Prenatal and childhood adverse life events, inflammation and depressive symptoms across adolescence

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Abstract

Background: No study has investigated the role of inflammation in explaining the association between early exposures to adverse life events and depressive symptoms in adolescence. **Method:** Using data from the Avon Longitudinal Study of Parents and Children, we tested if inflammatory markers [serum C-reactive protein (CRP) and interleukin 6 (IL-6)] at age 9 years mediated the association between adverse life events, measured separately for the prenatal (since the beginning of pregnancy) and the childhood (ages 0-9 years) periods, and the development of depressive symptoms at ages 10-17 years. Data (n=4,263) were analysed using mediation analysis in a latent growth curve modelling framework. **Results:** Depressive symptoms at the beginning of adolescence (age 10) were associated with the number of prenatal events, the number of events around birth and the increase in events over time in childhood (ages 0-9), even after adjustment for confounders. IL-6 partially mediated the association between increasing exposure to events over time in childhood and depressive symptoms at the beginning of adolescence. IL-6 did not mediate any other association between events and symptoms. There was no evidence for mediation by CRP, which was generally unrelated to events. **Limitations:** The small size of the mediation effect and the robust direct effects of events prenatally and around birth suggest there are multiple routes from early stressors to adolescent depression. **Conclusions:** In the general adolescent population, increasing exposure to psychosocial stressors over time during childhood is associated with the early onset of depressive symptoms, partly via increasing levels of plasma IL-6.

Keywords: Adolescence, adverse life events, ALSPAC, depression, IL-6, inflammation

Introduction

The early experience of adverse or stressful life events has been widely associated with adverse health outcomes[1, 2] such as the development of mental illness including major depressive disorder or psychosis spectrum disorders in adolescence and young adulthood.[3] In fact, stressful events experienced as early as during pregnancy have been found to increase the risk of mental health problems in the offspring[4]. Exposure to adverse life events, either prenatally or during childhood, can also increase plasma levels of inflammatory markers[4-10], in turn associated both with the onset of depressive symptoms and with treatment response.[11-14] For example, a recent meta-analysis found that elevated levels of inflammation are partially responsible for treatment resistance in individuals with depression.[15]

Thus, the link from early stressful life events to depression via inflammation is plausible but, to our knowledge, still unexplored in adolescence. One study tested the role of inflammation in the association between stressful life events and internalising symptoms, but did not investigate links beyond childhood, did not consider the role of prenatal life events and did not explore depressive symptomatology specifically[49]. No study has yet investigated if prenatal and childhood stressful life events are related to the development of depression across adolescence via increasing levels of inflammatory markers. This is a significant gap because knowledge about the roles of stressors and inflammation in adult depression cannot be assumed to directly apply to paediatric depression. First, due to shorter illness duration and less medical comorbidity, the associations between inflammation and depression in the youth population may be different from what has been documented in adults. Young people are typically free of illness burden or extensive histories of antipsychotic usage, linked to inflammation. Second, adult depression can have an age at onset in adulthood or childhood/adolescence, and there is some evidence that juvenile-onset (i.e., child- or adolescent-onset) and adult-onset depression do not have the same risk correlates and

precursors. For example, although genetic susceptibility to major depressive disorder does not differ between adult- and earlier-onset major depressive disorder [17], nor does early exposure to risk factors such as poverty[18, 19] and parental psychopathology, [20-22] some adverse childhood experiences such as maltreatment, including sexual/physical abuse and neglect, can accelerate onset into adolescence. [16]

Therefore, adding knowledge about the association between adverse life events and experiences, inflammatory marker levels and the onset and course of depression in adolescence would help build targeted interventions. Depression is a common and serious disorder of adolescence.[23] Lifetime prevalence of major depressive disorder increases dramatically from 1% of the population under age 12 to ~17%–25% by the end of adolescence[24], with the greatest surge in newly emergent cases in adolescence occurring after age 15 years.[25] It would also prevent future problems in other domains, given that juvenile-onset depression is associated with alterations in neural development[26], a host of later negative outcomes and pervasive dysfunction throughout life[27].

