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Psychiatric Symptoms in Adolescents: *FKBP5* Genotype - Early Life Adversity Interaction effects

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Abstract

Psychiatric disorders are multi-factorial and their symptoms overlap. Constitutional and environmental factors influence each other, and this contributes to risk and resilience in mental ill-health. We investigated functional genetic variation of stress responsiveness, assessed as *FKBP5* genotype, in relation to early life adversity and mental health in two samples of adolescents.

One population-based sample of 909 12-year-old adolescents (SESBiC) was assessed using the Life Incidence of Traumatic Events scale (LITE) and the Strengths and Difficulties Questionnaire (SDQ). One sample of 398 17-year-old adolescents, enriched for poly-victimized individuals (USSS), was assessed using the Juvenile Victimization Questionnaire (JVQ) and the Trauma Symptom Checklist for Children (TSCC). The *FKBP5* rs1360780 and rs3800373 polymorphisms were genotyped using a fluorescence-based competitive allele-specific PCR.

Most prominently among poly-victimized older male adolescents, the least common alleles of the polymorphisms, in interaction with adverse life events, were associated with psychiatric symptoms, after control of ethno-socio-economic factors. The interaction effect between rs3800373 and adverse life events on the TSCC sub-scales - anxiety, depression, anger, and dissociation - and with the rs1360780 on dissociation in the USSS cohort remained significant after Bonferroni correction.

This pattern of association is in line with the findings of clinical and neuroimaging studies, and implies interactive effects of *FKBP5* polymorphisms and early life environment on several psychiatric symptoms. These correlates add up to provide constructs that are relevant to several psychiatric symptoms, and to identify early predictors of mental ill-health.

Introduction

Adolescence is a key developmental period: the brain is still developing, and pubertal changes together with typical behavioural traits and environmental cues can influence susceptibility to the development of psychopathology and related behavioural symptoms [1]. Furthering knowledge of the underlying pathological mechanisms of psychiatric symptoms at an early age is of considerable relevance for timely identification of individuals at risk who might benefit from targeted intervention [2].

Behavioural problems and psychiatric symptoms are more common among children who have been abused and neglected [3]. In Sweden, victimisation is common among adolescents with an average of 2.5 events of victimization during the previous year, and of 4.5 events as lifetime victimization measured at the age of seventeen [4]. However, not all children who are exposed to early life adversity develop psychiatric symptoms. Suggestive evidence has been provided of the interactive contribution of the psychosocial background and genetic makeup of the child to proneness to clinical and subclinical psychiatric symptoms during adolescence [5].

Concomitant to experimental findings on laboratory animals, human studies demonstrate that early life adversity has a strong impact on stress response, behaviour, and ultimately mental health. Stress response is a crucial link between gene, environment and mental health, and is very influential, especially during critical periods of life, such as during brain development. Many psychiatric disorders are characterized by an impaired stress response. Interestingly, a robust constellation of findings on the effects of functional polymorphisms of the heat-shock protein 90 FK506 binding protein 5 (*FKBP5*) gene, in interaction with early life stress, on stress response as well as on psychopathology has emerged during the last decade [6, 7]. The FKBP5 protein is a co-chaperone regulator of (and regulated by) the glucocorticoid receptor, and thus in turn a modulator of the GR-mediated regulatory feed-back on the hypothalamic-pituitary-adrenal (HPA) axis in response to stress [6, 7].

Suggestive evidence of functionality has been provided for one polymorphism of the *FKBP5* gene, SNP rs1360780. Homozygosity for the minor allele T of the rs1360780 SNP is associated with twice the amount of FKBP5 protein levels in lymphocytes, relative to the other genotypes [8]. The T allele or TT genotype of the SNP rs1360780 (and consequentially the minor allele of the closely linked SNP rs3800373) has been associated with endocrine correlates: higher FKBP5 protein levels

[8]; higher *FKBP5* mRNA - plasma cortisol positive correlation [8-12]; reduced basal cortisol levels [8, 9, 12-15]; differential glucocorticoid-mediated response [12, 15, 16]; and heightened cortisol response after psychological stress in adults, adolescents and infants (in some cases moderated by childhood adversity) [10, 11, 17, 18]. Clinical associations have also been reported for this allele with better anti-depressant treatment response [8, 10, 19, 20]; greater risk of depression [12, 14, 15, 21, 22], suicidal tendencies [8, 23], comorbid anxiety-depression [24], post-traumatic stress disorder [6, 15, 23], and alcohol withdrawal severity [25]; as well as with major depression, aggressive behaviour, suicide attempts, and psychosis in an interaction with adverse life events [15, 26-28]. Moreover, associations with neural correlates of vulnerability to psychopathology have been demonstrated for this allele, such as higher amygdala reactivity [29], and increased threat and negative emotion-related amygdala reactivity in the context of childhood adversity [30]. Negative findings have, however, also been reported [7, 31]. Regarding GWAS studies, two *FKBP5* SNPs were among the top ten hits in a GWAS study on daily cortisol secretion, but they did not survive correction for multiple testing [13], whereas no GWAS-significant hits corresponding to SNPs in the *FKBP5* gene have been so far related to psychiatric phenotypes. All together, these evidences make the *FKBP5* a candidate gene, and the rs1360780 and rs3800373 SNPs candidate markers for studying the effect of constitutional factors of stress response and early adverse life events on adolescent mental health [32].

Aims of the study

The overall aim of the present study was to promote understanding of how early life stress and *FKBP5* polymorphisms related to stress response, interact and contribute to mental health in a population-based sample of adolescents, and among adolescents exposed to multiple victimisation experience. This study aimed to contribute to answering the following questions: How do environment and stress-related genetic factors interact in relation to propensity to the development of psychiatric symptoms?; Is the interactive effect relevant for several psychiatric symptoms?; Does sex play a role in these interactive influences?

Our hypothesis was that children who grew up in traumatic circumstances and with certain genetic variants have increased vulnerability to the development of psychiatric disorders, compared with those with protective or less environmentally sensitive genetic variants, and who have not experienced traumatic events. Furthermore, we hypothesize that the interaction between genotype and early life stress will be widespread, thus

indicating a vulnerability model beyond symptom categories.

Materials and methods

Two independent samples were used: i) adolescents from the 12-year follow-up of a longitudinal study of a large cohort monitored since birth (SESBiC), and ii) adolescents with a mean age of 17 from a large community-based cross-section study (USSS). Ethical approval was received for the SESBiC study (LU 439-93; LU 124-98; M51-07), and for the USSS study (69-07).

South East Sweden Birth Cohort study (SESBiC)

All mothers of children born between 1 May 1995 and 31 December 1996 in five counties in southern Sweden, with a population of just over 1% of all Swedes, were asked to participate in the SESBiC study and 1,723 mothers (88%) agreed to participate [33-37]. The present data come from the 12 year follow-up (mean age 12.0 years, $SD \pm 0.30$). An information letter and a consent form were sent to the parents (i.e. legal guardians). A separate, simplified information letter was enclosed for the child. Two children were deceased, 10 had moved out of the country and 24 had learning problems and were therefore unable to participate. A total of 909 individuals (53.9%) agreed to participate in the study. The follow-up was carried out at school where research assistants met with the children in small groups and assisted them in answering questionnaires and also in providing a sample of their saliva. The mothers were asked to fill in a package of questionnaires.

Upper Secondary School study (USSS)

The upper secondary school in Sweden offers three programmes: 1. Higher education preparatory programmes typically humanities, natural science, and social science ($\approx 44\%$ of all students); 2. Vocational programmes, typically health and social care, building and construction, hotel and tourism studies ($\approx 54\%$ of all students); and 3. Introductory programmes, typically preparatory education, programme-oriented individual options, vocational introduction, individual options, and language introduction, a resource-providing merit for students with different kinds of learning difficulties ($\approx 2\%$ of all students) [38]. A total of 51 out of 53 schools participated in the survey and a total of 6,096 students (78%) were present at the scheduled class-room survey.

The USSS adolescent sample was selected from this community-based sample ($\approx 5\%$), of students in the 2nd year upper secondary school system, evenly

distributed among structural parameters such as population, commuting patterns and economic structure [4] to represent the national average concerning, sex, birthplace, enrolment from other municipalities and educational programmes. Assuming that students taking the higher education preparatory programmes would have experienced less adverse life events [39], over-sampling of youths with more serious psychosocial problems was achieved by including more students from the vocational and introductory programmes (13% and 23% of all students in these programmes, respectively). The mean age was 17.2 years ($SD \pm 0.68$).

Questionnaires

The questionnaires were answered by the participants during class hours at school: data on demographics and psychosocial factors; history of the adolescent's exposure to lifetime adverse life events using the Life Incidence of Traumatic Events (LITE) [40] in the SESBiC study, and the Juvenile Victimization Questionnaire (JVQ) [41] in the USSS study. The majority of children has experienced at least one adverse life events during childhood and adolescence [42], and in Sweden 63.0% of children up to 12 years (no sex difference) had experienced at least one adverse life event during their lifetime, i.e. two or more events could be considered moderate exposure to trauma [43]. Of 17 year olds 84.1% (83.0% young men and 85.2% young women) had experienced at least one victimization event during their lifetime, and 10.3% had experienced 10 or more events and were categorized as poly-victimized (8.1% young men and 12.5% young women) [4]. Since adverse life events were so common, the number of life events at and above the 90th percentile was used in the analyses to catch the most vulnerable group. Symptoms of mental ill-health were assessed by scientifically validated screening tools: the Strengths and Difficulties Questionnaire (SDQ) [44] in the SESBiC study, and the Trauma Symptom Checklist for Children (TSCC) [45] in the USSS study. In the SESBiC study, data on immigrant status and the parent's occupation status were taken from questionnaires filled in by the mother. In the USSS study, participants were asked for information about birth-place, parents' birthplaces, parents' employment, and residence. LITE [43, 46], JVQ [4], SDQ [47], and TSCC [43, 46] are all scientifically validated screening tools with good psychometric properties that have been translated into Swedish and for which Swedish norms have been established.

