt, 2011). Recent animal and human studies have provided empirical support for this hypothesis (Daskalakis et al., 2012; Santarelli et al., 2014). For example, research shows that rats experiencing early maternal separation demonstrate better memory performance following proximal stress stimulation compared to rats without such separation (Zalosnik et al., 2014). Human studies also indicate that adolescents experiencing high childhood adversity have lower depression risk following high proximal stressful life events compared to those with low childhood adversity (Oldehinkel et al., 2014).

These two models diverge regarding how distal and proximal stress influence depression risk. However, recent evidence suggests they are not entirely opposing, and the nature of their interaction depends on individuals' innate genetic predispositions. Nederhof and Schmidt (2012) proposed an integrated model wherein individuals carrying sensitive genotypes are more susceptible to distal stress and consequently develop adaptive phenotypes, following a match-mismatch developmental pattern. Conversely, individuals with insensitive genotypes, unable to develop adaptive coping strategies, accumulate negative effects as stress levels increase, elevating depression risk. In other words, the applicability of the cumulative stress versus match-mismatch hypotheses depends on genetic makeup. Nevertheless, examination and validation of this assumption remain scarce, primarily derived from animal studies. For instance, van der Doelen et al. (2013) found that in male mice carrying 5-HTT+/- (functionally similar to the human 5-HTTLPR S allele), early-life stress and adult stress influenced depressive-like behavior in a match-mismatch pattern, while the distal-proximal stress interaction was not significant in other genotypes. To our knowledge, only one human study has attempted to test these models, using 5-HTTLPR polymorphism as the genetic index with childhood maltreatment and adult stressful life events as distal and proximal stressors, respectively, finding no significant three-way interaction with depression (Power et al., 2013). The non-significant E $\times$ E $\times$ G effect in that study may be attributable to the low explanatory power of single candidate gene loci. As Nederhof and Schmidt (2012) noted, lower genetic susceptibility reduces statistical power for testing theoretical models, and future research should incorporate multiple genes or multiple loci within genes.

The HPA axis system plays a crucial role in stress and depression development (Cai et al., 2015; Menke, 2019; Normann & Buttenschøn, 2019). Therefore, genetic variations regulating HPA axis function may represent important can-

didate genes moderating the stress-depression association. The FKBP5 gene (FK506 binding protein 5), located on chromosome 6p21.31, has received considerable attention. This gene regulates FKBP5 protein expression, which can bind to glucocorticoid receptors (GR) and competitively inhibit binding between glucocorticoids (GCs, primarily cortisol) and GR. Consequently, FKBP5 protein overexpression can lead to GR insensitivity and elevated cortisol levels (Tyrka et al., 2015; van Bodegom et al., 2017), subsequently impairing HPA axis negative feedback mechanisms (Tyrka et al., 2015), causing hippocampal atrophy (Frodl & O' Keane, 2013; Sapolsky, 2000), and ultimately triggering depression.

To date, research on FKBP5 gene interactions with stressful life events in depression has primarily adopted a single-locus approach. However, associations between single loci and most phenotypes are minimal (Nelemans et al., 2019), and single-locus × environment interaction research suffers from limited replicability (Duncan & Keller, 2011). As accumulating evidence demonstrates that depression has a polygenic or multi-locus genetic basis (Flint & Kendler, 2014; Mullins et al., 2015; Stocker et al., 2017), researchers have begun employing cumulative genetic score (CGS) methods to explain the cumulative genetic effects of relevant candidate genes on depression (e.g., Belsky et al., 2015; Stocker et al., 2017).

substantial empirical evidence indicates that FKBP5 gene polymorphisms rs1360780, rs3800373, and rs9296158 all moderate environmental sensitivity (Lavebratt et al., 2010; Piechaczek et al., 2019; Wang et al., 2018; Zimmermann et al., 2011), located in intron 2, the 3' untranslated region, and intron 5, respectively. Previous research shows that carriers of minor alleles including rs1360780 T, rs3800373 C, and rs9296158 A exhibit higher FKBP5 expression levels (Calabrò et al., 2019; White et al., 2012) and greater depression risk following adversity (Calabrò et al., 2019; Wang et al., 2018). Recent neuroimaging research found that a cumulative score of 10 HPA axis system genes including FKBP5, CRHR1, NR3C2, and NR3C1 moderated amygdala and hippocampal responses to fear-neutral facial stimuli in adolescents, responses closely associated with depression (Pagliaccio et al., 2015). Therefore, this study selected FKBP5 gene polymorphisms rs1360780, rs3800373, and rs9296158 to examine multi-locus genetic effects within a single gene, exploring whether the conditions for cumulative stress and match-mismatch hypotheses depend on individual genetic makeup.

