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The effects of childhood maltreatment, recent interpersonal and noninterpersonal stress, and HPA-axis multilocus genetic variation on prospective changes in adolescent depressive symptoms: A multiwave longitudinal study

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Abstract

Based on a multiwave, two-year prospective design, this study is the first to examine the extent to which multilocus hypothalamic–pituitary–adrenal axis (HPA axis)-related genetic variants, childhood maltreatment, and recent stress jointly predicted prospective changes in adolescent depressive symptoms. A theory-driven multilocus genetic profile score (MGPS) was calculated to combine the effects of six common polymorphisms within HPA-axis related genes (*CRHR1*, *NR3C1*, *NR3C2*, *FKBP5*, *COMT,* and *HTR1A*) in a sample of Chinese Han adolescents ($N = 827$; 50.2% boys; $M_{age} = 16.45 \pm 1.36$ years). The results showed that the three-way interaction of HPA-axis related MGPS, childhood maltreatment and recent interpersonal, but not noninterpersonal, stress significantly predicted prospective changes in adolescent depressive symptoms. For adolescents with high but not low HPA-axis related MGPS, exposure to severe childhood maltreatment predisposed individuals more vulnerable to recent interpersonal stress, exhibiting greater prospective changes in adolescent depressive symptoms. The findings provide preliminary evidence for the cumulative risk mechanism regarding gene-by-environment-by-environment ($G \times E_1 \times E_2$) interactions that underlie the longitudinal development of adolescent depressive symptoms and show effects specific to interpersonal stress.

Keywords: depression; polygenic; cumulative stress; HPA-axis; gene-by-environment-by-environment interaction

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Introduction

Depressive symptoms are a significant problem among adolescents worldwide (Thapar et al., 2012) and have long-term negative impacts on adolescent academic performance, interpersonal relationships, and mental and physical health (Hill et al., 2014; Thapar et al., 2012). It has been widely established that both early (LeMoult et al., 2020) and recent environmental stress (Yue et al., 2016) contribute to the etiology of adolescent depression. However, the mechanism by which early stress interacts with later stress to enhance the risk of depression is far from simple, and the extent to which it fits the cumulative stress or mismatch hypotheses has been controversial (Nederhof & Schmidt, 2012; Power et al., 2013). Under the cumulative stress hypothesis, childhood maltreatment makes individuals more vulnerable to recent stressful life events, increasing the risk of depression (McEwen, 2003), while under the mismatch hypothesis, exposure to (mild) early stress buffers youth from the adverse effects of recent stress, reducing the risk of depression (Schmidt, 2011; Seery et al., 2010). An integrated model proposes that programing sensitivity, the ability of an individual to program his or her (endo)phenotype based on environmental cues to improve adaptability to similar environmental conditions in the future, makes it possible to distinguish these two contradictory hypotheses (see Supplementary Materials Figure S1; Nederhof & Schmidt, 2012). That is, the cumulative stress hypothesis applies to individuals with low programing sensitivity, while the mismatch hypothesis applies to those with high programing sensitivity. Recent evidence suggests that hypothalamic–pituitary–adrenal (HPA) axis-related genetic variants, which play a crucial role in stress processing, may contribute to explaining the variance in programing sensitivity (e.g., Daskalakis et al., 2013; Nederhof & Schmidt, 2012; Starr et al., 2014, 2021). Nevertheless, few studies have examined the three-way interaction of HPA-axis related genetic variation and early and recent stress (i.e., gene-by-environment-by-environment interaction; $G \times E_1 \times E_2$) on adolescent depression, especially through a multilocus genetic profile score (MGPS) approach and based on a longitudinal design. Accordingly, adopting the MGPS approach, the current study aimed to test this integrated hypothesis by examining the effects of HPA-axis multilocus genetic variation, childhood maltreatment and recent interpersonal stress on the prospective changes in adolescent depressive symptoms with three waves of data across two years.

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Environmental stress and adolescent depressive symptoms

There is little doubt that not only recent stress (e.g., recent interpersonal or noninterpersonal stressful life events) but also early stress (e.g., childhood maltreatment) powerfully predicts adolescent depression (LeMoult et al., 2020; Yue et al., 2016). Furthermore, these two types of stress during different periods of life could interact with each other, increasing or decreasing the risk of depression in a manner of "cumulative stress" or "mismatch". According to the cumulative stress hypothesis, childhood maltreatment makes individuals more vulnerable to recent stressful life events, leading to an increased risk of depression (e.g., Nederhof & Schmidt, 2012). Notably, the "cumulative" concept here refers to a multiplicative, but not an additive, effect of early and recent stress. This hypothesis emphasizes that stress exposures during a lifetime are multiplicatively cumulative and result in the buildup of an allostatic load, enhancing the likelihood of developing a disease (McEwen, 2003). For instance, prior work reveals that youth with a history of childhood maltreatment report higher levels of depression than those without such a history when exposed to severe but not mild recent stress (McLaughlin et al., 2010; Rousson et al., 2020)[1].

In contrast, the mismatch hypothesis (or stress inoculation hypothesis; Hoogland & Ploeger, 2022; Schmidt, 2011; Seery et al., 2010; or steeling effect; Rutter, 2006) argues that (mild) childhood adversity during plastic developmental phases could trigger adaptive functions that increase the fitness of an individual under recent stress, decreasing the risk of depression. For instance, in a prospective study of Dutch adolescents, recent stressful life events have been found to significantly predict the onset of adolescent depression for those exposed to low and mean levels, but not high levels, of childhood adversity (Oldehinkel et al., 2014). Similarly, individuals exposed to a certain amount of adversity, including physical and sexual assault and trauma experiences, are more likely to develop resilience and be less affected by recent stress (Seery et al., 2010). Rats subjected to severe early neglect (24-hour maternal deprivation) exhibit better hippocampal synaptic plasticity and emotional learning under high-stress conditions in adulthood (Oomen et al., 2010). Taken together, these conflicting findings suggest substantial individual differences in how childhood maltreatment influences the effect of recent stressful life events on adolescent depression.

G × E_1 × E_2 and adolescent depressive symptoms

Both genetic and environmental factors underlie the etiology of depression (Feurer et al., 2017; Starr & Huang, 2019; Starr et al., 2019). Recent research further proposes a complex three-way interaction model whereby genetic variation and early and recent stress interact to explain individual differences in depression (Daskalakis et al., 2013; Power et al., 2013; Starr et al., 2014). One important work is an integrated hypothesis (see Figure S1; Nederhof & Schmidt, 2012), which highlights that cumulative stress and mismatch hypotheses may depend on genetic predispositions that explain individuals' programing sensitivity. Individuals with genetic predispositions reflecting high programing sensitivity could benefit from a match of early and recent stress by adaptive programing (i.e., mismatch). In contrast, individuals with genetic variants associated with low adaptive programing sensitivity would be less likely to be programed by early stress, resulting in continuous wear under the same situation (i.e., cumulative stress; Hoogland & Ploeger, 2022; Nederhof & Schmidt, 2012). Notably, this integrated model attempts to reconcile previous different models and points out that the programing sensitivity here in fact originates from (or also refers to) the concept of "plasticity" in previous models such as differential susceptibility (Belsky et al., 2007) and biological sensitivity to context (BSC; Boyce & Ellis, 2005). As highlighted by Nederhof & Schmidt (2012), the alleles exhibiting high plasticity to the environment might be those with high programing sensitivity.