Genetic analyses

DNA was extracted according to the manual from saliva samples collected using DNA Self Collection Kit (Oragene®). Genotyping of the polymorphism

of interest *FKBP5* SNPs rs1360780, and of the highly linked SNP rs3800373, was performed using a fluorescence-based competitive allele-specific PCR (KASPar) assay (KBioscience®). Allele discrimination was done using SNPviewer2®. The genotype calling was performed blind with regard to psychosocial data. To estimate the quality-rate of genotyping errors, a random repetition of ~13% of the population-based sample was carried out; the comparison indicated no inconsistencies. Diploypes were estimated with the EM algorithm using SNP & Variation Suite 7 (GoldenHelix®). Genotype frequencies are shown in Table S2. The genotypes were in Hardy-Weinberg equilibrium (Table S2).

SESBiC: Due to internal drop-out, the number of DNA samples from the 909 children that could be analysed, together with the data from the instruments, was $n = 888$ for *FKBP5* rs1360780 and $n = 902$ for rs3800373, and $n = 881$ in the haplotype analysis.

USSS: In all 398 adolescents participated. The number of DNA samples that could be analysed together with questionnaire data was $n = 394$ for both *FKBP5* rs1360780 and rs3800373 (four DNA samples were missing for each polymorphism) and $n = 390$ in the haplotype analysis. 81% of the students were in the vocational programme and 19% in the introductory programme.

Data analysis

The statistical power has been computed using the Genetic Power Calculator (<http://pengu.mgh.harvard.edu/~purcell/gpc/>) [48]. Since rs1360780 has been the SNP of interest and it is in high LD with rs3800373, the alpha was kept at 0.05. In an additive model, the power for a test with heritability equal to 1% is 85.3%. Thus, the size of the population-based cohort (SESBiC) outdoes the sample size of 781 subjects required for a power of 80%. On the other hand, the USSS cohort is underpowered, although it included more individuals reporting adverse life events, which is a known risk factor for psychopathology. Oversampling for individuals in the high-risk strata has certainly high efficacy in enhancing the power of a study. Group differences were tested using crosstab statistics and Pearson chi-square (χ^2) for categorical data and t-test (two-sided) for continuous variables. Bivariate correlations were tested, computing the Pearson's correlation coefficient. Genotype group differences in psychiatric symptoms were tested using univariate analysis of variance [49]. Multi-level statistical modelling was used to describe the interaction between environmental and genetic factors. The adjusted multivariate gene-by-environment

interaction models were analysed by applying general linear models (i.e. Univariate analysis of variance with Covariates, ANCOVA) using psychiatric symptoms as a scale variable. Genotypes were also grouped as homozygous and heterozygous for the T (rs1360780) or C (rs3800373) allele in one group and compared with homozygous for the major allele addressing the statistical constraint imposed by the low frequency of the minor alleles. Descriptive statistics were performed and presented for all three genotype groups. The environmental factor - adverse life events - was dichotomized according to [50], into less than 90th percentile vs. 90th percentile and above for the JVQ (10 or more events) and LITE (5 or more events) scales, respectively. As covariates: sex, living with both parents or not, immigrant status, and the parent's occupational status were used to account for known bias factors. Bonferroni correction was applied according to the number of psychobiologically meaningful questions tested and effectively independent SNPs, which is one marker when testing GxE association with the sub-scales (for the SDQ $p \leq 0.01$ and for the TSCC $p \leq 0.0083$). Effect sizes (Cohen's d) were calculated as the difference of mean score on SDQ/TSCC total scores for the risk genotype between no environmental load and high negative environmental load (90th percentile on LITE/JVQ) divided by the SD for the respective symptom measures. To further validate the findings, partial correlations, adjusted for sex, were performed to test for genotype-dependent environment effects on mental health using continuous measures, and to calculate the difference of correlation coefficients (ρ) [23]. Linkage disequilibrium (r^2 and D') and haplotypes were estimated with the EM algorithm using SNP & Variation Suite 7 (GoldenHelix®).

Results

Descriptive characteristics

Demographic and genotypic characteristics of the SESBiC (909 (50.1% boys) 12-year-old adolescents) and USSS (398 (54.3% boys) 17-year-old adolescents) cohorts are presented in the Supplementary Tables. In the SESBiC cohort the mean age was 12.0 ± 0.3 years, and in the USSS cohort 17.2 ± 0.7 years. The adolescents in the USSS cohort, as expected, had a higher percentage of parents born outside the country of residence ($p < 0.001$), a lower percentage had both parents with a job ($p < 0.001$), and fewer of them lived with their biological parents ($p = 0.002$) compared to the SESBiC cohort (Table S1). Genotype frequencies did not differ significantly between the two cohorts (Table S2).

The number of adverse life events among the adolescents in the SESBiC cohort (measured as LITE) ranged from 0 to 10 with a mean of 2.7 ± 1.8 , while in the USSS cohort (measured as JVQ) ranged from 0 to 32 with a mean of 4.6 ± 5.0 . The mean score of total psychiatric symptoms in the SESBiC cohort was 10.6 ($SD \pm 5.76$) (measured as SDQ total) and in the USSS cohort 31.6 ($SD \pm 23.50$) (measured as TSCC total) (Table S3 and S3b).

Adverse life events were as common in girls as in boys in the SESBiC cohort, whereas in the USSS cohort females reported more events ($p = 0.040$). Regarding mental health symptoms, in both cohorts sex differences were found in the SDQ and TSCC total scores as well as in the sub-scales, though more marked in the USSS cohort (table S3a and S3b). Symptoms of conduct and hyperactivity/inattention problems were more common among males, while symptoms of emotional problems and more positive prosocial behaviour were more often reported by females in the SESBiC cohort (Table 3a). Symptoms of sexual concerns were more common among males, while symptoms of anger, anxiety, depression, dissociation and posttraumatic stress were more often reported by females in the USSS cohort (Table 3b). The correlation between the SDQ total score in the SESBiC cohort and the different SDQ subscales was low to high ($r = 0.363 - 0.912$; $p < 0.001$), while between the TSCC total score in the USSS cohort and all the different TSCC subscales correlation was high ($r = 0.687 - 0.900$; $p < 0.001$) (Table S4).

Main effects

Both sexes reported higher scores on SDQ and TSCC total score and subscales if they had experienced adverse life events at or above the 90th percentile ($p = 0.044$ to $p < 0.001$ and $p = 0.023$ to $p < 0.001$; respectively). The main genotype effects on SDQ and TSCC scores were not present, except in the USSS cohort, where carriers of the minor allele of the *FKBP5* rs3800373 SNP reported significantly more psychiatric symptoms ($p = 0.037$; $F = 3.331$) (Table S5).

Gene-by-Environment interaction effects (GxE)

In the SESBiC cohort, a GxE interaction effect of *FKBP5* rs3800373 was observed on the SDQ total score ($p = 0.019$; $R^2 = 0.030$). Among adolescents with adverse life events at or above the 90th percentile, C carriers displayed the highest SDQ score, compared to the AA genotype, after adjustment for potential confounders. No sex differences were present, and genotype main effects were absent, as well as the effects of rs1360780 genotype (Table 1). At the sub-scales level, there

was only a nominally significant GxE effect on hyperactivity/inattention (Table S6a).

In the USSS cohort, both a gene main effect as well as a GxE interaction effect was found in the total TSCC score. The rs3800373 C carrier and rs1360780 T carrier genotypes were associated with higher scores among adolescents who had experienced adverse life events at or above the 90th percentile compared to individuals who were homozygous for the AA and CC genotypes, respectively, adjusted for potential confounders ($p = 0.001$; $R^2 = 0.260$ and $p = 0.007$; $R^2 = 0.254$; respectively) (Table 1). Nominally significant results in accordance with the same pattern of association were found for the sub-scales for Anger, Anxiety, Depression, Dissociation and Posttraumatic stress, but not for Sexual concerns (Table S6b). After Bonferroni correction, only the rs3800373xE effect on Anxiety, Depression, Anger, and Dissociation, and the rs1360780xE on Dissociation remained significant. When splitting the sample by sex because of its statistical significance in the model ($p < 0.001$), the GxE effects were only found in males (data not shown).

The effect sizes were highest for the rs3800373 and rs1360780 SNPs in the USSS cohort ($d = 0.76$ and $d = 0.56$, respectively), and moderate for the rs3800373 SNP in the SESBiC cohort ($d = 0.47$), with individuals carrying the minor allele of these SNPs having greater psychiatric symptoms than homozygous individuals for the most common alleles. In both cohorts, GxE effects were present in a genotype dose-dependent manner, with heterozygous individuals displaying intermediate scores in the presence of adverse life events, compared to both homozygous genotypes (Figure 1).