We aimed to add this knowledge by using longitudinal data from a large UK birth cohort study, the Avon Longitudinal Study of Parents and Children (ALSPAC). We investigated if higher levels of inflammatory markers in childhood (age 9 years) mediate the association between both prenatal and childhood (0-9 years) stressful life events and depressive symptoms across adolescence (ages 10-17 years). As is standard in epidemiological studies, we disentangled in our mediation analysis the total effect of the exposure (life events) on the outcome (depressive symptoms), the indirect effect of the exposure that acts through the mediator (inflammatory marker), and the direct effect of the exposure that is unexplained by that mediator.

Study design and participants

ALSPAC is a prospective cohort study designed to assess environmental factors during and after pregnancy that might affect the development, health, or wellbeing of the child [28] [http://www.bristol.ac.uk/alspac/researchers/our-data/; note that the study website contains details of all the data that is available through a fully searchable data dictionary and variable search tool]. It enrolled 14,541 pregnant women in the Bristol area of the UK between April 1991 and December 1992. From the first trimester of pregnancy parents completed postal questionnaires about themselves and the study child's health and development. Children were invited to attend annual assessment clinics, including face-to-face interviews and psychological and physical tests from age 7 years onwards. Additional children were recruited using the original enrolment definition from the participating children's age 7 years onwards, increasing the number to 15,445 to date.[29] Ethical approval for the study was obtained from the ALSPAC Ethics and Law Committee and the Local Research Ethics Committees. All participants provided written informed consent and there was no financial compensation (more details at www.alspac.bris.ac.uk). Our analytic sample included 4,263 children (singletons and first-born twins or triplets) who had data on inflammation at age 9 years [measured in ALSPAC with serum C-reactive protein (CRP) and interleukin 6 (IL-6)], did not report an infection at the time of blood collection or during the preceding week, and, to elucidate the link between inflammatory marker levels and risk of depression without the confounding burden of psychiatric comorbidities, had not received a diagnosis of any emotional disorder since age 7 years, when the Development and Well-Being Assessment (DAWBA) was first administered in ALSPAC.

Measures

Inflammation, age 9 years

In ALSPAC, inflammation in childhood was measured with CRP and IL-6, two well-established inflammatory markers[30], at age 9 years during a clinic visit. Consent for

biological samples has been collected in accordance with the Human Tissue Act (2004). Blood samples were collected from nonfasting participants and were immediately spun and frozen at −80°C. Inflammatory markers were assayed in 2008 after a median of 7.5 years in storage with no previous freeze-thaw cycles during this period. IL-6 (pg/mL) was measured by enzyme-linked immunosorbent assay (R&D Systems) and high-sensitivity CRP (mg/L) was measured by automated particle-enhanced immunoturbidimetric assay (Roche). All inter-assay coefficients of variation were less than 5%. In the full ALSPAC sample, IL-6 values ranged from 0.007 to 20.051 pg/mL (n = 5,072) while CRP values ranged from 0.01 to 67.44 mg/L (n = 5,082; for 60 of these children CRP values were over 10 mg/L). In order to increase the replicability and interpretability of the results, both CRP and IL-6 values were converted into tertiles in our analysis.

Life events

Pre- and post-natal stressful life events were measured in ALSPAC using, respectively, a 41- and a 43-event checklist (Supplementary Table S1), completed by the mother[31-34]. In our analysis we derived a total prenatal life events score using information from two time-points: at 18 weeks gestation (covering events since the beginning of the pregnancy) and at 8 weeks postpartum (covering events since 18 weeks gestation). At each time-point, a score of 0 was assigned if the event did not occur and a score of 1 if it did. We then calculated a total score capturing whether each of the 41 events ever occurred at any of the two time-points. Postnatally, stressful life events were measured at several time-points, including 21 months (covering events since the child was 8 months), 33 months (covering events since the child was 18 months), 47 months (covering events since the child was 33 months), 61 months (covering events since the child was 47 months), 73 months (covering events since the child was 61 months) and 110 months (covering events since the child was 73 months). At each time-point, a score of 0 was assigned if the event did not occur and a score of 1 if it did.