Accordingly, the HPA-axis related genes, that is, the genes that directly (e.g., *CRHR1*, *NR3C1*, *NR3C2*, and *FKBP5*) and indirectly (e.g., *COMT* and *HTR1A*) modulate HPA-axis functioning, have gained particular attention under this integrated hypothesis (Daskalakis et al., 2013; Nederhof & Schmidt, 2012). For instance, the BSC model and a series of previous studies have suggested that the alleles that trigger heightened stress reactivity may be more susceptible to the environment and considered plasticity alleles (e.g., Boyce & Ellis, 2005; Cao & Rijlaarsdam, 2023; McKenna et al., 2021; Pagliaccio et al., 2015; Starr & Huang, 2019). Consistently, childhood maltreatment was found to downregulate methylation at intron 7 of the *FKBP5* gene, particularly within its T-allele, but not GG-homozygote, carriers (Klengel et al., 2013). Particularly among these plasticity allele carriers, severe early maltreatment experiences induce hyperactivity of the right amygdala in response to threat-related stimuli (Di Iorio et al., 2017). Notably, these alterations regarding heightened stress reactivity and endophenotypes that occurred in plasticity alleles might be adaptive programing, functioning as maladaptive in later safe, predictable environments but as adaptive in later dangerous, unpredictable environments (Daskalakis et al., 2013; Nederhof & Schmidt, 2012). Accordingly, it seems that these plasticity alleles of the HPA-axis might predispose carriers to benefit from a match of early and recent stress. However, the work of Starr et al. (2014) focusing on *CRHR1* rs110402 × early stress × recent stress only partially supports this hypothesis: for A-allele carriers of *CRHR1* rs110402, which may reflect low programing sensitivity, early stress interacted with recent stress on adolescent depressive symptoms in a manner of cumulative stress, whereas for GG homozygous carriers, early stress failed to interact with recent stress. It should be noted that this work, together with the majority of extant G × E_1 × E_2 research, relies on a single-candidate gene design, which has disadvantages in obtaining adequate power and reliable results and gains increasing criticism for the neglect of polygenic underpinnings of depression (Border et al., 2019; Sullivan et al., 2012).

Several polygenetic approaches have been recommended in recent years, including (i) genome-wide association study (GWAS)-derived polygenic risk scores and (ii) theoretically driven multilocus genetic profile scores (MGPSs) that capture cumulative effects of candidate genes linked to a given biological system (Zhang & Belsky, 2022). The HPA-axis related MGPSs, for instance, have been consistently found to show greater power and predictive validity than individual genes examined in isolation (Cao & Rijlaarsdam, 2023; Di Iorio et al., 2017; Feurer et al., 2017; McKenna et al., 2021; Pagliaccio et al., 2015; Starr & Huang, 2019;

[1]It should be clarified that there is another widely used hypothesis in this area—stress sensitization. Stress sensitization posits that early stress increases an individual's sensitivity to later stress (Hammen et al., 2000; Stroud, 2020). This sensitivity is mainly reflected in the fact that early stress lowers an individual's threshold for depressive reactions to recent stress (Harkness et al., 2006). That is, youth with a history of adversity versus those without such a history would require only *mild* stress to trigger depression. In addition to this core nature, there are also some investigators who consider the multiplicative interaction of early and recent stress (in a manner of cumulative stress) described above as another manifestation of stress sensitization (e.g., McLaughlin et al., 2010; Rousson et al., 2020; Starr et al., 2021). Therefore, these two hypotheses have similarities but also slightly differ from each other.

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Starr et al., 2019). Unfortunately, to the best of our knowledge, only one study based on a cross-sectional design has utilized a polygenic approach to examine the G × E_1 × E_2 interaction regarding HPA-axis related genes on adolescent depressive symptoms (Starr et al., 2021). However, the results failed to support the integrated hypothesis. Specifically, for adolescents with high MGPS, which may reflect high programing sensitivity, recent stress was associated with depression at high but not low childhood adversity (i.e., in a cumulative stress manner), but for adolescents with low MGPS, there was no interaction between childhood adversity and recent stress (Starr et al., 2021). Despite not being limited to HPA-axis related genes, similar findings were reported in a prospective, multiwave longitudinal study that examined the interaction of a polygenic score of 13 genetic variants and childhood and adult material environmental quality on psychological distress (Keers & Pluess, 2017). The results showed that individuals with more genetic plasticity alleles were more vulnerable to adversity as adults, especially when they had a poor childhood environment. Notably, none of these G × E_1 × E_2 studies were conducted in Chinese Han samples. However, different ethnicities may differ in allele frequencies and functions (van IJzendoorn et al., 2012) and may have different family and social norms (Cho et al., 2021). For instance, physical punishment is often used as a form of discipline in traditional Chinese families, and is not perceived as harmful as it tends to be in U.S. families (Hayes et al., 2021). Thus, whether the interactive effect regarding HPA-axis related MGPS, childhood maltreatment and recent stress would become weaker or (even) different in Chinese Han samples deserves exploration.

Additional considerations of environmental stress and study design

It is worth noting that environmental stress is a heterogeneous construct, and interpersonal stress constitutes a robust candidate environment for G × E interactions (Vrshek-Schallhorn et al., 2014), especially for those involving HPA-axis related genes. Evidence has suggested that the HPA-axis system is particularly sensitive to interpersonal stress. For instance, heightened cortisol reactivity is found to be robustly elicited by laboratory stressors characterized by uncontrolled, social evaluative threat (Dickerson & Kemeny, 2004). Individuals with hyperactivity of the HPA-axis are more vigilant to social threat stimuli, especially after social stress induction (Roelofs et al., 2007). More importantly, recent research has provided preliminary evidence that HPA-axis related MGPS significantly moderates the effect of interpersonal, but not noninterpersonal, stress on depressive symptoms (Feurer et al., 2017; Starr & Huang, 2019) and related brain functions (Di Iorio et al., 2017). Accordingly, the current study sought to assess recent interpersonal and noninterpersonal stress to examine their respective interactions with HPA-axis related MGPS and childhood maltreatment.