Gene-dependent environment-psychiatric symptoms correlation effects

Partial correlations, adjusted for sex, substantially corroborated these findings in the USSS cohort but not in the SESBiC cohort. High correlations were present between the adverse life events and psychiatric symptoms among homozygous carriers of the minor allele of the rs3800373 and rs1360780 SNPs in the USSS cohort ($r = 0.888$; AA/CC: $Z = 4.45$; and $r = 0.841$; CC/TT: $Z = 4.19$, respectively) (Table S7). Thus, this confirms that the interaction effect between genotype and adverse life events in psychiatric symptoms is characterized by a genotype, as well as a stress-dosage load on number of psychiatric symptoms in adolescents.

Haplotype analyses

As the two markers are in high Linkage Disequilibrium ($D' = 0.98$ and $R^2 = 0.86$ in the SESBiC cohort, and $D' = 0.98$ and $R^2 = 0.82$ in the

IUSS cohort), the analyses were re-performed using diplotypes, which are inferred combinations of haplotypes (Table S8). As expected, a significant GxE was only present in the USSS cohort, with carriers of the diplotype containing the minor alleles of the rs3800373 and rs1360780 SNPs reporting more psychiatric symptoms if exposed to adverse life events ($p = 0.021$; $R^2 = 0.255$) (Table S9). Lastly, GxE effects were more evident in males (data not shown).

Discussion

The present study investigated the interactive effect of genetic factors of stress responsiveness and early life adversity on psychiatric symptoms in two independent samples, one population-based sample of young individuals (SESBiC cohort) and one sample of older adolescents enriched for poly-victimised adolescents (USSS cohort). The findings indicate that the least common functional variations of the *FKBP5* gene, a regulator of glucocorticoid receptor function, in an interaction with adverse life events, are associated with symptoms of mental ill-health among both 12 and 17 year-old adolescents. This is in line with previous findings in adults of a *FKBP5* genotype-by-childhood maltreatment interaction effect on clinical phenotypes, such as depression, PTSD, psychosis, suicide attempts, and on aggressive behaviour [18, 22, 23, 26, 28, 51, 52], suicidal tendencies, as well as on threat-related right dorsal amygdala reactivity in adolescents reactivity [30].

Key-features of psychiatric disorders, as defined by the DSM, are their phenotypical and biological heterogeneity, complexity, overlap, and interdependence. Our hypothesis was confirmed: adolescents who grew up in adverse circumstances, and with certain genetic variants, report a higher level of psychological symptoms, and that this interaction between genotype and early life stress illustrates a vulnerability model beyond symptom categories. On the other hand, those with protective or less environmental sensitive genetic variants and who have not experienced traumatic events display decreased propensity for the development of psychiatric disorders.

Too little attention has been devoted to children and adolescents as victims, in view of the social and personal burden of the consequences, and the cost-benefit advantage of early interventions. The emphasis in the present study has been on individuals at a particularly crucial time of life and of brain development (i.e. adolescence), and in fragile conditions (i.e. poly-victimisation). Furthermore, sex differences which are common in

psychiatric disease vulnerability, presentation, and outcomes, were also investigated.

To our knowledge, no *FKBP5* gene by adverse life events study on a broad spectrum of psychopathology symptoms has been performed for young adolescents and poly-victimised adolescents. Hence the present study presents novel findings valuable in the context of future meta-analyses. The similar results found in two independent and relatively large cohorts, despite the use of different self-reporting instruments, corroborate each other, providing support for their robustness. They are in line with the previous results of clinical, psychophysiological and neuroimaging studies for *FKBP5*, indicating the T allele as a risk factor for psychopathology [6, 8, 10, 12, 14, 15, 19-27, 29, 30]. In fact, the TT genotype of rs1360780, which leads to higher FKBP5 protein levels [7, 8], has been associated with an altered physiological stress response and heightened risk of stress [6-18]. Binder and colleagues suggested a putative enhancing function on gene transcription for the T allele by the formation of a TATA box, as well as differential chromatin conformations and interactions of long-range enhancers with the transcription start site, which would influence the response to the glucocorticoid receptor activation triggered by early life adversity [6].

Results were more prominent in males in the USSS cohort. Sex differences in neuroscience exist in relation to brain anatomy, function, chemistry and also the prevalence and nature of psychiatric disorders [53-55]. Substantial evidence indicates that psychosocial factors interact with sex/sex hormones, and with the genetic make-up, to influence individual stress response to environmental stressors, which is an important factor linked to psychiatric disorders. Indeed, several genetic association studies on psychiatric symptoms reported sex effects mediating gene-by-environmental interactions on adolescent mental health, thus stressing the importance of considering sex differences in biological psychiatry. In the present study, the GxE effects were more evident in males, although the direction of association was similar in females. Interestingly, the rs3800373 genotype displayed an effect in male but not female young adults on cortisol response to psychosocial stress [56]. The FKBP5 is a co-chaperone of the progesterone and androgen receptor complex [7], thus making sex dimorphic influences plausible.

Gene by environment interactions in psychiatry still debated today, and the biological underpinnings of these interactions remain largely unknown [2, 57, 58]. It has been suggested that the *FKBP5* gene variants examined in the present study have genetic and endocrine functional effects, and ultimately

predispose psychiatric disorders [7].. Differential cortisol recovery response after psychosocial stress has been demonstrated in young adults [10, 59], and also cortisol reactivity to stressful situations in infants, depending on the *FKBP5* genotype [11]. Binder and colleagues have recently provided evidence of a childhood trauma-dependent epigenetic mechanism [6], by demonstrating an allele-specific, childhood trauma, dependent DNA methylation of *FKBP5* for proneness to stress-related psychiatric disorders [6]. Thus, this makes it one putative mechanism behind the *FKBP5* gene by adverse life events effect on mental ill-health observed in the present study.

The SESBiC cohort is a representative sample of young Swedish adolescents [36], while the USSS cohort comprises a group of adolescents with a greater risk of having experienced more adverse life events [60, 61], as reflected by the fact that the $\geq 90^{\text{th}}$ percentile of ALE in the SESBiC cohort included 5 or more events, while in the USSS cohort it comprised 10 or more. The interaction between *FKBP5* SNP rs1360780 and rs3800373 with adverse life events was associated with mental health problems, after controlling for sex and ethno-socio-economic factors. This was particularly prominent among poly-victimized older male adolescents in the USSS cohort, probably due to a cumulative effect of age and multiple exposure to adverse life events which, combined with genetic vulnerability, results in more psychiatric symptoms. Time, type, and dosage of environmental exposure to adverse life events are certainly of importance for the development of posttraumatic symptomatology [3]. Retrospectively assessed early life adversity, in interaction with *FKBP5* genotype, but not adult trauma, has for instance been shown to predict psychopathology [62], thus stressing the importance of early investigations.

The strengths of the present study are: i) the inclusion of two relatively large independent samples; ii) the investigation of adolescent individuals, which permits identification of early predictors and reduces the retrospective time interval for early life adversity recall; iii) the use of well-validated screening instruments; iv) the study of a broad range of mental health symptoms; and v) the comparability of results obtained with different study set-ups, phenotypic outcome variables, and the nature of the environmental factor. The limitations include a relatively high drop-out of individuals who are often at the extreme end of the phenotypic spectrum; the cross-sectional retrospective study design; and the loss of statistical significance of the results related to the sub-scales if correction for multiple-testing is applied, although the genetic and statistical analyses have been unidirectionally hypothesis-driven.

In contrast with the proportion of phenotypic variance estimated by heritability studies, single common genetic variants have been shown to account for only a very small proportion of variance for complex behavioural and psychiatric phenotypes [63], presumably because environmental factors are not taken into account of. False positive findings, together with the low statistical power of many studies, have hindered our understanding of the pathological underpinning of psychiatric disorders and have restricted advances in clinical practice [2, 64]. As highlighted by Munafo et al. and Duncan and Keller [2, 65], well-powered direct replications are needed to arrive at a robust, reliable and reproducible GxE finding, and also converging evidence from preclinical and neuroimaging studies should corroborate a GxE finding. Accordingly, the present study attempted to test a previous hypothesis and replication of potential findings in an independent sample, by using a similar set-up with regard to phenotypic variable, genetic polymorphisms, statistical model, environmental moderator, and inclusion of both sexes. The two studies were planned independently, and therefore the assessment tools differ. While, on the one hand, the employment of the same tools in both studies would have been highly desirable, it is a strength of the present results that similar findings are obtained, independently of which measure is used. This mirrors what often happens in the literature, where multi-site studies employing the same protocol are rare and the resources demanding. It is true that methodological differences are often claimed as the explanatory factor for discrepancies in the results of many genetic association studies. However robust results should not be biased by such minor variances. It is important to note that, subsequently to correction for multiple-testing, most of the results related to the sub-scales can only be deemed nominally significant, thus calling for independent replications. On the other hand, since the dimensions of the sub-scales are interrelated, the total SDQ and TSCC scores serve the purpose of the present study better, i.e. to investigate genetic correlates of a psychiatric dimensional measure across its full range. The statistically significant findings in the USSS cohort were associated with moderate effect sizes. Only a very small proportion of phenotypic variance can be explained by single common genetic variants [63], and gene by gene interaction should also be considered [2], thus genetic score indexes and GWAS approaches are highly desirable.