Depressive symptoms

Depressive symptoms were assessed using the widely-used short (13-item) version of the Mood and Feelings Questionnaire (MFQ)[35]. This self-administered questionnaire (at ages 10.5, 12.5, 13.5, 16.5 and 17.5 years in ALSPAC) indexes symptoms experienced in the previous 2 weeks. Each item is scored 0 (not true), 1 (sometimes true) or 2 (true), giving a theoretical range of 0-26 for the total MFQ.

Confounders

We adjusted for several covariates known to be associated with children's levels of inflammatory markers, depressive symptoms and exposure to life events. These included gender, ethnicity (white, non-white), parental socio-economic status, which we approximated by maternal education (university degree or not), and obesity status (body mass index (BMI) above the 95th percentile for children of the same age).[36] BMI (weight (kg)/height (m)^2) was measured during the clinic visit at age 9 years.

Statistical analysis

All analyses were performed in STATA 15.0 (Stata Corporation, College Station, TX, 1997).[37] First, we fitted latent growth curve models to estimate the longitudinal trajectories of childhood events from ages 8 to 110 months and depressive symptoms from ages 10.5 to 17.5 years. The individual predicted values of the intercept (set at baseline) and the slope (rate of annual change) were then used in the mediation model. Using the predict command in STATA we generated predictions for the out-of-sample cases, i.e., the cases that were not used in the original estimation. This command uses the maximum likelihood with missing values (MLMV) estimation for those who had data on at least one time-point and single imputation for those missing information on all time-points. In this way, both the intercepts of childhood events and depressive symptoms (set at ages 8 months and 10.5 years,

respectively, as explained) and their slopes were estimated for our whole analytic sample (n=4,263). The individual predicted values of the intercepts and the slopes were then saved and used in regression models testing for mediation, separately by CRP and IL-6. Missingness among the confounders ranged from 1.1% (BMI) to 5.3% (maternal education). Confounders were then imputed (20 imputed datasets) using multiple imputation by chained equations (MICE)[38]. We assumed that missingness was dependent on observed data (missing at random). To predict missing data, we used all variables selected for analysis models. We imputed up to the analytic sample. We fitted models before and after adjustment for confounders, both in the imputed and the complete cases samples.

Results

Descriptive analysis

Table 1 shows the descriptive characteristics of our sample, including means and proportions for the exposures, outcomes and covariates. As can be seen, there seemed to be an increase in the number of depressive symptoms by age, especially after mid-adolescence, but a relative stability in the number of events experienced. Correlations between the main variables and between all exposures and outcomes were low to moderate, as expected (Supplementary Table S2).

Mediation models

The mediation model specified to predict the slope of depressive symptoms showed no association of symptoms with either life events or inflammatory markers (results available on request). However, the model specified to predict the intercept (set at age 10.5 years) of depressive symptoms showed that IL-6 at age 9 years mediated part of the effect of the slope of adverse life events (i.e., the growth of the number of events experienced after birth until age 9 years). The effect was robust to the adjustment for covariates (indirect effect: β=0·01,

p<0·05, 95% CI=0·000-0·004; total effect: β=0·04, p<0·01, 95% CI=0·013-0·073; direct effect: β=0·04, p<0·01, 95% CI=0·010-0·070)[a]. Table 2 presents the results (direct effects) of our mediation model for IL-6 before and after adjustment, and in both the imputed and the complete cases sample. The number of prenatal events had a significant effect on the intercept of the trajectory of depressive symptoms, as did the intercept of the trajectory of postnatal events, but IL-6 did not mediate either effect, as discussed. CRP did not mediate any of the associations between life events and the intercept of depressive symptoms, and was in fact unrelated to adverse life events (Supplementary Table S3).