Moreover, traditional analysis models of cross-sectional studies examine only genetic variation and interindividual variation in stress as predictors of individual differences in depressive symptoms, which tends to produce excessive Type I errors and biased parameter estimates (Peugh, 2010). Given the lack of longitudinal research exploring the integrated hypothesis in the G × E_1 × E_2 interaction of adolescent depressive symptoms, this study aimed to adopt a longitudinal, multilevel design, allowing person-centred modeling of recent stress and depressive symptoms.

Current study

Although the integrated model suggests that HPA-axis related genes reflecting different programing sensitivity may moderate how early stress interacts with recent stress in the prediction of adolescent depression, few studies have tested this three-way interaction, especially through a longitudinal, polygenic approach and focused on interpersonal stress. To address the gaps in the extant literature, using a multiwave, two-year prospective longitudinal design, the current study aimed to explore the three-way interaction of HPA-axis multilocus genetic variants, childhood maltreatment and recent interpersonal versus non-interpersonal stress on adolescent depressive symptoms in a sample of Chinese Han adolescents (see Figure S1). For this purpose, a theory-driven MGPS was used to capture the additive effect of six commonly studied HPA-axis related genetic variants: *CRHR1* rs110402, *NR3C1* rs41423247, *NR3C2* rs5522, *FKBP5* rs1360780, *COMT* rs4680, and *HTR1A* rs6295 (see details in the Methods section). Higher scores for MGPS indicated more alleles associated with heightened stress reactivity that may reflect high programing sensitivity. Hierarchical linear modeling (HLM), a person-centred modeling method, was adopted. We hypothesized that the HPA-axis related MGPS would interact with childhood maltreatment and recent interpersonal stress (but not non-interpersonal stress) in predicting prospective changes in adolescent depressive symptoms. Based on the integrated hypothesis, we expected that for adolescents with high MGPS that may reflect high programing sensitivity, the interaction between childhood maltreatment and interpersonal stress would be consistent with the mismatch hypothesis, whereas for adolescents with low MGPS, this interaction might work in a cumulative stress manner.

Methods

Participants and procedure

Participants were drawn from a longitudinal study investigating genetics and adolescent psychological adjustment in Shandong Province, China. A total of 827 adolescents (M_{age} = 16.45 ± 1.36 years; 50.2% boys) were primarily recruited from the first year (grade) of three technical secondary schools located in the cities of Jinan, Rizhao, and Tai'an. All participants were of Chinese Han ethnicity, without any Chinese reading or language difficulties, and had no current diagnosis of major physical, neurological, or pervasive developmental disorders. The median monthly family income was ¥ 3,000 (approximately $ 445)~ ¥ 6,000 (approximately $ 891). According to the Shandong Provincial Bureau of Statistics, the average monthly income of residents in Shandong Province was ¥ 2,633.08 (http://tjj.shandong.gov.cn/tjnj/nj2022/zk/zk/indexch.htm). The majority of parents graduated from primary school (mothers: 23.3%; fathers: 9.8%), junior high school (mothers: 47.7%; fathers: 47.3%), or high school (mothers: 20.7%; fathers: 31.2%), whereas only a small proportion of mothers (8.3%) and fathers (11.7%) received an education beyond high school.

At baseline (T1), all participants were instructed by research assistants to (i) complete questionnaires assessing childhood maltreatment, recent stress and depressive symptoms and (ii) provide buccal cells using the cheek swabs kit (Rellagene, Shanghai, China) for DNA extraction in their classroom. The participants were followed-up one year later (T2) and two years later (T3). At T2 and T3, participants completed questionnaires on

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recent stress and depressive symptoms again. This study was approved by the Ethics Committee of School of Nursing and Rehabilitation, Shandong University. Informed consent was obtained from all adolescents, their parents and school headmasters.

Measures

Childhood maltreatment

At T1, adolescents retrospectively reported their maltreatment experiences before the previous year using the Chinese version of the Childhood Trauma Questionnaire-Short Form (CTQ-SF; Bernstein et al., 1997). The CTQ-SF has been widely used and shows good reliability and validity in various community adolescent samples worldwide; it consists of 28 items, including 3 validity items. The remaining 25 items measure five subtypes: physical abuse (5 items, e.g., "Hit me so hard as to leave bruises"), emotional abuse (5 items, e.g., "Family said hurtful things"), sexual abuse (5 items, e.g., "Was touched sexually"), physical neglect (5 items, e.g., "No enough to eat"), and emotional neglect (5 items, e.g., "Made to feel important," a reverse-scored item). All items were rated on a 5-point scale from 1 ("Never") to 5 ("Always"). Sum scores were calculated, with higher scores reflecting more severe childhood maltreatment suffered by adolescents. The Cronbach's α of the CTQ-SF in the current study was .86.

Recent interpersonal and noninterpersonal stress

At T1, T2, and T3, the Adolescent Self-rating Life Events Checklist (ASLEC; Liu et al., 1997) developed for Chinese adolescents was used to assess the severity of interpersonal and noninterpersonal stressful life events experienced by adolescents during the previous year. The ASLEC has good validity and reliability among Chinese adolescents (Liu et al., 1997). Based on the original version, one item, "Pressure to enter higher education," was deleted since students in Chinese vocational high schools lack the possibility of entering higher education (e.g., university); one open-ended item, "Any other stressful events that you've experienced," was not included in the primary analysis as a result of a very low response rate. Accordingly, the ASLEC used in this study consists of 25 items assessing interpersonal stress (5 items, e.g., "I argued with my classmates") and four noninterpersonal stress: academic stress (5 items, e.g., "I failed in the examination"), being punished (7 items, e.g., "Being fined"), loss (4 items, e.g., "Stolen and lost things"), and adjustment stress (4 items, e.g., "Significant changes took place in living habits"). Participants rated the extent to which "this life event negatively affected you" on a 5-point Likert scale from 1 ("not at all") to 5 ("extremely severe") or indicated that the event had not happened ("did not happened" = 1). Sum scores were calculated as indices of the severity of interpersonal and noninterpersonal stress. The second order CFA supported the five-factor construct for interpersonal and noninterpersonal stress at both T1, T2, and T3 (T1: $\chi^2/df = 2.95$, CFI = .91, TLI = .89, RMSEA = .05, and SRMR = .05. T2: $\chi^2/df = 2.41$, CFI = .90, TLI = .87, RMSEA = .04, and SRMR = .06. T3: $\chi^2/df = 1.90$, CFI = .88, TLI = .87, RMSEA = .05, and SRMR = .07). Longitudinal metric invariance was established for interpersonal stress ($\Delta\chi^2$ (8) = 10.45, $p = .235$) and noninterpersonal stress ($\Delta\chi^2 \leq 19.48$, $p \geq .078$), except for loss ($\Delta\chi^2$ (6) = 19.39, $p = .004$), indicating that the measurement of the constructs was generally invariant across time.