To conclude, these findings heighten the generalisability of previous results in adult clinical samples, and of neuroimaging findings, to adolescents' behaviour and psychiatric symptoms.

Moreover, in line with more and more frequent evidence of overlapping genetic constructs across psychiatric disorders [49, 66], the present GxE findings suggest a shared GxE effect across different mental health problems, moving beyond strictly immobile psychiatric definitions and categories, towards a nosology informed by neurobiology of the disease [2], although no causality relationship could be addressed.

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Conflict of interest

None declared by any of the authors.

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References

1. Spear LP (2000) The adolescent brain and age-related behavioral manifestations. *Neurosci Biobehav Rev* 24:417-463
2. Munafo MR, Zammit S, Flint J (2014) Practitioner Review: A critical perspective on gene-environment interaction models - what impact should they have on clinical perceptions and practice? *J Child Psychol Psychiatry*
3. Heim C, Binder EB (2012) Current research trends in early life stress and depression: review of human studies on sensitive periods, gene-environment interactions, and epigenetics. *Exp Neurol* 233:102-111
4. Aho N, Gren-Landell M, Svedin CG (2014) The Prevalence of Potentially Victimizing Events, Poly-Victimization, and Its Association to Sociodemographic Factors: A Swedish Youth Survey. *J Interpers Violence*
5. Manuck SB, McCaffery JM (2014) Gene-environment interaction. *Annu Rev Psychol* 65:41-70
6. Klengel T, Mehta D, Anacker C, Rex-Haffner M, Pruessner JC, Pariante CM, Pace TW, Mercer KB, Mayberg HS, Bradley B, Nemeroff CB, Holsboer F, Heim CM, Ressler KJ, Rein T, Binder EB (2013) Allele-specific FKBP5 DNA demethylation mediates gene-childhood trauma interactions. *Nat Neurosci* 16:33-41
7. Zannas AS, Binder EB (2014) Gene-environment interactions at the FKBP5 locus: sensitive periods, mechanisms and pleiotropism. *Genes Brain Behav* 13:25-37
8. Binder EB, Salyakina D, Lichtner P, Wochnik GM, Ising M, Putz B, Papiol S, Seaman S, Lucae S, Kohli MA, Nickel T, Kunzel HE, Fuchs B, Majer M, Pfennig A, Kern N, Brunner J, Modell S, Baghai T, Deiml T, Zill P, Bondy B, Rupprecht R, Messer T, Kohnlein O, Dabitz H, Bruckl T, Muller N, Pfister H, Lieb R, Mueller JC, Lohmusaar E, Strom TM, Bettecken T, Meitinger T, Uhr M, Rein T, Holsboer F, Muller-Myhsok B (2004) Polymorphisms in FKBP5 are associated with increased recurrence of depressive episodes and rapid response to antidepressant treatment. *Nat Genet* 36:1319-1325
9. Menke A, Klengel T, Rubel J, Bruckl T, Pfister H, Lucae S, Uhr M, Holsboer F, Binder EB (2013) Genetic variation in FKBP5 associated with the extent of stress hormone dysregulation in major depression. *Genes Brain Behav* 12:289-296
10. Ising M, Depping AM, Siebertz A, Lucae S, Unschuld PG, Kloiber S, Horstmann S, Uhr M, Muller-Myhsok B, Holsboer F (2008) Polymorphisms in the FKBP5 gene region modulate recovery from psychosocial stress in healthy controls. *Eur J Neurosci* 28:389-398
11. Luijk MP, Velders FP, Tharner A, van Ijzendoorn MH, Bakermans-Kranenburg MJ, Jaddoe VW, Hofman A, Verhulst FC, Tiemeier H (2010) FKBP5 and resistant attachment predict cortisol reactivity in infants: gene-environment interaction. *Psychoneuroendocrinology* 35:1454-1461
12. Fujii T, Hori H, Ota M, Hattori K, Teraishi T, Sasayama D, Yamamoto N, Higuchi T, Kunugi H (2014) Effect of the common functional FKBP5 variant (rs1360780) on the hypothalamic-pituitary-adrenal axis and peripheral blood gene expression. *Psychoneuroendocrinology* 42:89-97

13. Velders FP, Kuningas M, Kumari M, Dekker MJ, Uitterlinden AG, Kirschbaum C, Hek K, Hofman A, Verhulst FC, Kivimäki M, Van Duijn CM, Walker BR, Tiemeier H (2011) Genetics of cortisol secretion and depressive symptoms: a candidate gene and genome wide association approach. *Psychoneuroendocrinology* 36:1053-1061
14. Zobel A, Schuhmacher A, Jessen F, Hofels S, von Widdern O, Metten M, Pfeiffer U, Hanes C, Becker T, Rietschel M, Scheef L, Block W, Schild HH, Maier W, Schwab SG (2010) DNA sequence variants of the FKBP5 gene are associated with unipolar depression. *Int J Neuropsychopharmacol* 13:649-660
15. Collip D, Myin-Germeys I, Wichers M, Jacobs N, Derom C, Thiery E, Lataster T, Simons C, Delespaul P, Marcelis M, van Os J, van Winkel R (2013) FKBP5 as a possible moderator of the psychosis-inducing effects of childhood trauma. *Br J Psychiatry* 202:261-268
16. Brent D, Melhem N, Ferrell R, Emslie G, Wagner KD, Ryan N, Vitiello B, Birmaher B, Mayes T, Zelazny J, Onorato M, Devlin B, Clarke G, DeBar L, Keller M (2010) Association of FKBP5 polymorphisms with suicidal events in the Treatment of Resistant Depression in Adolescents (TORDIA) study. *Am J Psychiatry* 167:190-197
17. Pagliaccio D, Luby JL, Bogdan R, Agrawal A, Gaffrey MS, Belden AC, Botteron KN, Harms MP, Barch DM (2014) Stress-system genes and life stress predict cortisol levels and amygdala and hippocampal volumes in children. *Neuropsychopharmacology* 39:1245-1253
18. Ajnakina O, Borges S, Di Forti M, Patel Y, Xu X, Green P, Stilo SA, Kolliakou A, Sood P, Marques TR, David AS, Prata D, Dazzan P, Powell J, Pariante C, Mondelli V, Morgan C, Murray RM, Fisher HL, Iyegbe C (2014) Role of Environmental Confounding in the Association between FKBP5 and First-Episode Psychosis. *Front Psychiatry* 5:84
19. Lekman M, Laje G, Charney D, Rush AJ, Wilson AF, Sorant AJ, Lipsky R, Wisniewski SR, Manji H, McMahon FJ, Paddock S (2008) The FKBP5-gene in depression and treatment response--an association study in the Sequenced Treatment Alternatives to Relieve Depression (STAR*D) Cohort. *Biol Psychiatry* 63:1103-1110
20. Kirchheiner J, Lorch R, Lebedeva E, Seeringer A, Roots I, Sasse J, Brockmoller J (2008) Genetic variants in FKBP5 affecting response to antidepressant drug treatment. *Pharmacogenomics* 9:841-846
21. Lavebratt C, Aberg E, Sjöholm LK, Forsell Y (2010) Variations in FKBP5 and BDNF genes are suggestively associated with depression in a Swedish population-based cohort. *J Affect Disord* 125:249-255
22. Appel K, Schwahn C, Mahler J, Schulz A, Spitzer C, Fenske K, Stender J, Barnow S, John U, Teumer A, Biffar R, Nauck M, Volzke H, Freyberger HJ, Grabe HJ (2011) Moderation of adult depression by a polymorphism in the FKBP5 gene and childhood physical abuse in the general population. *Neuropsychopharmacology* 36:1982-1991
23. Binder EB, Bradley RG, Liu W, Epstein MP, Deveau TC, Mercer KB, Tang Y, Gillespie CF, Heim CM, Nemeroff CB, Schwartz AC, Cubells JF, Ressler KJ (2008) Association of FKBP5 polymorphisms and childhood abuse with risk of posttraumatic stress disorder symptoms in adults. *Jama* 299:1291-1305
24. Minelli A, Maffioletti E, Cloninger CR, Magri C, Sartori R, Bortolomasi M, Congiu C, Bignotti S, Segala M, Giacomuzzi M, Gennarelli M (2013) Role of allelic variants of FKBP5-binding protein 51 (FKBP5) gene in the development of anxiety disorders. *Depress Anxiety* 30:1170-1176
25. Huang MC, Schwandt ML, Chester JA, Kirchhoff AM, Kao CF, Liang T, Tapocik JD, Ramchandani VA, George DT, Hodgkinson CA, Goldman D, Heilig M (2014) FKBP5 moderates alcohol withdrawal severity: human genetic association and functional validation in knockout mice. *Neuropsychopharmacology* 39:2029-2038
26. Bevilacqua L, Carli V, Sarchiapone M, George DK, Goldman D, Roy A, Enoch MA (2012) Interaction Between FKBP5 and Childhood Trauma and Risk of Aggressive Behavior. *Archives of General Psychiatry* 69:62-70
27. Szczepankiewicz A, Leszczynska-Rodziewicz A, Pawlak J, Narozna B, Rajewska-Rager A, Wilkosc M, Zaremba D, Maciukiewicz M, Twarowska-Hauser J (2014) FKBP5 polymorphism is associated with major depression but not with bipolar disorder. *J Affect Disord* 164:33-37
28. Roy A, Gorodetsky E, Yuan Q, Goldman D, Enoch MA (2010) Interaction of FKBP5, a stress-related gene, with childhood trauma increases the risk for