Discussion

Our study showed that, even in the general population, elevated IL-6 partially explained the association between increasing exposure to stressors over time in childhood and depressive symptoms at the beginning of adolescence. However, it found no evidence that the association between prenatal and very early (around birth) exposures to stressors and depressive symptoms was mediated by either IL-6 or CRP. Our results are therefore consistent with previous evidence about the link between exposure to stressors and later mental health problems but they also add to this evidence in three important ways: (i) they show an association between inflammation (IL-6) and depression in adolescence (ii) they point to a biological pathway (inflammation) via which increases in the number of stressors experienced over time in childhood may lead to depressive symptoms in adolescence, and (iii) they suggest that, rather than explaining the effect of prenatal maternal stress on risk of depression in adolescence, elevated IL-6 in children may be best be seen as an immediate response to a relative increase in the number of stressors encountered in childhood, in turn

[a] Using unstandardised regression coefficients, direct effect: b=0.43, SE=0.16; indirect effect: b=0.03, SE=0.01; total effect: b=0.46, SE=0.16.

associated with depressive symptoms in early adolescence. Thus, our findings about the association between recent exposures to psychosocial stressors and depressive symptoms in adolescence via inflammation, as well as about the direct link between prenatal exposures to psychosocial stressors and depressive symptoms in adolescence, can help to build targeted interventions to reduce the risk of major depressive disorder. [41, 42]

Our study has many strengths. To the best of our knowledge, it is the first general-population study to test the mediating role of inflammatory markers (IL-6 and CRP) in childhood in the association between exposure to stressors, both prenatally and postnatally across the childhood years, and the subsequent development of depressive symptoms across adolescence. Another significant strength is that it took a broad view of stressors, by considering both conditions or experiences marked by misfortune (either relative or absolute) [39] and events that are arguably developmental or universal challenges that most families or children encounter. Importantly, it also covered exposures to such psychosocial stressors comprehensively, including both events experienced by the expectant mother and her family throughout pregnancy and events experienced by the child and their family from birth until the end of the first decade of life, when inflammation was measured. Thus, it extends the existing evidence that has established associations in adolescence between inflammation, depression and recent exposures to stressors.[40]

Nonetheless, there are several important study limitations. First, inflammation was only assessed once. Second, given the observational design, causality cannot be inferred. Although our findings are in line with our expectations, we cannot discount residual confounding and reverse causality. It is also important to consider genetic mechanisms, which offer an alternative, noncausal interpretation of the associations we found between exposure to psychosocial stressors, elevated IL-6 and increased depressive symptoms in adolescents. For example, genetic pathways related to the immune system predict risk for several psychiatric

diagnoses including depression.[43] In turn, early expressions of liability to these conditions might increase the risk of exposure to psychosocial stressors, in line with a stress generation explanation[44] according to which depression or depressogenic vulnerabilities can increase susceptibility to stressful events that are at least in part influenced by the individual. Third, the life events checklist did not cover specifically events experienced by the child, so our analyses may underestimate the impact of personal events on both inflammatory marker levels and depressive symptoms. Fourth, the mediation effect found was small, which underlines the direct impact of adverse life events on depressive symptoms and suggests that there are multiple routes from exposure to psychosocial stressors to depression in adolescence. It also underscores the importance of taking development into account when examining links between early exposure to stressors or adversity and circulating markers of inflammation. For example, a recent meta-analysis[45] showed that the association between early life adversity and both CRP and IL-6 appears to be negligible across youth samples ($z = 0.06$ for both; confidence intervals, respectively, were [−0.01, 0.14] and [−0.17, 0.30]). By contrast, a meta-analysis investigating the association between childhood trauma exposure and circulating inflammatory markers in adulthood, published only 3 years ago in 2016[5], showed small, yet significant, effect sizes overall; $z = 0.10$ for CRP, $z = 0.08$ for IL-6, and $z = 0.23$ for TNF-α (measured in too few studies with paediatric populations to make definitive conclusions and not available in our sample). Development across childhood and adolescence must also be considered. As Kuhlman et al. note[45], it is possible that in children upregulations in the HPA-axis may be masking the pro-inflammatory phenotype. The substantial alteration of the HPA-axis during pubertal development[46], on the other hand, coupled with the emergence at around the same time of risky behaviours that increase inflammation such as smoking, drinking and unhealthy eating, suggests that differences in patterns between child and adolescent samples are likely. Future studies measuring

glucocorticoid concentrations in children and adolescents in addition to inflammatory markers could help test this. Finally, we considered, for our analysis, stressful life events as strictly environmental exposures. However, the now established genetic overlap between stressful life events and major depressive disorder suggests that their relationship may not be directionally causal but a consequence of common genetic effects that influence both.[47] Nonetheless, most of the stressors in the checklist we used are likely independent events (i.e., beyond the control of the child); the genetic overlap between major depressive disorder and independent life events is lower than for dependent life events [47, 48] (such as relationship problems or job loss, that may be, in part, the result of a person's own behaviour).