HPA-axis multilocus genetic variation

At T1, buccal cells were used for genomic DNA extraction and genotyping using the Sequenom chip-based MALDI-TOF mass spectrometry platform (San Diego, CA, USA; Starr & Huang, 2019). Based on BSC theory and previous research (Feurer et al., 2017; Pagliaccio et al., 2014; Starr & Huang, 2019; Starr et al., 2021) and consistent with Cao & Rijlaarsdam (2023), four core SNPs that directly regulate HPA-axis function (*CRHR1* rs110402, *NR3C1* rs41423247, *NR3C2* rs5522, and *FKBP5* rs1360780) and two peripheral SNPs that indirectly regulate HPA-axis function (*COMT* rs4680 and *HTR1A* rs6295) were included to calculate the HPA-axis related MGPS. No SNPs significantly deviated from Hardy-Weinberg equilibrium ($\chi^2 \leq 2.04$, $p \geq .153$), and the minor allele frequencies for all SNPs were greater than 5% (see Supplementary Table S1). Following preestablished procedures of Cao & Rijlaarsdam (2023), the MGPS was aggregated based on the previously established effects of individual SNPs on HPA-axis regulation, where a higher MGPS indicated a larger number of alleles previously associated with heightened stress reactivity of the HPA-axis that may reflect high programing sensitivity (Ellis & Boyce, 2008; Nederhof & Schmidt, 2012).

Depressive symptoms

At T1, T2, and T3, the Chinese version of the Children's Depression Inventory (CDI; Kovacs, 1992) was used by adolescents to assess the severity of their depressive symptoms. The CDI is a 27-item self-report measure of depressive symptoms among children and adolescents that has been widely used and shows good validity and reliability, especially in nonclinical populations (e.g., Brendgen et al., 2005). For each item, the participants identified which of the three statements best described themselves in the past two weeks (e.g., 0 = "I am sad occasionally," 1 = "I am sad many times" and 2 = "I am sad all the time"). Sum scores were calculated for each participant, with higher scores reflecting higher levels of depressive symptoms (T1: $M = 15.59 \pm 7.46$, range = 0 ~ 48; T2: $M = 13.54 \pm 8.01$, range = 0 ~ 46; T3: $M = 12.55 \pm 7.93$, range = 0 ~ 40). In this study, the CDI-based single-factor construct of depressive symptoms showed a good fit to the data at three annual waves (T1: $\chi^2/df = 2.28$, CFI = .89, TLI = .87, RMSEA = .04, and SRMR = .04. T2: $\chi^2/df = 2.26$, CFI = .90, TLI = .89, RMSEA = .04, and SRMR = .05. T3: $\chi^2/df = 1.70$, CFI = .90, TLI = .89, RMSEA = .04, and SRMR = .05). Longitudinal metric invariance was established ($\Delta\chi^2$ (54) = 71.22, $p = .058$), indicating that the measurement of adolescent depressive symptoms was invariant across time. According to the threshold (CDI score≥19) recommended by Kovacs (1992), our sample reported higher prevalence rates of clinical depressive symptoms than global general adolescents (20%; Shorey et al., 2022) at all three waves (T1: 33.41%; T2: 26.03%; T3: 25.86%; $ps < .01$). In addition, there were 16.8% adolescents who reported elevated or persistent clinical depressive symptoms from T1 to T3. The Cronbach's αs of the CDI in the three waves were .86 ~ .89.

Missing data

Given the presence of missing data (childhood maltreatment at T1: 0.8%; recent stress at T1, T2 and T3: 0.8%, 11.6%, 47.5%, respectively; depressive symptoms at T1, T2 and T3: 1.2%, 8.9% and 47.2%, respectively), the pattern of missing data was checked. To be noted, the high attrition rate at T3 was primarily due to the off-campus internships of these technical secondary school students. All students were required to participate in internships, but the internship periods were located at different times for

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different students depending on their academic schedule. Nonrespondents at T2 and T3 did not differ from respondents in the demographic variables and study variables (sex, age, childhood maltreatment, recent stress at T1 and T2, and depressive symptoms at T1 and T2) after multiple correction. Little's missing completely at random (MCAR) test did not yield significant results ($\chi^2 = 2.85, p > .999$). The expectation-maximization algorithm was therefore used to address missing data, as this approach has been demonstrated to be suitable for missing data that is MCAR or missing at random in longitudinal studies, with stable interpolation results, good robustness and small bias (Newman, 2003).

Statistical analysis

Main analyses

Five main analytical steps were conducted in Mplus 7.4. First, the equal gene model was conducted using HLM to ensure that the additive approach (i.e., MGPS) for aggregating the effect of each polymorphism was appropriate (Stocker et al., 2017; see Supplementary Information "Equal gene model" section for details). Second, descriptive statistics and bivariate correlations among all study variables were calculated. Third, HLM was further conducted to examine the effects of HPA-axis related MGPS, childhood maltreatment, recent stress and their three-way interaction on prospective changes in adolescent depressive symptoms (Feurer et al., 2017). Adolescent sex and age were included as covariates (Hill et al., 2014; Thapar et al., 2012). Separate analyses were conducted for each stress domain. Bonferroni correction was used to control for multiple testing among $G \times E_1 \times E_2$ interactions. Accordingly, the threshold of the p value was .010 (.05/5, i.e., five stress domains). All within-subject predictors, including lagged depressive symptoms and recent interpersonal and noninterpersonal stress, were centered at group means, while all between-subject predictors, including childhood maltreatment and MGPS, were centered at grand means. All models were corrected for nonnormal distributions by maximum likelihood estimation with robust standard errors, which is more sensitive to model misspecification than bootstrapping (Finney & DiStefano, 2013).

Specifically, (i) an unconditional model was conducted without any within-subject or between-subject predictors to obtain intraclass correlation (ICC, the proportion variance that is between subjects versus the total variance) of adolescent depressive symptoms; (ii) within-subject predictors were added into the unconditional model to test the prospective association between recent stress and adolescent depressive symptoms; and (iii) between-subject predictors and cross-level interactions were added to investigate whether the prospective association between recent stress and adolescent depressive symptoms varied over time as a function of adolescent sex, age, childhood maltreatment and MGPS (i.e., the three-way interaction between childhood maltreatment, recent stress and MGPS on the prospective changes in adolescent depressive symptoms). The specific modeling procedures were consistent with Feurer et al. (2017) and described in the Supplementary Information "Multilevel models" section.

As a measure of effect size (Snijders & Bosker, 1994), R_1^2 and R_2^2 were estimated by fitting the empty model (to estimate the outcome variance) and the full model (random-intercept-only model; the model of substantive interest) and represented the level-1 and level-2 modeled proportion of variances, respectively, which can be calculated using the formula:

$R_1^2 = 1 - (\sigma^2$ full model $+ \sigma^2_{u0}$ full model$)/(\sigma^2$ empty model $+ \sigma^2_{u0}$ empty model),

$R_2^2 = 1 - [(\sigma^2$ full model $/ n) + \sigma^2_{u0}$ full model$]/[(\sigma^2$ empty model $/ n) + \sigma^2_{u0}$ empty model],

σ^2 is the residual variance in level-1; σ^2_{u0} is the residual variance in level-2; and n is the group size.