Regarding distal and proximal stress for adolescent depression, extensive research shows that during childhood, parent-child relationships dominate children' s interpersonal worlds (Furman & Buhrmester, 1992; Helsen et al., 2000), with parenting representing an important predictor of depression (McLeod et al., 2007; Yap & Jorm, 2015). Low positive or supportive parenting and high negative parenting predict increased depression risk (Schleider & Weisz, 2017). During adolescence, peer networks expand (Prinstein & La Greca, 2002), peer interactions become more frequent (Buhrmester & Furman, 1987), and peers become more important sources of social support and belonging (Steinberg, 2014),

making peer victimization a significant predictor of depression during this period (Adrian et al., 2019; Schoeler et al., 2018). Moreover, research indicates that harsh parenting explains more variance in childhood depression than peer victimization (Bilsky et al., 2013; Cole et al., 2015). Based on these findings, this study employed childhood maternal negative parenting and adolescent peer victimization as distal and proximal adversity indicators, respectively.

Numerous studies demonstrate gender differences in gene-environment interactions on depression and related neural structures and functions (Chang et al., 2017; Pagliaccio et al., 2015; Wang et al., 2019). FKBP5 is a chaperone protein for progesterone and androgen receptor complexes (Zannas & Binder, 2013), and its expression and interactions with the environment may be influenced by sex hormones, resulting in gender differences. For instance, research shows that the interaction between rs1360780 polymorphism and negative life experiences on depression is significant only in males (Lavebratt et al., 2010). Comasco et al. (2015) also found that FKBP5 polymorphisms rs1360780 and rs3800373 interact more significantly with early-life adversity to predict depression in male versus female adolescents. Additionally, previous research indicates that gender is closely associated with levels of negative parenting and peer victimization, with males potentially experiencing more negative parenting (Keshavarz & Mounts, 2017; Tenenbaum & Leaper, 2003) and peer victimization (Crick & Bigbee, 1998) than females, and the variability of environmental variables influences gene-environment interactions (Belsky & Beaver, 2011; Wang et al., 2019). Therefore, examining the three-way interaction among FKBP5 multi-locus additive score, maternal negative parenting, and peer victimization separately by gender is warranted.

Given that no previous research has examined gender patterns in cumulative stress and match-mismatch hypotheses, we refrain from making explicit hypotheses regarding gender differences.

In summary, to explore the mechanisms underlying adolescent depression, this study investigates the interactive effects of maternal negative parenting, peer victimization, and FKBP5 multi-locus additive score on adolescent depression, testing which theoretical model (cumulative stress vs. match-mismatch) best explains the observed patterns. This research extends cumulative stress and match-mismatch hypotheses into the multi-locus genetic domain and explores gender differences, contributing to a deeper understanding of adolescent depression pathogenesis and providing more specific and targeted recommendations for intervention practice.

2.1 Participants

Participants were drawn from a large-scale longitudinal project. This genetic study was independently conducted using the database from that project, aiming to examine the effects of distal and proximal stress on adolescent depression, thus employing a three-year longitudinal design. At T1 (sixth grade), maternal

negative parenting and adolescent depressive symptoms were assessed (N = 2,114, Mage = 12.31 ± 0.47 years, 51.7% male). At T2 (ninth grade), adolescent depression was reassessed and peer victimization data were collected. The three-year interval between assessments resulted in 291 participants lost to attrition (13.8%). Attrition analyses revealed no significant differences between retained and lost participants in T1 age (t(2091) = 0.04, p = 0.97) or T1 socioeconomic status (SES) (t(2112) = -0.69, p = 0.49). However, lost participants reported higher maternal negative parenting (t(2112) = 2.23, p = 0.03) and depression levels (t(2112) = 3.75, p < 0.001) at T1 and were more likely to be male (^{2} = 6.18, df = 1, p = 0.01). Demographic characteristics and statistical results for T1 and T2 are presented in Table 1 .

At T2, participants were distributed across 39 classes in 14 schools. Previous research indicates that gene-environment interactions typically yield effect sizes of 0.01-0.02 when reaching significance (e.g., Starr et al., 2014; Wang et al., 2019). Based on this range, power analysis using G*Power 3.1.9.2 indicated that approximately 395-787 participants were needed to achieve 80% statistical power ($\alpha = 0.05$). However, due to attrition, only 970 participants had genetic data available, including 473 males (48.8%) with a mean age of 15.32 years (SD = 0.47).