- attempting suicide. *Neuropsychopharmacology* 35:1674-1683
29. Fani N, Gutman D, Tone EB, Almlil L, Mercer KB, Davis J, Glover E, Jovanovic T, Bradley B, Dinov ID, Zamanyan A, Toga AW, Binder EB, Ressler KJ (2013) FKBP5 and attention bias for threat: associations with hippocampal function and shape. *JAMA Psychiatry* 70:392-400
 30. White MG, Bogdan R, Fisher PM, Munoz KE, Williamson DE, Hariri AR (2012) FKBP5 and emotional neglect interact to predict individual differences in amygdala reactivity. *Genes Brain Behav* 11:869-878
 31. Dunn EC, Solovieff N, Lowe SR, Gallagher PJ, Chaponis J, Rosand J, Koenen KC, Waters MC, Rhodes JE, Smoller JW (2014) Interaction between genetic variants and exposure to Hurricane Katrina on post-traumatic stress and post-traumatic growth: a prospective analysis of low income adults. *J Affect Disord* 152-154:243-249
 32. Binder EB (2009) The role of FKBP5, a co-chaperone of the glucocorticoid receptor in the pathogenesis and therapy of affective and anxiety disorders. *Psychoneuroendocrinology* 34 Suppl 1:S186-195
 33. Agnafors S, Comasco E, Bladh M, Sydsjo G, Dekeyser L, Orelund L, Svedin CG (2013) Effect of gene, environment and maternal depressive symptoms on pre-adolescence behavior problems - a longitudinal study. *Child Adolesc Psychiatry Ment Health* 7:10
 34. Agnafors S, Sydsjo G, Dekeyser L, Svedin CG (2013) Symptoms of depression postpartum and 12 years later-associations to child mental health at 12 years of age. *Matern Child Health J* 17:405-414
 35. Hook B, Cederblad M, Berg R (2006) Prenatal and postnatal maternal smoking as risk factors for preschool children's mental health. *Acta Paediatr* 95:671-677
 36. deKeyser L, Svedin CG, Agnafors S, Bladh M, Sydsjo G (2014) Multi-informant reports of mental health in Swedish-born children of immigrants and children born to non-immigrants - the SESBiC-study. *BMC Pediatr* 14:95
 37. Dekeyser L, Svedin CG, Agnafors S, Sydsjo G (2011) Self-reported mental health in 12-year-old second-generation immigrant children in Sweden. *Nord J Psychiatry* 65:389-395
 38. Skolverket (2011) <http://www.skolverket.se/om-skolverket/andra-sprak-och-lattlast/in-english/the-swedish-education-system>. In, Stockholm
 39. Breslau N, Kessler RC, Chilcoat HD, Schultz LR, Davis GC, Andreski P (1998) Trauma and posttraumatic stress disorder in the community: the 1996 Detroit Area Survey of Trauma. *Arch Gen Psychiatry* 55:626-632
 40. Greenwald R, Rubin A (1999) Assessment of posttraumatic symptoms in children: Development and preliminary validation of parent and child scales. *Research on Social Work Practice* 9:61-75
 41. Finkelhor D, Hamby SL, Ormrod R, Turner H (2005) The Juvenile Victimization Questionnaire: reliability, validity, and national norms. *Child Abuse Negl* 29:383-412
 42. Finkelhor D, Turner H, Ormrod R, Hamby SL (2009) Violence, abuse, and crime exposure in a national sample of children and youth. *Pediatrics* 124:1411-1423
 43. Gustafsson PE, Nilsson D, Svedin CG (2009) Polytraumatization and psychological symptoms in children and adolescents. *Eur Child Adolesc Psychiatry* 18:274-283
 44. Lundh LG, Wangby-Lundh M, Bjarehed J (2008) Self-reported emotional and behavioral problems in Swedish 14 to 15-year-old adolescents: a study with the self-report version of the Strengths and Difficulties Questionnaire. *Scand J Psychol* 49:523-532
 45. Briere J, Johnson K, Bissada A, Damon L, Crouch J, Gil E, Hanson R, Ernst V (2001) The Trauma Symptom Checklist for Young Children (TSCYC): reliability and association with abuse exposure in a multi-site study. *Child Abuse Negl* 25:1001-1014
 46. Nilsson D, Gustafsson PE, Svedin CG (2010) Self-reported potentially traumatic life events and symptoms of post-traumatic stress and dissociation. *Nord J Psychiatry* 64:19-26
 47. Smedje H, Broman JE, Hetta J, von Knorring AL (1999) Psychometric properties of a Swedish version of the "Strengths and Difficulties Questionnaire". *Eur Child Adolesc Psychiatry* 8:63-70
 48. Purcell S, Cherny SS, Sham PC (2003) Genetic Power Calculator: design of linkage and association genetic mapping studies of complex traits. *Bioinformatics* 19:149-150
 49. Lee SH, Ripke S, Neale BM, Faraone SV, Purcell SM, Perlis RH, Mowry BJ, Thapar A, Goddard ME, Witte JS, Absher D, Agartz I, Akil H, Amin F, Andreassen OA,

- Anjorin A, Anney R, Anttila V, Arking DE, Asherson P, Azevedo MH, Backlund L, Badner JA, Bailey AJ, Banaschewski T, Barchas JD, Barnes MR, Barrett TB, Bass N, Battaglia A, Bauer M, Bayes M, Bellivier F, Bergen SE, Berrettini W, Betancur C, Bettecken T, Biederman J, Binder EB, Black DW, Blackwood DHR, Bloss CS, Boehnke M, Boomsma DI, Breen G, Breuer R, Bruggeman R, Cormican P, Buccola NG, Buitelaar JK, Bunney WE, Buxbaum JD, Byerley WF, Byrne EM, Caesar S, Cahn W, Cantor RM, Casas M, Chakravarti A, Chambert K, Choudhury K, Cichon S, Cloninger CR, Collier DA, Cook EH, Coon H, Cormand B, Corvin A, Coryell WH, Craig DW, Craig IW, Crosbie J, Cuccaro ML, Curtis D, Czamara D, Datta S, Dawson G, Day R, De Geus EJ, Degenhardt F, Djurovic S, Donohoe GJ, Doyle AE, Duan JB, Dudbridge F, Duketis E, Ebstein RP, Edenberg HJ, Elia J, Ennis S, Etain B, Fanous A, Farmer AE, Ferrier IN, Flickinger M, Fombonne E, Foroud T, Frank J, Franke B, Fraser C, et al. (2013) Genetic relationship between five psychiatric disorders estimated from genome-wide SNPs. *Nat Genet* 45:984-992
50. Finkelhor D, Ormrod RK, Turner HA, Hamby SL (2005) Measuring poly-victimization using the juvenile victimization questionnaire. *Child Abuse & Neglect* 29:1297-1312
 51. Xie P, Kranzler HR, Poling J, Stein MB, Anton RF, Farrer LA, Gelernter J (2010) Interaction of FKBP5 with childhood adversity on risk for post-traumatic stress disorder. *Neuropsychopharmacology* 35:1684-1692
 52. Zimmermann P, Bruckl T, Nocon A, Pfister H, Binder EB, Uhr M, Lieb R, Moffitt TE, Caspi A, Holsboer F, Ising M (2011) Interaction of FKBP5 gene variants and adverse life events in predicting depression onset: results from a 10-year prospective community study. *Am J Psychiatry* 168:1107-1116
 53. Cahill L (2006) Why sex matters for neuroscience. *Nat Rev Neurosci* 7:477-484
 54. Cosgrove KP, Mazure CM, Staley JK (2007) Evolving knowledge of sex differences in brain structure, function, and chemistry. *Biol Psychiatry* 62:847-855
 55. Toffoletto S, Lanzenberger R, Gingnell M, Sundstrom-Poromaa I, Comasco E (2014) Emotional and cognitive functional imaging of estrogen and progesterone effects in the female human brain: a systematic review. *Psychoneuroendocrinology* 50:28-52
 56. Mahon PB, Zandi PP, Potash JB, Nestadt G, Wand GS (2013) Genetic association of FKBP5 and CRHR1 with cortisol response to acute psychosocial stress in healthy adults. *Psychopharmacology (Berl)* 227:231-241
 57. Feder A, Nestler EJ, Charney DS (2009) Psychobiology and molecular genetics of resilience. *Nat Rev Neurosci* 10:446-457
 58. Caspi A, Moffitt TE (2006) Gene-environment interactions in psychiatry: joining forces with neuroscience. *Nat Rev Neurosci* 7:583-590
 59. Buchmann AF, Holz N, Boecker R, Blomeyer D, Rietschel M, Witt SH, Schmidt MH, Esser G, Banaschewski T, Brandeis D, Zimmermann US, Laucht M (2014) Moderating role of FKBP5 genotype in the impact of childhood adversity on cortisol stress response during adulthood. *Eur Neuropsychopharmacol* 24:837-845
 60. Gren-Landell M, Aho N, Carlsson E, Jones A, Svedin CG (2013) Posttraumatic stress symptoms and mental health services utilization in adolescents with social anxiety disorder and experiences of victimization. *Eur Child Adolesc Psychiatry* 22:177-184
 61. Gren-Landell M, Aho N, Andersson G, Svedin CG (2011) Social anxiety disorder and victimization in a community sample of adolescents. *J Adolesc* 34:569-577
 62. Binder EB, Nemeroff CB (2010) The CRF system, stress, depression and anxiety-insights from human genetic studies. *Mol Psychiatry* 15:574-588
 63. Gratten J, Wray NR, Keller MC, Visscher PM (2014) Large-scale genomics unveils the genetic architecture of psychiatric disorders. *Nat Neurosci* 17:782-790
 64. Heininga VE, Oldehinkel AJ, Veenstra R, Nederhof E (2015) I just ran a thousand analyses: benefits of multiple testing in understanding equivocal evidence on gene-environment interactions. *PLoS One* 10:e0125383
 65. Duncan LE, Keller MC (2011) A critical review of the first 10 years of candidate gene-by-environment interaction research in psychiatry. *Am J Psychiatry* 168:1041-1049
 66. Smoller JW, Craddock N, Kendler K, Lee PH, Neale BM, Nurnberger JI, Ripke S, Santangelo S, Sullivan PF, Consortium PG (2013) Identification of risk loci with shared effects on five major psychiatric