Fourth, simple slope analyses were performed to examine the pattern of significant $G \times E_1 \times E_2$ interactions, by which to what extent the interactions were consistent with the integrated hypothesis was tested. Finally, the role of each individual SNP in the $G \times E_1 \times E_2$ interactions was investigated.

Sensitivity analyses

To test the robustness of the findings in our primary analyses, the $G \times E_1 \times E_2$ interactions were explored again when (i) the open-ended item of ASLEC was included; (ii) the interactive effects of covariates with both genetic and environmental variables were controlled for (Dick et al., 2015); (iii) the quadratic effects for genetic and environmental stress were controlled for (Dick et al., 2015); (iv) the exposure number of childhood maltreatment and recent stress (see details in Supplementary Information and Table S2) was used; and (v) the four dimensions of noninterpersonal stress (academic stress, being punished, loss, and adjustment stress) were standardized and averaged. In addition, using our available data, we were able to establish an approximation of the MGPS established by Di Iorio et al. (2017), including *CRHR1* rs110402, *FKBP5* rs1360780 and *NR3C2* rs5522/rs4635799 haplotype (see Supplementary Table S3). Our approximation MGPS only differed in the *NR3C2* rs5522 but not the *NR3C2* rs5522/rs4635799 haplotype. We also reran models with this approximation MGPS.

Results

Preliminary analyses

The results of disaggregated and equal gene models are shown in Table S4. The constrained equal gene models did not significantly differ from the freely estimated disaggregated models ($\Delta R_1^2 \leq .006$, $\Delta R_2^2 \leq .003$, $ps \geq .914$). This finding suggested that the six SNPs included in the HPA-axis related MGPS did not significantly depart from equal effects across each other regarding their main effects and interactive effects, supporting additivity across the effects of six SNPs. That is, these analyses provided statistical evidence for the use of the MGPS in this study.

Descriptive statistics and bivariate correlations for all study variables are presented in Table 1. The severity of depressive symptoms was correlated at three annual waves ($rs \geq .55$, $p < .001$) and showed a significant decline from T1 to T3 ($F(2, 1652) = 106.09$, $p < .001$, $\eta^2 = .114$), consistent with previous findings about the developmental trajectory of depressive symptoms from mid- to late adolescence (17 ~ 19 years old; Schubert et al., 2017). Recent interpersonal and noninterpersonal stress demonstrated a significant, medium- to large-magnitude stability over time ($.24 \leq rs \leq .61$, $ps \leq .001$). The HPA-axis related MGPS was unrelated to childhood maltreatment ($r = -.01$, $p = .867$) or recent interpersonal and noninterpersonal stress at T1, T2 and T3 ($|r|s \leq .07$, $ps \geq .054$), demonstrating the absence of significant gene-environment correlations (rGE). The HPA-axis related MGPS was also unrelated to adolescent depressive symptoms at three annual waves ($|r|s \leq .06$, $ps \geq .103$). In contrast, childhood maltreatment and recent interpersonal and noninterpersonal stress were positively correlated with

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Table 1. Descriptive statistics and bivariate correlations for study variables

Variables	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
1. MGPS	—																			
2. CM	−.01	—																		
3. IS_T1	.01	.39***	—																	
4. IS_T2	.03	.27***	.44***	—																
5. IS_T3	−.004	.28***	.44***	.57***	—															
6. AS_T1	−.01	.32***	.71***	.37***	.40***	—														
7. AS_T2	.07	.22***	.33***	.69***	.43***	.42***	—													
8. AS_T3	−.001	.24***	.39***	.49***	.74***	.49***	.56***	—												
9. BP_T1	−.04	.35***	.69***	.36***	.29***	.75***	.32***	.30***	—											
10. BP_T2	.03	.21***	.30***	.59***	.43***	.32***	.70***	.43***	.33***	—										
11. BP_T3	−.01	.22***	.38***	.49***	.66***	.37***	.51***	.68***	.39***	.60***	—									
12. LO_T1	.01	.20***	.56***	.27***	.24***	.54***	.24***	.16***	.60***	.23***	.23***	—								
13. LO_T2	.07	.15***	.24***	.52***	.38***	.22***	.52***	.33***	.25***	.58***	.48***	.27***	—							
14. LO_T3	.01	.18***	.25***	.28***	.56***	.16***	.30***	.45***	.17***	.37***	.62***	.24***	.51***	—						
15. ADS_T1	.01	.32***	.62***	.29***	.30***	.53***	.21***	.28***	.55***	.23***	.33***	.53***	.15***	.24***	—					
16. ADS_T2	.04	.20***	.24***	.56***	.32***	.24***	.57***	.39***	.23***	.50***	.38***	.17***	.50***	.23***	.32***	—				
17. ADS_T3	.04	.27***	.38***	.46***	.59***	.32***	.46***	.56***	.32***	.46***	.57***	.26***	.45***	.51***	.44***	.61***	—			
18. DEP_T1	−.06	.49***	.45***	.37***	.36***	.37***	.29***	.34***	.38***	.27***	.33***	.26***	.17***	.24***	.44***	.27***	.40***	—		
19. DEP_T2	−.05	.42***	.35***	.51***	.47***	.33***	.43***	.43***	.30***	.36***	.37***	.21***	.27***	.31***	.30***	.46***	.42***	.64***	—	
20. DEP_T3	.01	.38***	.29***	.43***	.50***	.20***	.36***	.43***	.17***	.29***	.38***	.12***	.25***	.33***	.20***	.36***	.44***	.55***	.74***	—
21. sex, girls	.07*	.02	.02	.05	.02	.02	.08*	.03	.01	.02	−.01	−.01	.01	−.01	.01	.02	.04	.10**	.06	.02
22. age	.002	−.05	−.05	−.06	−.07*	−.04	−.004	−.04	−.03	−.03	−.03	−.01	−.002	−.03	.02	.02	−.01	−.03	−.08*	−.09*
M/%	2.76	36.57	8.94	8.41	7.38	7.98	7.07	6.66	10.50	9.12	8.77	6.40	5.95	5.63	6.32	5.91	5.45	15.58	13.62	12.48
SD	1.08	10.12	3.94	3.71	2.60	3.24	2.42	2.01	5.22	3.46	2.80	3.18	2.79	2.05	2.70	2.32	1.67	7.43	7.80	6.61

Note: $N = 827$; Sex: boys = 0, girls = 1; MGPS = Multilocus genetic profile score; CM = Childhood maltreatment; IS = Interpersonal stress; LO = Loss; AS = Academic stress; BP = Being punished; ADS = Adjustment stress; DEP = Depressive symptoms.
*$p < .05$; **$p < .01$; ***$p < .001$.

adolescent depressive symptoms ($rs \geq .12$, $ps < .001$). Compared to boys, girls exhibited higher levels of academic stress at T2 and more severe depressive symptoms at T1 ($rs \geq .07$, $ps \leq .041$). Adolescent age was negatively correlated with interpersonal stress at T3 and depressive symptoms at T2 and T3 ($|r|$s $\leq .07$, $ps \leq .046$).