At T2, participants with genetic data (N = 970) did not differ significantly from those without (N = 853) in gender (^{2} = 2.73, df = 1, p = 0.09), age, T1 SES, T1 maternal negative parenting, T2 peer victimization, or T2 depression (|ts| < 1.66, ps > 0.05). However, participants with genetic data had lower T1 depression scores (t(1821) = 2.23, p = 0.03). This study included only adolescents who participated in genotyping. The study was approved by the ethics committee of ×× University.

Table 1 Sample Characteristics

Variable	T1 (N = 2,114)	T2 (N = 1,823)	Test Statistics
Gender (male)	1,092 (51.7%)	922 (50.6%)	^{2} = 0.46, df = 1, p = 0.50
Age (years)	12.31 ± 0.47	15.31 ± 0.47	t(3895) = -0.69, p = 0.99

Note: SES was calculated following Akkoyun-Farinez et al. (2018).

2.2 Measures

2.2.1 Adolescent Depression Depressive symptoms were assessed using the Children' s Depression Inventory (CDI) (Kovacs, 1992), which has been widely used in depression research, particularly in non-clinical samples, and demonstrates good reliability and validity (Wu et al., 2012; Zhang et al., 2016). The CDI comprises 27 items assessing depressive symptoms over the past two weeks

(e.g., "unhappy," "pessimistic"). Items are rated on a 3-point scale from 0 to 2, representing "sometimes," "often," and "always," with higher scores indicating greater depression severity. In this study, Cronbach' s α coefficients were 0.87 and 0.89 at T1 and T2, respectively, with mean scores of 0.18 (SD = 0.21) and 0.27 (SD = 0.25).

2.2.2 Maternal Negative Parenting Maternal negative parenting was assessed using the Chinese version of the Child-Rearing Practices Report (CRPR) (Chen et al., 2010), completed by mothers. This questionnaire demonstrates good psychometric properties in Chinese child and adolescent samples (Chen et al., 2002). Maternal negative parenting includes two dimensions: "rejection" (4 items, e.g., "I often forget things I should do for my child") and "punishment" (7 items, e.g., "I believe physical punishment is the best way to discipline a child"). Items are rated on a 5-point scale from 0 ("completely uncharacteristic") to 4 ("completely characteristic"), with higher scores indicating higher levels of negative parenting. Cronbach' s α was 0.65 in this study, with a mean of 1.17 (SD = 0.44).

2.2.3 Peer Victimization Peer victimization was measured using the Chinese revised version of the Multidimensional Peer Victimization Scale (MPVS; Mynard & Joseph, 2000), which demonstrates good reliability and validity (Ji et al., 2011). The scale comprises three dimensions: "physical victimization" (3 items, e.g., "This semester, other students threatened to hit me"), "verbal victimization" (3 items, e.g., "This semester, other students called me stupid"), and "relational victimization" (8 items, e.g., "This semester, other students turned people against me"). Adolescents self-reported on a 4-point scale from 0 ("never happened") to 3 ("happened often"), with higher scores indicating greater victimization. Cronbach' s α was 0.92 in this study, with a mean of 0.33 (SD = 0.41).

2.3 Genetic Testing

DNA was extracted from saliva samples. Polymorphisms rs1360780, rs3800373, and rs9296158 were genotyped using the Sequenom (San Diego, CA, USA) chip-based matrix-assisted laser desorption/ionization time-of-flight (MALDI-TOF) mass spectrometry platform. Genotyping results were read in real-time by MassARRAY RT 3.0.0.4 software and analyzed by MassARRAY Typer 3.4 software. Quality control procedures included duplicate checks (5%), negative controls, and minor allele frequency checks, with a genotyping success rate exceeding 97%.

2.4 Procedure

Prior to data collection, informed consent was obtained from adolescents, their parents, and schools. Adolescents' depression and peer victimization data were

collected in classrooms with school cooperation, while mothers completed questionnaires at school to report their parenting practices. Trained graduate students collected saliva samples in classrooms after instructing participants to avoid eating, smoking, drinking, or chewing gum for 30 minutes prior to collection. The ratio of experimenters to adolescents was at least 1:8, with collection taking approximately 30 minutes. Samples were then sent to a biotechnology company for DNA extraction, purification, and genotyping.