disorders: a genome-wide analysis. *Lancet*
381:1371-1379

Figure 1

Relation of adverse life events (<90th percentile vs. ≥90th percentile) to estimated marginal means of psychiatric symptoms in adolescents (SDQ in the SESBiC cohort and TSCC in the USSS cohort) by *FKBP5* genotype

SESBiC cohort

GxE: $p = 0.054$ (**0.019[#]**)

GxE: $p = 0.232$ (**0.127[#]**)

USSS cohort

GxE: $p = 0.004$ (**0.001[#]**)

GxE: $p = 0.032$ (**0.007[#]**)

SDQ: Strength and Difficulties Questionnaire; TSCC: Trauma Symptom Checklist for Children

GxE: gene-by-environment interaction effect based on GLM model adjusted for: sex, living with both parents v.s. separated parents, both parents working v.s. ≥ one parent not working, both parents born in Sweden v.s. ≥ one parent born outside Sweden

[#] model with 2 levels of genotypes, homozygous for the most common allele vs. carriers of the minor allele

Table 1

Interaction effect of *FKBP5* genotype and adverse life events associated with psychiatric symptoms (assessed with SDQ and TSCC) in adolescents of the SESBiC and USSS cohorts, respectively

Cohort	<i>FKBP5</i> genotypes	Participants	ALE <90th percentile	Participants	ALE ≥90th percentile	GLM [§]	R ²
		N	mean SDQ score (SD)	N	mean SDQ score (SD)	<i>p</i>	
SESBiC	rs3800373					E = 0.010 ; G = 0.307; GxE = 0.048	0.018
	AA	449	10.5 (6.01)	68	11.4 (5.65)	E < 0.001 [#] ; G = 0.156 [#] ; GxE = 0.019 [#]	0.030 [#]
	AC	272	9.9 (5.37)	57	13.4 (5.06)		
	CC	52	10.2 (5.56)	4	14.8 (6.02)		
	rs1360780					E = 0.010 ; G = 0.424; GxE = 0.237	0.014
	CC	422	10.5 (5.93)	61	11.6 (5.70)	E = 0.003 [#] ; G = 0.320 [#] ; GxE = 0.127 [#]	0.025 [#]
	CT	281	10.1 (5.62)	61	12.9 (5.29)		
	TT	58	10.3 (5.62)	5	14.8 (5.22)		
		N	mean TSCC score (SD)	N	mean TSCC score (SD)	<i>p</i>	
USSS	rs3800373					E < 0.001 ; G = 0.005 ; GxE = 0.004	0.260
	AA	187	28.8 (19.84)	26	48.3 (26.77)	E < 0.001 [#] ; G = 0.004 [#] ; GxE = 0.001 [#]	0.260 [#]
	AC	136	25.8 (19.56)	20	61.7 (37.73)		
	CC	18	32.5 (18.50)	7	71.0 (12.78)		
	rs1360780					E < 0.001 ; G = 0.017 ; GxE = 0.032	0.251
	CC	169	27.7 (19.68)	25	48.9 (27.13)	E < 0.001 [#] ; G = 0.005 [#] ; GxE = 0.007 [#]	0.254 [#]
	CT	146	27.3 (20.09)	20	61.0 (34.14)		
	TT	25	30.7 (17.60)	9	65.0 (16.33)		

[§] GLM adjusted for: sex, living with both parents v.s. separated parents; both parents working v.s. ≥one parent not working; both parents born in Sweden v.s. ≥one parent born outside Sweden

[#] model with 2 levels of genotypes, homozygous for the most common allele vs. carriers of the minor allele

G: genotype main effect; E: environment main effect; GxE: gene-by-environment interaction effect; SDQ: Strength and Difficulties Questionnaire; TSCC: Trauma Symptom Checklist for Children; ALE: adverse life events

Supplementary Table 1
Demographic characteristics

	SESBiC (%)	USSS (%)	<i>p</i> [†]
Sex			
Male	50.1	54.3	n.s.
Secondary school			
Higher education preparatory programs		0	
Vocational program		81	
Introductory program		19	
Graduate school			
standard program	100		
Both parents born in Sweden	88.6	80.2	<0.001
Living with both biological parents	75.5	67.3	=0.002
Both parents working	81.2	61.6	<0.001

Chi square test

Supplementary Table 2

Descriptive statistics of *FKBP5* SNPs

Cohort	dbSNP ID	Genotype frequencies		Minor Allele Frequency	HWE <i>P</i>	Call rate %
		Males N (%)	Females N (%)			
SESBiC	rs3800373					96.7
	AA	258 (49.9)	259 (50.1)			
	AC	164 (49.9)	165 (50.2)	25% (M)	0.48 (M)	
	CC	31 (55.4)	25 (44.6)	24% (F)	0.85 (F)	
	rs1360780					95.4
	CC	235 (48.7)	248 (51.3)	28% (M)	0.60 (M)	
USSS	CT	173 (50.6)	169 (49.4)	25% (F)	0.80 (F)	
	TT	36 (57.1)	27 (42.9)			
	rs3800373					95.5
	AA	110 (51.6)	103 (48.4)			
	AC	93 (59.6)	63 (40.4)	27% (M)	0.12 (M)	
	CC	12 (48.0)	13 (52.9)	29% (F)	0.56 (F)	
	rs1360780					95.5
	CC	103 (53.1)	91 (46.9)			
	CT	95 (57.2)	71 (42.8)	30% (M)	0.44 (M)	
	TT	17 (50.0)	17 (50.0)	25% (F)	0.44 (F)	

F: females; HWE: Hardy-Weinberg Equilibrium; M: males; SNPs: single-nucleotide polymorphisms

Supplementary Table 3a

Strength and Difficulties Questionnaire (SDQ) scores by sex in the SESBiC cohort

SDQ	Sex	N	Mean	SD	<i>p</i> [†]
TOTAL	Male	455	11	6	0.044
	Female	454	10.2	5.5	
Conduct	Male	455	1.7	1.5	0.002
	Female	454	1.4	1.2	
Emotional	Male	455	1.8	1.7	<0.001
	Female	454	2.6	1.9	
Hyperactivity / inattention	Male	455	3.8	2.1	<0.001
	Female	454	3.1	1.9	
Peer problems	Male	455	1.8	1.5	0.101
	Female	454	1.6	1.4	
Pro-social behaviour	Male	455	7.7	1.7	<0.001
	Female	454	8.6	1.4	

TOTAL is the sum of the subscales: Conduct, Emotional, Hyperactivity / inattention and Peer problems

[†]: T-test

Supplementary Table 3b

Trauma Symptom Checklist for Children (TSCC) scores by sex in the USSS cohort

	Sex	N	Mean	SD	<i>p</i> [†]
TOTAL	Male	216	25,6	21,92	<0.001
	Female	182	38,7	23,38	
ANX	Male	216	3,0	3,66	<0.001
	Female	182	6,6	4,42	
DEP	Male	216	3,1	3,92	<0.001
	Female	182	6,6	4,92	
ANG	Male	216	4,6	4,90	0.044
	Female	182	5,6	4,52	
PTS	Male	216	4,9	4,98	<0.001
	Female	182	9,0	5,70	
DIS	Male	216	4,8	4,64	<0.001
	Female	182	7,5	4,87	
SC	Male	216	6,2	5,07	0.048
	Female	182	5,2	4,30	

TOTAL is the sum of the subscales: ANX: anxiety; DEP: depression; ANG: anger; PTS: posttraumatic stress; DIS: dissociation; SC: sexual concerns

[†]T-test

Supplementary Table 4

Correlation between total and subscales scores for SDQ and TSCC in adolescents of the SESBiC and USSS cohorts, respectively

SDQ		conduct	emotional	hyperactivity / inattention	peer problems	pro-social behaviour	
	N	909	909	909	909	909	
TOTAL SCORE	<i>r</i>	0.673**	0.595**	0.912**	0.333**	-0.363**	
TSCC		ANX	DEP	ANG	PTS	DIS	SC
	N	398	398	398	398	398	398
TOTAL SCORE	<i>r</i>	0.850**	0.873**	0.835**	0.893**	0.900**	0.687**

r: Pearson correlation coefficient

Supplementary table 5

Impact of *FKBP5* genotype on psychiatric symptoms in adolescents by TSCC and SDQ and total score and sub-scales in the SESBiC and USSS cohorts, respectively