Associations between HPA-axis multilocus genetic variation, childhood maltreatment, recent stress and adolescent depressive symptoms

First, the unconditional model showed that the ICC was .606, suggesting that 60.6% of the variance in adolescent depressive symptoms was due to differences between adolescents, and 39.4% was due to within-person fluctuations over time. Then, when recent stress in level-1 was included in the unconditional model, the results showed that both interpersonal and noninterpersonal stress positively predicted prospective changes in adolescent depressive symptoms ($b \geq 0.30$, $SE \leq 0.06$, $p < .001$).

Next, we tested the integrated models using HLM as described earlier. As shown in Table 2, for the $G \times E_1 \times E_2$ models regarding interpersonal stress, the level-1 and level-2 modeled proportions of variance were 24.3% and 25.3%, respectively. Specifically, childhood maltreatment positively predicted prospective changes in adolescent depressive symptoms ($b = 0.003$, $SE = 0.001$, $p = .009$, 95% CI [0.001, 0.01]). The two-way interaction between childhood maltreatment and recent interpersonal stress on prospective changes in adolescent depressive symptoms was also significant ($b = 0.02$, $SE = 0.004$, $p < .001$, 95% CI [0.01, 0.03]). Further simple slope analysis revealed that the positive association between recent interpersonal stress and prospective changes in adolescent depressive symptoms was stronger for adolescents with high versus low levels of childhood maltreatment ($b = 0.57$, $SE = 0.06$, $p < .001$, 95% CI [0.46, 0.68] versus $b = 0.20$, $SE = 0.06$, $p = .002$, 95% CI [0.07, 0.32]), consistent with the cumulative stress hypothesis.

More importantly, the three-way interaction of HPA-axis related MGPS, childhood maltreatment and recent interpersonal stress significantly predicted prospective changes in adolescent depressive symptoms ($b = 0.01$, $SE = 0.004$, $p = .004$, 95% CI [0.004, 0.02]). Simple slope tests (see Figure 1) revealed that the two-way interaction of childhood maltreatment × recent interpersonal stress was significant at high MGPS ($M + SD$) ($b = 0.03$, $SE = 0.01$, $p < .001$, 95% CI [0.02, 0.04]); for these high MGPS adolescents, recent interpersonal stress significantly predicted prospective changes in depressive symptoms at high but not low childhood maltreatment ($b = 0.61$, $SE = 0.09$, $p < .001$, 95% CI [0.44, 0.78] versus $b = -0.02$, $SE = 0.09$, $p = .815$, 95% CI [−0.20, 0.16]), consistent with the cumulative stress hypothesis. However, the two-way interaction of childhood maltreatment × recent interpersonal stress was not significant at low MGPS ($M - SD$) ($b = 0.01$, $SE = 0.01$, $p = .317$, 95% CI [−0.004, 0.01]), suggesting the absence of a cumulative stress or mismatch effect. Thus, the interaction form of HPA-axis related MGPS × childhood maltreatment × recent interpersonal stress did not function as the integrated hypothesis.

For the $G \times E_1 \times E_2$ models regarding noninterpersonal stress (including academic stress, being punished, loss, and adjustment stress), none of the three-way interactions were significant ($ps \geq .020$; see Supplementary Table S5). However, it should be noted that although the $G \times E_1 \times E_2$ interactions were found only in the domain of interpersonal, but not noninterpersonal, stress, the Wald test did not demonstrate any significant differences between the interactions regarding these two types of stress ($\chi^2 \leq 0.69$, $df = 1$, $ps \geq .408$).

Table 2. Hierarchical linear modeling predicting changes in depressive symptoms from three-way interaction between HPA-axis multilocus genetic score, childhood maltreatment, and recent interpersonal stress

		b	*SE*	*p*	95% CI of *b*
Dep_t intercept (β_{0j})	Intercept (γ_{00})	17.34	2.16	< .001	[13.11, 21.57]
	CM (γ_{01})	0.31	0.02	< .001	[0.27, 0.35]
	MGPS (γ_{02})	−0.24	0.17	.156	[−0.58, 0.09]
	MGPS × CM (γ_{03})	0.01	0.02	.540	[−0.03, 0.05]
	sex (γ_{04})	1.08	0.38	.004	[0.34, 1.71]
	age (γ_{05})	−0.24	0.13	.067	[−0.50, 0.02]
Dep_{t-1} slope (β_{1j})	Intercept (γ_{10})	−0.19	0.01	< .001	[−0.22, −0.17]
	CM (γ_{11})	0.003	0.001	.009	[0.001, 0.01]
	MGPS (γ_{12})	0.003	0.01	.760	[−0.02, 0.03]
	MGPS × CM (γ_{13})	0.002	0.001	.088	[0.00, 0.004]
IS slope (β_{2j})	Intercept (γ_{20})	0.38	0.04	< .001	[0.30, 0.47]
	CM (γ_{21})	0.02	0.004	< .001	[0.01, 0.03]
	MGPS (γ_{22})	−0.08	0.04	.046	[−0.17, −0.001]
	MGPS × CM (γ_{23})	0.01	0.004	.004[a]	[0.004, 0.02]
R_1^2				.243	
R_2^2				.253	

Note: MGPS = Multilocus genetic profile score; CM = Childhood maltreatment; IS = Interpersonal stress.
[a]The significance threshold of *p* for $G \times E_1 \times E_2$ interactions was .010 (.050/5 testing; five stress domains).

Test of individual SNPs

When examining the role of individual SNPs, none of the SNPs yielded a significant interaction with childhood maltreatment and recent interpersonal stress (Table 3). Furthermore, the "*n*–1" test (Starr et al., 2021; Vrshek-Schallhorn et al., 2014) revealed that excluding one of the six individual SNPs from the MGPS at a time did not change the significant $G \times E_1 \times E_2$ interaction regarding interpersonal stress (see Supplementary Table S6). These results indicate that the MGPS results were not driven by any individual SNPs and that the MGPS showed greater power and predictive validity than individual SNPs examined in isolation.