2.5 Data Processing and Analysis

Data were processed and analyzed using SPSS 22.0. Pearson product-moment correlations examined associations among study variables. Prior to correlation analysis, FKBP5 polymorphisms were coded. Given that minor allele carriers of FKBP5 show higher protein expression levels (Calabrò et al., 2019; White et al., 2012) and greater depression risk following adversity, while major allele carriers show lower depression levels (Calabrò et al., 2019; Wang et al., 2018), we hypothesized that major alleles are protective. Individuals carrying more major alleles may be more likely to develop adaptive stress coping strategies following distal adversity, consistent with characteristics of susceptible individuals in Nederhof and Schmidt' s (2012) integrated model. Based on this rationale and following previous research (Kang et al., 2012), we coded the three FKBP5 loci based on minor allele count: rs1360780: TT = 0, CT = 1, CC = 2; rs3800373: CC = 0, CA = 1, AA = 2; rs9296158: AA = 0, AG = 1, GG = 2. A multi-locus additive score was calculated by summing across loci (males: $N_0 = 30$, $N_1 = 1$, $N_2 = 11$, $N_3 = 171$, $N_4 = 11$, $N_5 = 42$, $N_6 = 207$; females: $N_0 = 20$, $N_1 = 4$, $N_2 = 9$, $N_3 = 170$, $N_4 = 9$, $N_5 = 38$, $N_6 = 247$). Few participants had scores of 1, 2, or 4, so following previous research (Beaver & Belsky, 2012; Cicchetti & Rogosch, 2012; Wade et al., 2015), individuals with scores of 0, 1, and 2 were combined into one group (coded as 1), those with score 3 were coded as 2, those with scores 4 and 5 were combined as 3, and those with score 6 were coded as 4. The final sample included 42/33, 171/170, 53/47, and 207/247 male/female participants with CGS of 1, 2, 3, and 4, respectively, with higher scores indicating greater genetic sensitivity.

To test the cumulative stress and match-mismatch hypotheses, hierarchical regression analyses were conducted separately by gender with T2 depression as the outcome. Predictors were entered as follows: (1) T1 depression as a control variable in the first block; (2) CGS (G), T1 maternal negative parenting (E_1), and T2 peer victimization (E_2) in the second block; (3) G × E_1, G × E_2, and E_1 × E_2 in the third block; and (4) E_1 × E_2 × G in the fourth block. Maternal negative parenting and peer victimization were standardized prior to analysis, and Bootstrap resampling with 10,000 iterations was employed. To control Type I error rates, the Benjamini–Hochberg (1995) procedure was applied to correct regression results for multiple comparisons. Given attrition and gender-stratified analyses reducing sample size and potentially compromising statistical power, internal consistency analysis (see Wang et al., 2020) was used to further verify

findings. Additionally, to ensure reliability of the multi-locus additive score, analyses were conducted using single FKBP5 loci and cumulative scores of any two loci as genetic indices to examine $E_1 \times E_2 \times G$ effects, with linear gene effect testing performed (see Cao & Zhang, 2019; Stocker et al., 2017).

3. Results

3.1 Descriptive Analyses

Genotype distributions for the three FKBP5 loci were: rs1360780: CC = 55.9% (542 individuals), CT = 38.5% (374 individuals), TT = 5.6% (54 individuals); rs3800373: AA = 56.4% (547 individuals), CA = 38.1% (370 individuals), CC = 5.5% (53 individuals); rs9296158: GG = 47.3% (459 individuals), AG = 44.5% (432 individuals), AA = 8.2% (79 individuals). Observed and expected values showed good fit (rs1360780: ^{2} = 1.02, df = 2, p = 0.31; rs3800373: ^{2} = 0.88, df = 2, p = 0.35; rs9296158: ^{2} = 2.64, df = 2, p = 0.10), all conforming to Hardy-Weinberg equilibrium.

Means, standard deviations, and correlations among main variables are presented in Table 2 . Independent samples t-tests revealed that male adolescents reported significantly higher maternal negative parenting (t(968) = 3.17, p = 0.002) and peer victimization (t(968) = 6.63, p < 0.001) than females, justifying gender-stratified analyses. The FKBP5 multi-locus additive score was not significantly correlated with maternal negative parenting or peer victimization, ruling out gene-environment correlation. Both maternal negative parenting and peer victimization showed significant positive correlations with T1 and T2 depression, leading us to control for T1 depression in subsequent analyses. The two depression assessments were significantly positively correlated, with paired samples t-test indicating significantly higher T2 than T1 depression (t(969) = -11.49, p < 0.001).