Cohort	<i>FKBP5</i> genotypes	N	Psychiatric symptoms N (SD)	F	p
SESBiC SDQ Total	rs3800373			0.041	0.96
	AA	517	10.6 (5.96)		
	AC	329	10.5 (5.47)		
	CC	56	10.5 (5.67)		
	rs1360780			0.014	0.986
	CC	483	10.6 (5.91)		
	CT	342	10.6 (5.66)		
	TT	63	10.7 (5.68)		
SDQ Conduct	rs3800373			0.028	0.972
	AA	517	1.5 (1.39)		
	AC	329	1.5 (1.33)		
	CC	56	1.5 (1.28)		
	rs1360780			0.081	0.922
	CC	483	1.5 (1.38)		
	CT	342	1.6 (1.36)		
	TT	63	1.5 (1.29)		
SDQ Emotional	rs3800373			1.986	0.138
	AA	517	2.3 (1.88)		
	AC	329	2.1 (1.78)		
	CC	56	1.8 (1.61)		
	rs1360780			1.339	0.263
	CC	483	2.3 (1.84)		
	CT	342	2.2 (1.84)		
	TT	63	1.9 (1.66)		
SDQ Hyperactivity//inattention	rs3800373			0.196	0.822
	AA	517	3.4 (2.10)		
	AC	329	3.4 (1.98)		
	CC	56	3.6 (2.21)		
	rs1360780			0.358	0.699
	CC	483	3.4 (2.09)		
	CT	342	3.4 (2.02)		
	TT	63	3.7 (2.18)		
SDQ Peer problems	rs3800373			0.24	0.787
	AA	517	1.7 (1.48)		
	AC	329	1.8 (1.42)		
	CC	56	1.6 (1.51)		
	rs1360780			0.451	0.637
	CC	483	1.7 (1.46)		
	CT	342	1.8 (1.43)		
	TT	63	1.7 (1.61)		
SDQ Prosocial	rs3800373			0.913	0.402
	AA	517	8.2 (1.63)		
	AC	329	8.1 (1.65)		
	CC	56	8.0 (1.65)		
	rs1360780			1.169	0.311
	CC	483	8.2 (1.58)		
	CT	342	8.1 (1.71)		
	TT	63	8.0 (1.60)		
USSS TSCC Total	rs3800373			3.331	0.037
	AA	213	31.1 (21.70)		
	AC	156	30.4 (25.54)		
	CC	25	43.3 (24.38)		
	rs1360780			2.313	0.1
	CC	194	30.4 (21.90)		
	CT	166	31.4 (25.36)		
	TT	34	39.8 (22.94)		
TSCC Anxiety	rs3800373			0.815	0.443
	AA	213	4.5 (4.25)		
	AC	156	4.6 (4.65)		

	CC	25	5.7 (4.14)		
	rs1360780			0.939	0.392
	CC	194	4.4 (4.32)		
	CT	166	4.7 (4.54)		
	TT	34	5.5 (4.21)		
TSCC Depression	rs3800373			1.453	0.235
	AA	213	4.6 (4.60)		
	AC	156	4.6 (4.95)		
	CC	25	6.2 (4.72)		
	rs1360780			0.812	0.445
	CC	194	4.4 (4.67)		
	CT	166	4.7 (4.84)		
	TT	34	5.6 (4.78)		
TSCC Anger	rs3800373			3.761	0.024
	AA	213	5.1 (4.70)		
	AC	156	4.7 (4.63)		
	CC	25	7.5 (5.55)		
	rs1360780			1.909	0.15
	CC	194	5.0 (4.56)		
	CT	166	4.9 (4.90)		
	TT	34	6.6 (5.11)		
TSCC Post-trauma	rs3800373			4.052	0.018
	AA	213	6.7 (5.30)		
	AC	156	6.3 (5.96)		
	CC	25	9.8 (6.71)		
	rs1360780			2.349	0.097
	CC	194	6.5 (5.30)		
	CT	166	6.6 (5.98)		
	TT	34	8.8 (6.27)		
TSCC Dissociation	rs3800373			1.441	0.238
	AA	213	6.1 (4.32)		
	AC	156	5.8 (5.53)		
	CC	25	7.6 (5.86)		
	rs1360780			0.568	0.567
	CC	194	5.9 (4.39)		
	CT	166	6.1 (5.45)		
	TT	34	6.9 (5.32)		
TSCC Sexual concerns	rs3800373			3.229	0.041
	AA	213	5.5 (4.40)		
	AC	156	5.8 (5.03)		
	CC	25	8.0 (5.61)		
	rs1360780			3.866	0.022
	CC	194	5.4 (4.53)		
	CT	166	5.7 (4.87)		
	TT	34	7.9 (5.16)		

Supplementary tables 6a and 6b

Interaction of *FKBP5* genotype and level of adverse life events predict psychiatric symptoms in adolescents by by TSCC and SDQ sub-scales in the SESBiC and USSS cohorts, respectively

a) SESBiC cohort

SDQ	FKBP5 Genotypes	Participants N	ALE <90th percentile mean (SD)	Participants N	ALE ≥90th percentile mean (SD)	$p^{\$}$	$R^2^{\#}$
Conduct	rs3800373					E =0.173; G =0.663; GXE =0.519	0.033
	AA	449	1.5 (1.40)	68	1.7 (1.27)		
	AC	272	1.4 (1.29)	57	2.0 (1.41)	E = 0.041[#] ; G =0.376 [#] ; GXE =0.235 [#]	0.035 ⁵
	CC	52	1.4 (1.28)	4	2.0 (1.41)		
	rs1360780					E =0.091; G =0.541; GXE =0.514	0.033
	CC	422	1.5 (1.39)	61	1.7 (1.32)		
Emotional	CT	281	1.5 (1.34)	61	2.0 (1.38)	E =0.073 [#] ; G =0.315 [#] ; GXE =0.279 [#]	0.033 [#]
	TT	58	1.5 (1.29)	5	2.2 (1.30)		
	rs3800373					E =0.198; G =0.666; GXE =0.856	0.057
	AA	449	2.2 (1.84)	68	2.6 (2.06)		
	AC	272	2.0 (1.76)	57	2.5 (1.84)	E = 0.009[#] ; G =0.444 [#] ; GXE =0.562 ^{#2}	0.057 [#]
	CC	52	1.8 (1.59)	4	2.3 (2.06)		
Hyperactivity /inattention	rs1360780					E =0.113; G =0.931; GXE =0.761	0.057
	CC	422	2.2 (1.81)	61	2.5 (2.09)		
	CT	281	2.1 (1.84)	61	2.5 (1.84)	E = 0.016[#] ; G =0.928 [#] ; GXE =0.427 [#]	0.056 [#]
	TT	58	1.8 (1.64)	5	2.6 (1.95)		
	rs3800373					E = 0.010 ; G =0.083; GXE = 0.019	0.046
	AA	449	3.4 (2.12)	68	3.6 (1.94)		
Peer problems	AC	272	3.2 (1.95)	57	4.5 (1.80)	E = 0.003[#] ; G = 0.040[#] ; GXE = 0.008[#]	0.045
	CC	52	3.5 (2.20)	4	5.03(1.71)		
	rs1360780					E = 0.016 ; G =0.277; GXE =0.229	0.040
	CC	422	3.4 (2.11)	61	3.4 (2.114)		
	CT	281	3.3 (2.01)	61	3.3 (2.01)	E = 0.011[#] ; G =0.268 [#] ; GXE =0.145 [#]	0.039 [#]
	TT	58	3.5 (2.19)	5	5.03(1.58)		
Prosocial behaviour	rs3800373					E =0.404; G =0.808; GXE =0.600	0.019
	AA	449	1.7 (1.44)	68	1.6 (1.43)		
	AC	272	1.7 (1.39)	57	1.7(1.40)	E = 0.037[#] ; G =0.566 [#] ; GXE =0.339 [#]	0.018 [#]
	CC	52	1.6 (1.52)	4	1.7 (1.58)		
	rs1360780					E =0.089; G =0.820; ; GXE =0.665	0.018
	CC	422	1.6 (1.43)	61	2.1 (1.62)		
Prosocial behaviour	CT	281	1.7 (1.41)	61	2.0 (1.56)	E = 0.040[#] ; G =0.815 [#] ; GXE =0.642 [#]	0.018 [#]
	TT	58	1.7 (1.60)	5	2.4 (1.95)		
	rs3800373					E = 0.039 ; G =0.081; GXE =0.194	0.091
	AA	449	8.2 (1.63)	68	8.0 (1.62)		
	AC	272	8.2 (1.57)	57	7.5 (1.89)	E =0.073 [#] ; G =0.053 [#] ; GXE =0.138 [#]	0.090 [#]
	CC	52	8.1 (1.61)	4	6.8 (0.96)		
Prosocial behaviour	rs1360780					E =0.076; G =0.066; GXE =0.173	0.087
	CC	422	8.2 (1.58)	61	8.1 (1.61)		
	CT	281	8.2 (1.66)	61	7.5 (1.85)	E =0.125 [#] ; G = 0.027[#] ; GXE =0.080 [#]	0.086 [#]
	TT	58	8.2 (1.61)	5	7.2 (1.30)		

[§] GLM adjusted for: sex, living with both parents v.s. separated parents; both parents working v.s. ≥one parent not working; both parents born in Sweden v.s. ≥one parent born outside Sweden.