Sensitivity analyses

The three-way interaction of HPA-axis related MGPS, childhood maltreatment and recent interpersonal stress in our primary analysis remained robust when (i) an open-ended item of ASLEC was included (see Supplementary Table S7), (ii) the interactive effects of covariates with both genetic and environmental variables were controlled for (Table S8), and (iii) the quadratic effects for genetic and environmental stress were controlled for (Table S9). However, such a three-way interaction did not survive when the exposure number, but not the severity, of childhood maltreatment and recent stress was used (Table S10) or when the approximation MGPS including only three SNPs that was established in previous research (Di Iorio et al., 2017) was adopted (Table S11). The

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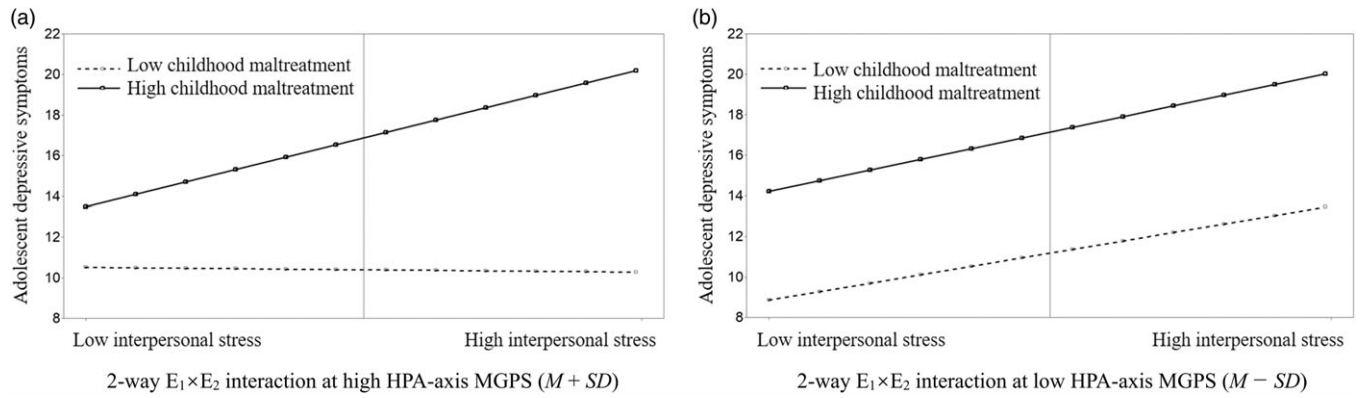

Figure 1. Prediction of changes in depressive symptoms by recent interpersonal stress as a function of childhood maltreatment at (a) high HPA-axis MGPS ($M + SD$) and (b) low HPA-axis MGPS ($M - SD$). MGPS = multilocus genetic profile score.

Table 3. Hierarchical linear modeling predicting changes in depressive symptoms from three-way interaction between HPA-axis individual SNPs, childhood maltreatment, and recent interpersonal stress

Model	SNP	*b*	*SE*	*p*[a]	95% CI of *b*
G × CM × IS	*CRHR1* rs110402	0.004	0.01	.705	[−0.02, 0.02]
	NR3C1 rs41423247	0.02	0.01	.006	[0.01, 0.04]
	NR3C2 rs5522	0.01	0.01	.608	[−0.01, 0.02]
	FKBP5 rs1360780	0.01	0.01	.253	[−0.01, 0.03]
	COMT rs4680	0.01	0.01	.346	[−0.01, 0.02]
	HTR-1A rs6295	0.02	0.02	.165	[−0.01, 0.05]

Note: G = Each SNP; CM = Childhood maltreatment; IS = Interpersonal stress.
[a]The significance threshold of p for $G \times E_1 \times E_2$ interactions was .002, i.e., .050/(30 testing; 5 stress domains × 6 SNPs).

three-way interaction regarding noninterpersonal stress remained nonsignificant when an averaged score of four dimensions of noninterpersonal stress was utilized (Table S12). In addition, neither of the interactions of quadratic childhood maltreatment × recent stress ($b = 0.0001$, $SE = 0.0001$, $p = .304$) or MGPS × quadratic childhood maltreatment × recent stress ($b = 0.0001$, $SE = 0.0001$, $p = .181$) fell significant.

Discussion

Using a multiwave, two-year prospective design, the current study was the first to explore the three-way interaction of HPA-axis multilocus genetic variants, childhood maltreatment and recent interpersonal versus noninterpersonal stress on adolescent depressive symptoms in a sample of Chinese Han adolescents. We highlighted here four main findings. First, HPA-axis related MGPS significantly interacted with the severity (but not exposure number) of childhood maltreatment and recent interpersonal stress in predicting prospective changes in adolescent depressive symptoms. Second, this three-way interaction did not function as the integrated hypothesis: for high but not low MGPS adolescents, the interaction between childhood maltreatment and interpersonal stress was significant and worked in a manner of cumulative stress. Third, when using the individual-gene approach, none of the three-way interactions survived. Fourth, only recent interpersonal stress, but not noninterpersonal stress, including academic stress, being punished, loss, and adjustment stress, drove the $G \times E_1 \times E_2$ interaction.

Specifically, although the integrated model and recent evidence suggest that HPA-axis related genes reflecting programing sensitivity may moderate how early stress interacts with recent stress to affect adolescent depression, few studies have tested this three-way interaction, especially (i) through a longitudinal, polygenic approach and (ii) focused on interpersonal stress and (iii) in a non-Western sample. Adopting a multiwave longitudinal design and person-centred HLM, the current study preliminarily revealed that HPA-axis related MGPS significantly interacted with childhood maltreatment and recent interpersonal stress in predicting prospective changes in adolescent depressive symptoms. Moreover, the pattern of interaction between childhood maltreatment and interpersonal stress was consistent with the cumulative stress hypothesis for high, but not low, HPA-axis related MGPS adolescents. Accordingly, the $G \times E_1 \times E_2$ interaction contradicted the integrated hypothesis. That is, we did not find evidence supporting heightened genetic susceptibility regarding HPA-axis related MGPS toward adaptive programing in response to childhood maltreatment and recent interpersonal stress.

However, it should be noted that Nederhof and Schmidt (2012) proposed that the size of individual programing effects requires a combination of genetic variation, developmental experience and the timing of experience. Prenatal, childhood and adolescence (especially early adolescence) periods are all considered important windows of opportunity (Andersen, 2003; Levin & Liu, 2021) when individuals are highly susceptible to phenotypic programing and may redirect abnormal development to normal processes. Evidence from pharmacology also suggests that the window of opportunity often exists in early adolescence rather than in middle or late adolescence, which could reduce the risk later in life. For instance, prepubertal exposure to methylphenidate could increase individual aversive properties and decrease the response to cocaine, but postpubertal or adult exposure to such stimulants would increase the response to later challenges (Andersen, 2003). Because the childhood maltreatment we measured was reported retrospectively, it may not be possible to accurately measure maltreatment at the window of opportunity. On top of that, it should be kept in mind that evidence on the integrated hypothesis is currently sparse, with the main evidence primarily focusing on animal models with strict environmental control (Daskalakis et al., 2013), and therefore it remains to be explored and validated in

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human studies. As highlighted by Hoogland & Ploeger (2022), humans are developing in complex and quickly changing environments, and it seems impossible to program children for all the different stressors that they may encounter during life. Additionally, there is another view that childhood maltreatment might be considered as a severe, but not moderate, stressor, which is more likely to overwhelm an individual's ability to manage recent stress rather than to trigger adaptive functions, i.e., exerting only cumulative stress effect (Shapero et al., 2015). More research is needed to tackle these controversies and meanwhile it should be important to note that, as proposed by Liu (2015), individual differences may exist in what constitutes a moderate stressor that triggers stress inoculation/ steeling effect.