Table 2 Correlations Among Variables

Variable	T1 Maternal Negative Parenting	T2 Peer Victimization	T1 Depression	T2 Depression
T1 Maternal Negative Parenting	–	0.15** / 0.12**	0.11* / 0.34***	0.12** / 0.15**
T2 Peer Victimization	0.24*** / 0.24***	–	0.19*** / 0.39***	0.48*** / 0.49***

Variable	T1 Maternal Negative Parenting	T2 Peer Victimization	T1 Depression	T2 Depression
T1 Depression	0.19*** / 0.24***	0.39*** / 0.48***	–	0.42*** / 0.45***
T2 Depression	0.22*** / 0.26***	0.27*** / 0.26***	0.49*** / 0.48***	–

Note: $p < 0.05$, ***p < 0.01,*** $p < 0.001$. Correlations below the diagonal are for males, above for females. Values before/after slash represent male/female information. CGS = cumulative genetic score.

3.2 Effects of FKBP5 Multi-Locus Additive Score, Maternal Negative Parenting, and Peer Victimization on Adolescent Depression

As shown in Tables 3 and 4 , after controlling for T1 depression, main effects of FKBP5 multi-locus additive score and maternal negative parenting were non-significant, while peer victimization significantly positively predicted T2 depression in both males and females. Crucially, a significant three-way interaction among maternal negative parenting, peer victimization, and CGS emerged only for male adolescents. Specifically, among males with CGS = 4 (high-sensitivity group), the maternal negative parenting × peer victimization interaction was significant (β = -0.15, t = -2.50, p = 0.01). Maternal negative parenting significantly negatively predicted depression under high peer victimization (β = -0.13, t = -2.36, p = 0.02) and marginally positively predicted depression under low peer victimization (β = 0.04, t = 1.85, p = 0.07) (interaction pattern shown in Figure 1 [Figure 1: see original paper]), consistent with the match-mismatch hypothesis. For males with CGS = 1 (β = 0.14, t = 1.14, p = 0.26), 2 (β = 0.08, t = 1.26, p = 0.21), and 3 (β = -0.16, t = -1.10, p = 0.28), the peer victimization × maternal negative parenting interaction was not significant. To simplify the model, these individuals were combined into a low CGS group (β = 0.01, t = 0.24, p = 0.81), with the interaction pattern shown in Figure 1, where maternal negative parenting and peer victimization tended to function according to the cumulative stress hypothesis.

Table 3 Effects of FKBP5 Multi-Locus Additive Score, Maternal Negative Parenting, and Peer Victimization on Male Adolescent Depression

Predictor	β	95% CI	p
T1 Depression	0.58	[0.47, 0.69]	<0.001
T1 Maternal Negative Parenting	0.01	[-0.02, 0.02]	0.237
T2 Peer Victimization	0.08	[0.06, 0.11]	<0.001

Predictor	β	95% CI	p
CGS × T1 Maternal Negative Parenting	0.01	[-0.01, 0.02]	0.115
CGS × T2 Peer Victimization	0.01	[-0.03, 0.01]	0.012
T1 Maternal Negative Parenting × T2 Peer Victimization	0.01	[-0.01, 0.03]	0.115
CGS × T1 Maternal Negative Parenting × T2 Peer Victimization	-0.04	[-0.04, -0.003]	0.012**

Note: Bold values remain significant after Benjamini–Hochberg (1995) correction. 95% CIs were obtained via Bootstrap method.

Table 4 Effects of FKBP5 Multi-Locus Additive Score, Maternal Negative Parenting, and Peer Victimization on Female Adolescent Depression

Predictor	β	95% CI	p
T1 Depression	0.62	[0.51, 0.74]	<0.001
T1 Maternal Negative Parenting	0.01	[-0.01, 0.02]	0.234
T2 Peer Victimization	0.07	[0.05, 0.11]	<0.001
CGS × T1 Maternal Negative Parenting	0.01	[-0.02, 0.02]	0.081
CGS × T2 Peer Victimization	0.01	[-0.02, 0.01]	0.081
T1 Maternal Negative Parenting × T2 Peer Victimization	0.01	[-0.03, 0.04]	0.081
CGS × T1 Maternal Negative Parenting × T2 Peer Victimization	-0.01	[-0.04, 0.02]	0.081

Figure 1 Interaction Between Maternal Negative Parenting and Peer Victimization on Depression in Male Adolescents with High and Low CGS