Conclusion

These limitations notwithstanding, our findings suggest that increasing exposure to psychosocial stressors over time during the first decade of life is related to depressive symptoms in early adolescence, partly via increased levels of inflammatory markers. Importantly, the same or very similar psychosocial stressors experienced prenatally and around birth are also related to depressive symptoms in early adolescence, but not via elevated inflammation, suggesting qualitatively different pathways to adolescent depression by early and recent psychosocial stressors.

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References

1. Croft J, Heron J, Teufel C, Cannon M, Wolke D, Thompson A, et al. Association of trauma type, age of exposure, and frequency in childhood and adolescence with psychotic experiences in early adulthood *Jama Psychiat*. 2019; **76**:79-80.

2. Danese A, Moffitt TE, Harrington H, Milne BJ, Polanczyk G, Pariante CM, et al. Adverse childhood experiences and adult risk factors for age-related disease: depression, inflammation, and clustering of metabolic risk markers *Archives of pediatrics & adolescent medicine*. 2009; **163**:1135-43.

3. Assari S, Lankarani MM. Association between stressful life events and depression; intersection of race and gender *Journal of racial and ethnic health disparities*. 2016; **3**:349-56.

4. Kingsbury M, Weeks M, MacKinnon N, Evans J, Mahedy L, Dykxhoorn J, et al. Stressful life events during pregnancy and offspring depression: evidence from a prospective cohort study *Journal of the American Academy of Child & Adolescent Psychiatry*. 2016; **55**:709-16. e2.

5. Baumeister D, Akhtar R, Ciufolini S, Pariante CM, Mondelli V. Childhood trauma and adulthood inflammation: a meta-analysis of peripheral C-reactive protein, interleukin-6 and tumour necrosis factor-α *Mol Psychiatr*. 2016; **21**:642.

6. Chen E, Hanson MD, Paterson LQ, Griffin MJ, Walker HA, Miller GE. Socioeconomic status and inflammatory processes in childhood asthma: the role of psychological stress *Journal of Allergy and Clinical Immunology*. 2006; **117**:1014-20.

7. Danese A, Caspi A, Williams B, Ambler A, Sugden K, Mika J, et al. Biological embedding of stress through inflammation processes in childhood *Mol Psychiatr*. 2011; **16**:244-6.

8. Dowd JB, Zajacova A, Aiello AE. Predictors of inflammation in US children aged 3–16 years *American journal of preventive medicine*. 2010; **39**:314-20.

9. Jokela M, Virtanen M, Batty GD, Kivimäki M. Inflammation and specific symptoms of depression *Jama Psychiat*. 2016; **73**:87-8.

10. Miller AH, Raison CL. The role of inflammation in depression: from evolutionary imperative to modern treatment target *Nature Reviews Immunology*. 2016; **16**:22.

11. Delany FM, Byrne ML, Whittle S, Simmons JG, Olsson C, Mundy LK, et al. Depression, immune function, and early adrenarche in children *Psychoneuroendocrino*. 2016; **63**:228-34.

12. Khandaker GM, Pearson RM, Zammit S, Lewis G, Jones PB. Association of Serum Interleukin 6 and C-Reactive Protein in Childhood With Depression and Psychosis in Young Adult Life A Population-Based Longitudinal Study *Jama Psychiat*. 2014; **71**:1121-8.

13. Khandaker GM, Stochl J, Zammit S, Goodyer I, Lewis G, Jones PB. Childhood inflammatory markers and intelligence as predictors of subsequent persistent depressive symptoms: a longitudinal cohort study *Psychological medicine*. 2018; **48**:1514-22.

14. Khandaker GM, Zammit S, Burgess S, Lewis G, Jones PB. Association between a functional interleukin 6 receptor genetic variant and risk of depression and psychosis in a population-based birth cohort *Brain, behavior, and immunity*. 2018; **69**:264-72.