Notably, compared to adolescents carrying fewer plasticity alleles of HPA-axis related genes, adolescents carrying more plasticity alleles were found to be applicable to the cumulative stress hypothesis. This result was in line with the finding of one recent study (Starr et al., 2021) that the interaction of childhood adversity and recent stress functioned in a cumulative stress manner only among adolescents ($M_{age} = 15.90$) with more but not fewer plasticity alleles of HPA-axis related genes. These results seem to provide preliminary evidence that genetic susceptibility further strengthens the cumulative stress hypothesis. In other words, in the complex psycho-biological development of individuals, exposure to severe childhood maltreatment could increase vulnerability to later stress (Huh et al., 2014; Rousson et al., 2020), especially for individuals with high genetic susceptibility. Notably, the social support systems of these adolescents might be disrupted to some extent after exposure to 2-hits of environmental (interpersonal) stress (Starr et al., 2019) and these factors may together enhance the risk for maladaptation (depressive episodes) in life.

Additionally, none of the $G \times E_1 \times E_2$ interactions regarding individual SNPs were significant after multiple correction, suggesting that the predictive validity and power of the MGPS was stronger than that of any individual SNPs. This finding provided further support for the claim that the genetic plasticity towards the interaction between early and recent environmental stress underlying psychopathology may manifest as an additive effect of multiple genetic variants (Feurer et al., 2017; Starr et al., 2021).

Regarding specific domains of stress, we found that only recent interpersonal stress, but not noninterpersonal stress drove the $G \times E_1 \times E_2$ interaction. This result was in line with previous research showing that HPA-axis MGPS moderates the effect of interpersonal, but not noninterpersonal, stress on depressive symptoms (Feurer et al., 2017; Starr & Huang, 2019) and related brain functions (Di Iorio et al., 2017). These findings together support that HPA-axis related genetic variants may be particularly sensitive to interpersonal stress and jointly increase the risk of depression, expanding the view of interpersonal theories of depression (Hames et al., 2013). Humans are a social species with a strong, fundamental, and widespread urge to establish and preserve healthy interpersonal relationships; therefore, sensitivity to interpersonal stress is a feature of humans that favors maximizing reproduction (Baumeister & Leary, 1995). Meanwhile, childhood maltreatment itself is also an important form of early interpersonal childhood adversity. According to attachment theory, early interpersonal relationships and communication patterns can serve as internal working models (secure vs. insecure) for future interpersonal relationships (Bowlby, 1982) and could more strongly affect adolescent emotional regulation and HPA-axis reactivity than noninterpersonal relationships (Poole et al., 2018; Stroud et al., 2016). Nevertheless, it should be pointed out that although the $G \times E_1 \times E_2$ interactions were found only in the domain of interpersonal stress, the differences between interactions regarding interpersonal versus noninterpersonal stress did not reach a significant level, and this remains to be validated.

The current study featured several strengths, including (i) the use of a theory-driven biological pathway-based HPA-axis related MGPS, (ii) interpersonal and noninterpersonal stress comparison, (iii) a multiwave, two-year prospective study design, (iv) HLM person-centred modeling, (v) a non-Western sample, and (vi) a series of sensitivity analyses. Based on these strengths, this study provided the first evidence that multiolocus HPA-axis genetic variation significantly interacted with the severity of childhood maltreatment and recent interpersonal stress to predict prospective changes in adolescent depressive symptoms in a cumulative stress manner.

Several limitations of this study should be carefully considered. First, as most genetic studies based on relatively large samples do (Duncan & Keller, 2011), self-report checklists were used to measure early and recent stress, which would result in significant disadvantages (Harkness & Monroe, 2016). For example, the self-report checklist does not rate stress exposure independently of stress response or depressive episodes, is unable to distinguish acute from chronic stressors or major from minor stressors, and does not provide anchor examples for rating standardization, resulting in greater measurement errors. Thus, blind-rater contextual interviews are strongly recommended to improve the current findings (Harkness & Monroe, 2016; Starr et al., 2021). Meanwhile, childhood maltreatment was assessed retrospectively, which may introduce recall bias. Although there is little possibility for the overlap of exposure time between childhood maltreatment and recent stress, future research can benefit from prospective assessment of early stress from prenatal to adolescence. Second, although our study used a theoretically driven MGPS, which goes beyond the traditional approach of individual candidate genes, the number of SNPs included was still small. Different SNPs are often used in research to yield different MGPSs (see Table S3), which is not conducive to comparisons between studies and the synthesis of evidence. Accordingly, standardization of procedures for constructing and replicating the MGPSs is greatly warranted (Pagliaccio et al., 2015). Otherwise, polygenic scores derived from GWAS can be used in the future, although this approach may not substantially increase the power versus the MGPS approach (Peyrot et al., 2014). Finally, this study focused on a community rather than a clinical sample, so the extent to which the results can be generalized to participants with a clinical diagnosis remains to be explored. Despite this, as highlighted by the transdiagnostic models, depression is not an all-or-none phenomenon (Dalgleish et al., 2020), and the current findings might inform preventive intervention strategies for community adolescents before they develop diagnostic depressive disorders. For instance, the findings emphasize the need to reduce early and recent interpersonal stress, which may serve as a modifiable risk factor in the pathway to depressive symptoms especially for those carrying more HPA-axis plasticity alleles. In addition, the current findings should be treated with caution before replication in independent samples and other racial and cultural groups. In particular, future research should utilize data from more time points with low levels of attribution to validate this complex multilevel model about three-way $G \times E_1 \times E_2$ interactions, which could decrease the potential risk of just identification, overfitting, poor generalization and poor power. Further, more time point data are beneficial for the examination of other growth models (Wright, 2017).

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Conclusion

This study provides preliminary evidence that HPA-axis multilocus genetic variants jointly interact with childhood maltreatment and recent interpersonal stress in the prediction of prospective changes in adolescent depressive symptoms, and the specific interaction form functions in the manner of cumulative stress. These findings highlight the cumulative risk mechanism regarding $G \times E_1 \times E_2$ interactions that underlie the longitudinal development of adolescent depressive symptoms and show effects specific to interpersonal stress.

Supplementary material. The supplementary material for this article can be found at https://doi.org/10.1017/S0954579424000269.

Data availability statement. The data that support the findings of this study are available from the corresponding author upon reasonable request.

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