3.3 Internal Consistency Analysis

To examine the stability and reliability of the $E_1 \times E_2 \times G$ findings, we randomly split the total sample into two subsamples. The three-way interaction was significant in both subsample 1 (β = -0.44, t = -1.99, p = 0.048) and subsample 2 (β = -0.48, t = -2.76, p = 0.006) for male adolescents. Further analysis revealed that when CGS = 4, the maternal negative parenting × peer victimization interaction was significant (subsample 1: β = -0.20, t = -2.25, p = 0.03;

subsample 2: β = -0.17, t = -2.00, p = 0.048). Under high peer victimization, maternal negative parenting marginally or significantly negatively predicted depression (subsample 1: β = -0.13, t = -1.89, p = 0.06; subsample 2: β = -0.19, t = -2.03, p = 0.045), with maternal negative parenting and peer victimization influencing depression in a match-mismatch pattern. However, under low peer victimization, maternal negative parenting's predictive effect was non-significant (subsample 1: β = 0.06, t = 1.98, p = 0.051; subsample 2: β = 0.04, t = 1.29, p = 0.20). When CGS < 4, the interaction was non-significant (subsample 1: β = -0.03, t = -0.34, p = 0.74; subsample 2: β = 0.01, t = 0.14, p = 0.89), with the interaction pattern tending to follow the cumulative stress hypothesis.

3.4 Supplementary Analyses

To validate the multi-locus approach, supplementary analyses were conducted. First, after controlling for T1 depression, single-locus effects of each FKBP5 polymorphism were examined. Results showed significant E × E × G effects for males (rs1360780: ΔR^2 = 0.012, β = -0.39, t = -3.01, p = 0.02; rs3800373: ΔR^2 = 0.008, β = -0.30, t = -2.41, p = 0.02; rs9296158: ΔR^2 = 0.006, β = -0.24, t = -2.15, p = 0.03). Second, two-locus cumulative effects were analyzed after excluding each locus in turn, revealing significant E × E × G effects for males (rs1360780 and rs3800373: ΔR^2 = 0.007, β = -0.45, t = -2.31, p = 0.03; rs1360780 and rs9296158: ΔR^2 = 0.011, β = -0.39, t = -2.80, p = 0.02; rs3800373 and rs9296158: ΔR^2 = 0.008, β = -0.33, t = -2.47, p = 0.02). These analyses indicate that while single-locus effects were significant, no single locus showed a dominant effect, supporting the multi-locus additive approach (Huang & Starr, 2019; Pearson-Fuhrhop et al., 2014; Vrshek-Schallhorn et al., 2015). Finally, linear gene model testing revealed that the full model explained more variance than the linear gene effect model, but the difference in R^2 was not significant (ΔR^2 = 0.014, F(12, 444) = 2.81, p > 0.05), indicating that the three FKBP5 loci did not significantly deviate from linear gene effects. Therefore, linear coding (0, 1, 2) based on minor allele count and multi-locus additive scoring were appropriate.

4 Discussion

This longitudinal study examined the interactive effects of distal and proximal stressors (maternal negative parenting and peer victimization) and a cumulative score of three FKBP5 loci (rs1360780, rs3800373, and rs9296158) on adolescent depression, testing which theoretical model (cumulative stress vs. match-mismatch) best explained the observed patterns.

Results revealed a significant three-way interaction among males but not females. In male adolescents, when individuals carried more susceptibility alleles (CGS = 4), maternal negative parenting and peer victimization influenced depression in a match-mismatch pattern. Specifically, depression was higher when individuals experienced high peer victimization combined with low maternal negative

parenting, but lower when high peer victimization was paired with high maternal negative parenting. For individuals carrying fewer susceptibility alleles (CGS = 1, 2, and 3), the maternal negative parenting × peer victimization interaction was not significant, but the two factors tended to influence depression according to the cumulative stress hypothesis—depression was lower when both maternal negative parenting and peer victimization were low, and higher when both were high. Thus, both cumulative stress and match-mismatch hypotheses can explain adolescent depression mechanisms, applying to individuals with lower and higher FKBP5 multi-locus additive scores, respectively, though these results were limited to males.

The match-mismatch and cumulative stress hypotheses share a focus on individual responses to environmental factors but differ in that the former posits higher developmental plasticity, with individuals developing adaptive stress coping strategies following distal adversity, while the latter suggests lower plasticity, with individuals showing minimal or no adaptive responses to distal adversity. Importantly, these hypotheses are not mutually exclusive; their applicability depends on genetic makeup. Genetic predisposition partially determines how distal and proximal stressors affect individuals (Nederhof & Schmidt, 2012), a proposition supported by our findings. Regarding genetic influences on development, Belsky and colleagues (Belsky et al., 2009; Belsky & Pluess, 2013) propose that genes influence environmental sensitivity, with certain genotypes rendering individuals more susceptible to positive and/or negative environments and thus showing greater plasticity. Nederhof and Schmidt (2012) also emphasize that individuals carrying certain genotypes (susceptibility genotypes) may be more responsive to distal environments, with distal environments more likely to induce adaptive phenotypic programming that guides development toward adaptation to expected (similar) subsequent environments, but results in maladaptation when facing mismatched proximal environments. Individuals without or with fewer susceptibility alleles may follow a cumulative stress pattern, being less influenced by adaptive programming from distal environments, such that detrimental effects of distal and proximal adversities accumulate, increasing allostatic load and ultimately resulting in higher depression levels. Our findings align with Nederhof and Schmidt' s integrated model of gene-distal-proximal environment interactions.