15. Strawbridge R, Arnone D, Danese A, Papadopoulos A, Vives AH, Cleare A. Inflammation and clinical response to treatment in depression: a meta-analysis *European Neuropsychopharmacology*. 2015; **25**:1532-43.

16. Andersen SL, Teicher MH. Stress, sensitive periods and maturational events in adolescent depression *Trends in neurosciences*. 2008; **31**:183-91.

17. Halldorsdottir T, Piechaczek C, Soares de Matos AP, Czamara D, Pehl V, Wagenbuechler P, et al. Polygenic Risk: Predicting Depression Outcomes in Clinical and Epidemiological Cohorts of Youths *American Journal of Psychiatry*. 2019:appi. ajp. 2019.18091014.

18. Joinson C, Kounali D, Lewis G. Family socioeconomic position in early life and onset of depressive symptoms and depression: a prospective cohort study *Soc Psych Psych Epid*. 2017; **52**:95-103.

19. Shanahan L, Copeland WE, Costello EJ, Angold A. Child-, adolescent-and young adult-onset depressions: differential risk factors in development? *Psychological medicine*. 2011; **41**:2265-74.

20. Gilman SE, Kawachi I, Fitzmaurice GM, Buka SL. Socio-economic status, family disruption and residential stability in childhood: relation to onset, recurrence and remission of major depression *Psychological medicine*. 2003; **33**:1341-55.

21. Hill J, Pickles A, Rollinson L, Davies R, Byatt M. Juvenile-versus adult-onset depression: multiple differences imply different pathways *Psychological medicine*. 2004; **34**:1483-93.

22. Jaffee SR, Moffitt TE, Caspi A, Fombonne E, Poulton R, Martin J. Differences in early childhood risk factors for juvenile-onset and adult-onset depression *Archives of general psychiatry*. 2002; **59**:215-22.

23. Emslie GJ, Mayes TL, Ruberu M. Continuation and maintenance therapy of early-onset major depressive disorder *Pediatric Drugs*. 2005; **7**:203-17.

24. Kessler RC, Avenevoli S, Merikangas KR. Mood disorders in children and adolescents: an epidemiologic perspective *Biological psychiatry*. 2001; **49**:1002-14.

25. Hankin BL, Abramson LY, Moffitt TE, Silva PA, McGee R, Angell KE. Development of depression from preadolescence to young adulthood: emerging gender differences in a 10-year longitudinal study *Journal of abnormal psychology*. 1998; **107**:128.

26. Luby JL, Belden AC, Jackson JJ, Lessov-Schlaggar CN, Harms MP, Tillman R, et al. Early childhood depression and alterations in the trajectory of gray matter maturation in middle childhood and early adolescence *Jama Psychiat*. 2016; **73**:31-8.

27. McLeod G, Horwood L, Fergusson D. Adolescent depression, adult mental health and psychosocial outcomes at 30 and 35 years *Psychological medicine*. 2016; **46**:1401-12.

28. Boyd A, Golding J, Macleod J, Lawlor DA, Fraser A, Henderson J, et al. Cohort Profile: the 'children of the 90s'--the index offspring of the Avon Longitudinal Study of Parents and Children *Int J Epidemiol*. 2013; **42**:111-27.

29. Fraser A, Macdonald-Wallis C, Tilling K, Boyd A, Golding J, Smith GD, et al. Cohort Profile: The Avon Longitudinal Study of Parents and Children: ALSPAC mothers cohort *International Journal of Epidemiology*. 2013; **42**:97-110.

30. Del Giudice M, Gangestad SW. Rethinking IL-6 and CRP: Why they are more than inflammatory biomarkers, and why it matters *Brain, behavior, and immunity*. 2018; **70**:61-75.

31. Barnett BE, Hanna B, Parker G. Life event scales for obstetric groups *J Psychosom Res*. 1983; **27**:313-20.

32. Brown GW, Harris TO, Peto J. Life Events and Psychiatric-Disorders .2. Nature of Causal Link *Psychological Medicine*. 1973; **3**:159-76.