[#] model with 2 levels of genotypes, homozygous for the most common allele vs. carriers of the minor allele

E: environment main effect; GXE: gene-by-environment interaction effect; SDQ: Strength and Difficulties Questionnaire; ALE: adverse life events

b) USSS cohort

TSCC	FKBP5 Genotypes	Participants N	ALE <90th percentile mean (SD)	Participants N	ALE ≥90th percentile mean (SD)	$p^{\S}$	$R^{2\#}$
Anxiety	rs3800373					E <0.001; G =0.034; GXE =0.025	0.283
	AA	187	4.2 (3.92)	26	7.2 (5.50)		
	AC	136	3.9 (3.70)	20	9.8 (6.82)	E =<0.001 [#] ; G =0.011 [#] ; GXE =0.008 [#]	0.270 [#]
	CC	18	4.7 (3.60)	7	8.4 (4.47)		
	rs1360780					E <0.001; G =0.047; GXE =0.075	0.278
	CC	169	4.0 (3.95)	25	7.3 (5.62)		
	CT	146	4.0 (3.71)	20	9.5 (6.82)	E =<0.001 [#] ; G =0.013 [#] ; GXE =0.023 [#]	0.265 [#]
Depression	rs3800373					E <0.001; G =0.001; GXE <0.001	0.280
	AA	187	4.3 (4.35)	26	6.5 (5.87)		
	AC	136	3.7 (3.77)	20	10.5 (7.58)	E <0.001 [#] ; G <0.001 [#] ; GXE <0.001 [#]	0.266 [#]
	CC	18	4.4 (3.99)	7	10.9 (3.08)		
	rs1360780					E <0.001; G =0.003; GXE <0.001	0.275
	CC	169	4.1 (4.38)	25	6.6 (5.97)		
	CT	146	4.0 (3.78)	20	10.1 (7.71)	E <0.001 [#] ; G <0.001 [#] ; GXE <0.001 [#]	0.260 [#]
Anger	rs3800373					E <0.001; G =0.039; GXE =0.038	0.185
	AA	187	4.7 (4.43)	26	8.3 (5.44)		
	AC	136	3.9 (3.79)	20	10.1 (6.16)	E <0.001 [#] ; G =0.078 [#] ; GXE =0.008 [#]	0.162
	CC	18	5.4 (4.02)	7	12.9 (5.52)		
	rs1360780					E <0.001; G =0.191; GXE =0.258	0.169
	CC	169	4.4 (4.19)	25	8.5 (5.39)		
	CT	146	4.3 (4.28)	20	9.9 (6.32)	E <0.001 [#] ; G =0.102 [#] ; GXE =0.084 [#]	0.153 [#]
Post-trauma	rs3800373					E <0.001; G =0.007; GXE =0.011	0.319
	AA	187	6.0 (4.94)	26	11.5 (5.39)		
	AC	136	5.3 (5.16)	20	12.8 (7.09)	E <0.001 [#] ; G =0.039 [#] ; GXE =0.010 [#]	0.295 [#]
	CC	18	6.7 (4.86)	7	17.6 (3.78)		
	rs1360780					E <0.001; G =0.045; GXE =0.072	0.311
	CC	169	5.8 (4.85)	25	11.6 (5.48)		
	CT	146	5.8 (5.31)	20	12.8 (7.06)	E <0.001 [#] ; G =0.032 [#] ; GXE =0.053 [#]	0.294
Dissociation	rs3800373					E <0.001; G =0.020; GXE =0.001	0.266
	AA	187	5.6 (3.96)	26	9.4 (5.34)		
	AC	136	4.8 (4.31)	20	12.6 (7.87)	E <0.001 [#] ; G =0.005 [#] ; GXE <0.001 [#]	0.251 [#]
	CC	18	5.1 (4.30)	7	14.0 (4.32)		
	rs1360780					E <0.001; G =0.023; GXE =0.008	0.259
	CC	169	5.4 (3.96)	25	9.4 (5.44)		
	CT	146	5.2 (4.37)	20	12.6 (7.87)	E <0.001 [#] ; G =0.005 [#] ; GXE =0.003 [#]	0.244 [#]
Sexual concerns	rs3800373					E <0.001; G =0.060; GXE =0.376	0.093
	AA	187	5.2 (4.19)	26	7.4 (5.43)		
	AC	136	5.3 (4.26)	20	9.2 (7.95)	E <0.001 [#] ; G =0.068 [#] ; GXE =0.123 [#]	0.069 [#]
	CC	18	7.1 (5.62)	7	10.3 (5.25)		
	rs1360780					E <0.001; G =0.087; GXE =0.266	0.094
	CC	169	5.1 (4.30)	25	7.4 (5.54)		
	CT	146	5.2 (4.09)	20	9.3 (7.91)	E <0.001 [#] ; G =0.072 [#] ; GXE =0.219 [#]	0.065 [#]
Sexual concerns	TT	25	7.5 (5.15)	9	8.89 (5.35)		

[§]GLM adjusted for: sex, living with both parents v.s. separated parents; both parents working v.s. ≥one parent not working; both parents born in Sweden v.s. ≥one parent born outside Sweden.

[#] model with 3 levels of SNPs and with 2 levels

E: environment main effect; GXE: gene-by-environment interaction effect; TSCC: Trauma Symptom Checklist for Children; ALE: adverse life events

Supplementary Table 7

FKBP5 rs3800373 and rs1360780 genotype-dependent correlation effects between adverse life events and psychiatric symptoms assessed with SDQ and TSCC in adolescents of the SESBiC and USSS cohorts, respectively

	rs3800373			rs1360780		
	AA	AC	CC	CC	CT	TT
SESBiC						
N	517	329	56	483	342	63
<i>r</i>	0.125**	0.314***	0.052	0.128**	0.273***	0.083
<i>Z value for difference</i>	AA/AC=2.82 ^b	AC/CC=1.86	AA/CC=0.51	CC/CT=2.14 ^a	CT/TT=1.42	CC/TT=0.33
USSS						
N	213	156	25	194	166	34
<i>r</i>	0.392***	0.542***	0.888***	0.391***	0.593***	0.841***
<i>Z value for difference</i>	AA/AC=1.81	AC/CC=3.53 ^b	AA/CC=4.45 ^b	CC/CT=1.78	CT/TT=3.17 ^b	CC/TT=4.19 ^b

r: Pearson correlation coefficient, *= $p < 0.05$, **= $p < 0.01$, ***= $p < 0.001$

^a $p < 0.05$, ^b $p < 0.01$

Supplementary Table 8

Diplotypes of *FKBP5* SNPs

Cohort	rs3800373- rs1360780 diplotypes	Diplotype frequencies	
		Males N (%)	Females N (%)
SESBIC	Homozygous for major allele at both loci	229 (48.4)	244 (51.6)
	All other combinations	183 (51.7)	171 (48.3)
	Homozygous for minor allele at both loci	30 (55.6)	24 (44.4)
USSS	Homozygous for major allele at both loci	100 (52.4)	91 (47.6)
	All other combinations	102 (58.6)	72 (41.4)
	Homozygous for minor allele at both loci	12 (48.0)	13 (52.0)

Supplementary Table 9

Interaction effect of *FKBP5* diplotypes and adverse life events associated with psychiatric symptoms (assessed with SDQ and TSCC) in adolescents of the SESBiC and USSS cohorts, respectively

Total score	<i>FKBP5</i> rs3800373 - rs1360780 diplotypes	Participants	ALE <90th percentile	Participants	ALE ≥90th percentile	p [§]	R ²
		N		N			
SDQ							
			mean SDQ score (SD)		mean SDQ score (SD)		
	Homozygous for major allele at both loci	413	10.4 (5.94)	60	11.6 (5.75)	E < 0.017 ; G =0.478 GxE = 0.275	0.014
	All other combinations	291	10.1 (5.61)	63	12.9 (5.21)		
	Homozygous for minor allele at both loci	50	10.2 (5.63)	4	14.8 (6.02)		
TSCC							
			mean TSCC score (SD)		mean TSCC score (SD)		
	Homozygous for major allele at both loci	166	27.9 (19.66)	25	48.9 (27.13)	E < 0.001 ; G = 0.007 GxE = 0.021	0.255
	All other combinations	153	27.0 (20.03)	21	60.3 (37.31)		
	Homozygous for minor allele at both loci	18	32.5 (19.75)	7	71.0 (12.78)		

§ GLM adjusted for: sex, living with both parents v.s. separated parents; both parents working v.s. ≥one parent not working; both parents born in Sweden v.s. ≥one parent born outside Sweden. ALE: adverse life events

Supplementary Figure

Relation of adverse life events (<90th percentile vs. ≥90th percentile) to estimated marginal means of psychiatric symptoms in adolescents (SDQ in the SESBiC cohort and TSCC in the USSS cohort) by *FKBP5* diplototypes

SDQ: Strength and Difficulties Questionnaire; TSCC: Trauma Symptom Checklist for Children

GxE: gene-by-environment interaction effect based on GLM model adjusted for: sex, living with both parents v.s. separated parents, both parents working v.s. than one parent not working, both parents born in Sweden v.s. more than one parent born outside Sweden

Homozygous for major allele at both loci

All other combinations

Homozygous for minor allele at both loci