Although few studies have examined genetic moderation of cumulative stress and match-mismatch hypotheses, neurobiochemical evidence supports our results. Lower FKBP5 multi-locus additive scores correspond to greater FKBP5 secretion, which binds excessively to GR and reduces cortisol binding capacity, elevating GC concentrations. Following high negative parenting, increased GC release during stress responses leads to excessively high GC concentrations, causing persistent HPA axis hyperactivity, impaired negative feedback (Matosin et al., 2018; Tyrka et al., 2015), and altered hippocampal pyramidal neuron morphology (Woolley et al., 1990). These biochemical changes may reduce adaptive stress coping strategies (Criado-Marrero et al., 2018; Matosin et al., 2018), such as cognitive reappraisal, acceptance, and problem-solving (Aldao & Nolen-

Hoeksema, 2012; Moritz et al., 2016). Over time, when adolescents face high peer victimization later in development, continued neural system impairment prevents use of adaptive strategies, resulting in high depression. Conversely, when overall adversity is low (low negative parenting and low peer victimization), stress system damage is minimal and depression remains low. This may represent the neurobiological basis for the cumulative stress pattern observed when FKBP5 multi-locus additive scores are low.

In adolescents with higher FKBP5 multi-locus additive scores, relatively lower FKBP5 expression allows adequate GC-GR binding, enabling effective negative feedback to suppress cortisol release and maintain normal HPA axis function and hippocampal development. Under these normal developmental conditions, adolescents experiencing maternal negative parenting may be more likely to develop adaptive stress coping strategies (Kuhn et al., 2015), thereby increasing resilience or resistance to subsequent adverse environments (Daskalakis et al., 2013; Romeo, 2015). If individuals later encounter peer victimization matching their maternal negative parenting levels, they can effectively utilize established coping mechanisms. Research shows that distal adversity can adaptively shape cognitive functioning, such that when facing proximal stress, individuals show better shifting function, enabling flexible, rapid, and efficient responses to recurring adverse environments and promoting positive development (Mittal et al., 2015). Conversely, under mismatched conditions—such as low maternal negative parenting combined with high peer victimization in our study—individuals cannot benefit from coping strategies established through distal adversity and may even experience compromised development due to inappropriate responses (Herbison et al., 2017). In other words, adolescents experiencing low maternal negative parenting may be unprepared to respond appropriately to high peer victimization, resulting in higher depression. A recent MRI study found smaller hippocampal volume in individuals with mismatched distal and proximal stress levels compared to matched individuals (Paquola et al., 2017), with hippocampal morphology representing an important predictor of adolescent depression (Rao et al., 2010). Additionally, our study found marginally higher depression in adolescents experiencing high maternal negative parenting and low peer victimization compared to those experiencing low levels of both ($\beta = 0.04$, $t = 1.85$, $p = 0.07$), partially supporting the match-mismatch hypothesis. Nederhof et al. (2014) similarly found that individuals experiencing low early adversity and high recent stress showed significantly higher depression risk than those experiencing low levels of both, while individuals experiencing high early life stress showed equivalent depression risk regardless of recent stress levels. We speculate two reasons for these findings: First, individuals may more readily adapt to mild or moderate stress (i.e., low rather than high maternal negative parenting in our study) than extreme stress (Santarelli et al., 2014). Second, compared to mismatched conditions, individuals experiencing high distal adversity and low proximal stress may show less health deterioration than those experiencing low distal adversity and high proximal stress, demonstrating relatively better developmental functioning (Frankenhuis & Del Giudice, 2012).