33. Brown GW, Sklair F, Harris TO, Birley JLT. Life-events and psychiatric disorders1 Part 1: some methodological issues *Psychological Medicine*. 2009; **3**:74.

34. Honnor MJ, Zubrick SR, Stanley FJ. The role of life events in different categories of preterm birth in a group of women with previous poor pregnancy outcome *Eur J Epidemiol*. 1994; **10**:181-8.

35. Ancold A, Stephen C. Development of a short questionnaire for use in epidemiological studies of depression in children and adolescents *Age (years)*. 1995; **6**.

36. Lobstein T, Baur L, Uauy R. Obesity in children and young people: a crisis in public health *Obesity reviews*. 2004; **5**:4-85.

37. StataCorp. Stata Statistical Software: Release 15. College Station, TX: StataCorp LLC; 2017.

38. Azur MJ, Stuart EA, Frangakis C, Leaf PJ. Multiple imputation by chained equations: what is it and how does it work? *International journal of methods in psychiatric research*. 2011; **20**:40-9.

39. Slopen N, Kubzansky LD, McLaughlin KA, Koenen KC. Childhood adversity and inflammatory processes in youth: a prospective study *Psychoneuroendocrino*. 2013; **38**:188-200.

40. Kautz MM, Coe CL, McArthur BA, Mac Giollabhui N, Ellman LM, Abramson LY, et al. Longitudinal changes of inflammatory biomarkers moderate the relationship between recent stressful life events and prospective symptoms of depression in a diverse sample of urban adolescents *Brain, Behavior, and Immunity*. 2019.

41. Davey CG, McGorry PD. Early intervention for depression in young people: a blind spot in mental health care *The Lancet Psychiatry*. 2018.

42. Harrington R, Clark A. Prevention and early intervention for depression in adolescence and early adult life *European archives of psychiatry and clinical neuroscience*. 1998; **248**:32-45.

43. Barnes J, Mondelli V, Pariante CM. Genetic contributions of inflammation to depression *Neuropsychopharmacology*. 2017; **42**:81.

44. Liu RT, Alloy LB. Stress generation in depression: A systematic review of the empirical literature and recommendations for future study *Clinical psychology review*. 2010; **30**:582-93.

45. Kuhlman KR, Horn SR, Chiang JJ, Bower JE. Early life adversity exposure and circulating markers of inflammation in children and adolescents: A systematic review and meta-analysis *Brain, behavior, and immunity*. 2019.

46. Gunnar MR, Wewerka S, Frenn K, Long JD, Griggs C. Developmental changes in hypothalamus–pituitary–adrenal activity over the transition to adolescence: normative changes and associations with puberty *Development and psychopathology*. 2009; **21**:69-85.

47. Clarke T-K, Zeng Y, Navrady L, Xia C, Haley C, Campbell A, et al. Genetic and environmental determinants of stressful life events and their overlap with depression and neuroticism *Wellcome open research*. 2018; **3**.

48. Bemmels HR, Burt SA, Legrand LN, Iacono WG, McGue M. The heritability of life events: an adolescent twin and adoption study *Twin Research and Human Genetics*. 2008; **11**:257-65.

49. Flouri E, Francesconi M, Papachristou E, Midouhas E, Lewis G. Stressful life events, inflammation and emotional and behavioural problems in children: A population-based study. Brain, behavior, and immunity. 2019 Feb 23.