Notably, the three-way interaction was not significant among female adolescents. We speculate this may relate to several factors. First, hormones may regulate FKBP5 gene expression, causing gender differences in interaction effects (Hubler & Scammell, 2004). Magee (2005) demonstrated that androgens can directly regulate FKBP5 through interactions between estrogen receptors and a distal enhancer located 65 kb downstream of the transcription start site in intron 5 of the FKBP5 gene. Thus, hormones may modulate FKBP5 expression, leading to gender-specific E × E × G effects. Second, HPA axis sensitivity shows gender differences (Hollanders et al., 2017). For example, HPA axis response differs significantly before and after antidepressant treatment in males (with hyperactivity resolving post-treatment) and predicts treatment response, whereas no such difference appears in females (Binder et al., 2009). Since the three FKBP5 loci selected in our study importantly regulate HPA axis function, this may contribute to gender-specific interaction results. Third, variability in environmental variables may influence detection of interactions (Wang et al., 2019). Female adolescents in our sample showed relatively low levels and limited variability in maternal negative parenting and peer victimization, potentially insufficient for detecting significant E × E × G effects. Furthermore, our findings may suggest that genetic factors influencing environmental response patterns differ between males and females, with FKBP5 being more closely associated with male environmental sensitivity, thus preventing revelation of these theoretical models' applicability to female depression. Indeed, research shows that genes influencing environmental sensitivity differ by gender; for example, Lavebratt et al. (2010) found that FKBP5 rs1360780 moderated male but not female sensitivity to negative environments. Our results do not imply that cumulative stress and match-mismatch hypotheses are inapplicable to female adolescents; future research should investigate unique genes related to female environmental sensitivity and their interactions with distal and proximal adversities showing greater variability, to further elucidate whether these hypotheses are moderated by such genes and enrich our understanding of depression mechanisms to provide comprehensive, timely, and effective assistance for both genders.

This study examined the interactive effects of maternal negative parenting, peer victimization, and FKBP5 multi-locus additive score on adolescent depression, exploring gender differences and testing cumulative stress versus match-mismatch hypotheses to advance understanding of depression mechanisms. Results demonstrate gender differences in adolescent depression mechanisms and show that distal-proximal stress interaction patterns differ depending on genetic background. Moreover, findings emphasize the importance of adaptive phenotypic plasticity in depression mechanism research, potentially opening new avenues for etiological investigation. As Homberg (2012) stated, "Depression may be far more complex than researchers anticipate, and slow progress in depression etiology research may result from adhering to the mainstream view that specific gene loci combined with stress inevitably cause depression while neglecting the adaptive nature of distal stress for individual development." Additionally, this study has important implications for adolescent depression intervention, suggest-

ing that treatment should consider genetic background and give equal attention to distal and proximal adversities. The single-gene multi-locus approach further provides reliable evidence for gene-environment interactions in adolescent depression.

Several limitations warrant mention. First, although FKBP5 polymorphisms rs1360780, rs3800373, and rs9296158 show linkage disequilibrium (e.g., Piechaczek et al., 2019) and could be analyzed as haplotypes, we did not conduct haplotype analysis. As noted, Nederhof and Schmidt' s (2012) integrated model emphasizes that higher genetic sensitivity distinguishes between cumulative stress and match-mismatch hypotheses, with Power et al. (2013) providing empirical support. Notably, single-gene multi-locus additive effects are based on the additivity hypothesis for complex psychopathology genetics, focusing on the number of susceptibility alleles carried across both chromosomes (i.e., individual genetic sensitivity), whereas haplotype analysis primarily focuses on linkage patterns among adjacent loci on the same chromosome and their association with psychopathology. Given our research aims, haplotype analysis was not conducted. Second, based on the HPA axis' s important regulatory role in stress response and depression, we examined cumulative effects of three FKBP5 loci closely associated with HPA axis function. The non-significant main effect and low explanatory power (1.2%) of the three-way interaction reflect "missing heritability," where molecular genetics explains less variance than quantitative genetics estimates (Maher, 2008). This phenomenon is common in molecular genetic studies of depression and other psychological traits (Dahl et al., 2019; Genin, 2020; Lopizzo et al., 2015), resulting from complex depression genetics, limitations of current research paradigms, DNA methylation effects, and interactions among multiple genes/loci and environments (Lopizzo et al., 2015). Although the gene-environment interaction explained only 1% of variance, its significance for developmental behavioral genetics should not be underestimated (Evans, 1985; Hasan & Afzal, 2019). Future research should employ high-quality designs combining candidate gene and genome-wide association approaches (Nelemans et al., 2019) to further explore multi-gene/locus and multi-environment interactions in depression under solid theoretical guidance. Third, the E × E × G effect size of 1.2% in males yielded low statistical power. Although results were corrected for multiple comparisons and internal consistency analyses showed stable findings, future research should replicate these results in other samples, particularly larger ones.

References

Note: The reference list is preserved exactly as provided in the original text, maintaining all formatting, author names, and publication details.

Note: Figure translations are in progress. See original paper for figures.