Table 1. Descriptive statistics of all study variables in the analytic sample (n=4,263)			
Continuous variables			
	N	**M (SD)**	
Prenatal adverse life events	3,717	5.53 (2.92)	
Adverse life events 8 months	3,808	3.85 (2.51)	
Adverse life events 21 months	3,674	4.46 (2.82)	
Adverse life events 33 months	3,572	5.05 (3.04)	
Adverse life events 47 months	3,584	4.74 (3.16)	
Adverse life events 61 months	3,472	4.42 (2.97)	
Adverse life events 73 months	3,428	3.84 (2.83)	
Adverse life events 110 months	3,562	4.68 (3.14)	
MFQ score 10.5 years	3,802	3.90 (3.36)	
MFQ score 12.5 years	3,423	3.80 (3.73)	
MFQ score 13.5 years	3,099	4.62 (4.18)	
MFQ score 16.5 years	2,319	5.61 (5.43)	
MFQ score 17.5 years	2,185	6.40 (5.21)	
Inflammatory markers			
		M (SD)	**Range**
IL-6 total sample (pg/mL)*	4,263	1.21 (1.48)	0.00-20.05
First IL-6 tertile (pg/mL)	1,421	0.38 (0.12)	0.00-0.57
Second IL-6 tertile (pg/mL)	1,421	0.79 (0.14)	0.57-1.09
First and second IL-6 tertile (pg/mL)	2,842	0.58 (0.24)	0.00-1.09
Third (upper) IL-6 tertile (pg/mL)	1,421	2.46 (2.04)	1.09-20.05
CRP (mg/L)*	4,263	0.62 (1.97)	0.01-45.17
First CRP tertile (mg/mL)	1,423	0.08 (0.02)	0.01-0.13
Second CRP tertile (mg/mL)	1,419	0.21 (0.05)	0.14-0.34
First and second CRP tertile (mg/mL)	2,842	0.15 (0.07)	0.01-0.34
Third (upper) CRP tertile (pg/mL)	1,421	1.58 (3.20)	0.35-45.17
Categorical variables			
	N	**%**	
Obesity, 9 years	206	4.89	
Mother is university-educated	657	17.34	
Female	2,071	48.63	
Non-white	155	4.01	
Notes: MFQ=Mood and Feelings Questionnaire; IL-6 = interleukin 6; CRP = C-reactive protein. *Mean values for IL-6 and CRP are presented here for information			

Table 2. Mediation (by IL-6) model of depressive symptoms (MFQ)

Direct paths	Imputed Cases (n=4,263)			Complete Cases (n=3,438)		
	b	SE	95% CI	b	SE	95% CI
Unadjusted model						
Prenatal ALE → IL-6	0.00	0.00	-0.00 - 0.00	0.00	0.00	-0.00 - 0.00
Slope of postnatal ALE → IL-6	0.25**	0.06	0.12 - 0.38	0.23**	0.06	0.10 - 0.37
Intercept of postnatal ALE → IL-6	0.00	0.00	-0.00 - 0.01	0.00	0.00	-0.00 - 0.01
Prenatal ALE → Intercept of MFQ	0.01*	0.00	0.00 - 0.03	0.01*	0.00	0.00 - 0.03
Slope of postnatal ALE → Intercept of MFQ	0.44**	0.16	0.12 - 0.76	0.45**	0.17	0.11 - 0.79
Intercept of postnatal ALE → Intercept of MFQ	0.08**	0.01	0.06 - 0.11	0.08**	0.01	0.06 - 0.11
IL-6 → Intercept of MFQ	0.14**	0.03	0.07 - 0.22	0.11**	0.04	0.03 - 0.19
Fully adjusted model [1]						
Prenatal ALE → IL-6	0.00	0.00	-0.00 - 0.00	0.00	0.00	-0.00 - 0.00
Slope of postnatal ALE → IL-6	0.25**	0.06	0.12 - 0.38	0.23**	0.06	0.10 - 0.37
Intercept of postnatal ALE → IL-6	0.00	0.00	-0.00 - 0.01	0.00	0.00	-0.00 - 0.01
Prenatal ALE → Intercept of MFQ	0.01*	0.00	0.00 - 0.03	0.01	0.00	-0.001 - 0.03
Slope of postnatal ALE → Intercept of MFQ	0.43**	0.16	0.11 - 0.76	0.46*	0.17	0.10 - 0.81
Intercept of postnatal ALE → Intercept of MFQ	0.08**	0.01	0.06 - 0.11	0.10**	0.01	0.07 - 0.12
IL-6 → Intercept of MFQ	0.10**	0.03	0.03 - 0.18	0.07	0.04	-0.00 - 0.16

Notes: IL-6 = interleukin 6 (upper tertile vs. rest); ALE = Adverse life events; MFQ=Mood and Feelings Questionnaire (short form); SE=Standard error; 95% CI=95% Confidence interval. [1] Adjusted for: gender, ethnicity, maternal education, and BMI

* p<.05 ** p<.01