Supplementary materials

Supplementary Appendix 1. Data analysis plan

The association between maternal haemoglobin during pregnancy and mid-childhood educational attainment

1. Introduction

40% of pregnant women globally are anaemic, making anaemia the most common pregnancy complication worldwide, with the highest prevalence in low- and middle-income countries (LMIC)[1,2]. However even in high-income obstetric settings, haemoglobin (Hb) concentrations below the WHO definition of anaemia (<110 g/L)[1] are commonly observed. Hb concentrations fall physiologically during pregnancy as the expansion of the plasma volume exceeds the concomitant increase in red cell mass, meaning that measured Hb concentrations do not always reflect total oxygen carrying capacity especially later in pregnancy.

Associations have been reported between low maternal Hb concentrations during pregnancy and increased risk of adverse outcomes, including maternal and neonatal mortality, stillbirth, preterm birth (PTB), and low birth weight (BW)[2–4]. However, some studies suggest that high Hb concentrations are also associated with more pregnancy complications[5], particularly PTB and small-for gestational age (SGA)[6,7], as well as gestational diabetes mellitus (GDM)[8]. A U-shaped association between maternal haemoglobin and birth outcomes has therefore been postulated, although this has not been universally observed[9].

There is little available high-quality evidence exploring the relationship between maternal anaemia and later neurodevelopmental outcomes for the child, although recent UK guidelines suggest that improving maternal iron status may improve neurodevelopmental outcomes. Recent systematic reviews suggest either no associations between maternal iron status and child cognition[10] or possible detrimental effects of both maternal iron deficiency and excess during pregnancy with offspring neurodevelopment during childhood[11]. Given that recent guidance suggests an active and early approach to maternal iron supplementation in pregnancy[12], the nature of the relationship between maternal Hb and later childhood educational outcomes represents an important knowledge gap.

This study aims to investigate the associations between maternal haemoglobin levels during pregnancy in early and late pregnancy with offspring academic performance aged 5-7.

2. Research questions

1. Is there a linear or non-linear association between maternal Hb levels during pregnancy (at 12/representing early pregnancy, 28 weeks/representing late pregnancy, and delta Hb between early and late pregnancy) and not attaining expected educational standards between age 5-7?
2. Are any observed associations mediated by adverse birth outcomes (PTB, SGA, low birth weight, GDM)? Is there an association between maternal Hb and educational outcomes in pregnancies without adverse birth outcomes?
3. Is there any association between maternal anaemia and not attaining expected educational standards between age 5-7?

3. Hypothesis testing

Hypothesis : Maternal Hb levels in late pregnancy (28 weeks) are associated with childhood educational attainment, at least at age 5

Significance level (α) : 0.05

4. Defining datasets

1. Pregnancy Outcome Prediction Study (POPS) dataset, Department of Obstetrics & Gynaecology, University of Cambridge
    - POPS was a prospective cohort study of 4512 nulliparous women attending antenatal visits at the Rosie Maternity Hospital, Cambridge, England between January 2008-July 2012[13]

    There were 4 POPS study visits during pregnancy[13]: 1) at 12 weeks consisted of taking informed consent and phlebotomy, 2) at 20 weeks consisted of fetal biometry, phlebotomy, uterine and umbilical Doppler, 3) at 28 weeks consisted of fetal biometry, phlebotomy, uterine and umbilical Doppler, and 4) at 36 weeks consisted of fetal biometry, phlebotomy, uterine and umbilical Doppler.
    Data to use: maternal Hb during pregnancy, other pregnancy/antenatal factors (fetal growth parameters from antenatal ultrasounds, other biomarkers), maternal data (age, morbidities, parity, occupation, partner status, etc), perinatal information (gestational age, infant sex, birthweight, etc), other baseline demographics (including socioeconomic parameters).

4164 participants who had live born singleton infants were eligible for inclusion. List cleaning reduced this to 3722 mother/baby pairs who were confirmed alive and traceable at study commencement. 45 pairs opted out therefore linkage was attempted for 3677 pairs.

2. National Pupil Database (NPD) datasets, Department for Education
    - Data to use: early years, phonics, key stage 1
3. NHS digital data datasets
    - Data to use: hospital episode statistics (HES) at outpatient clinic, inpatient ward, or A&E unit to identify children with significant morbidities that can interfere with their academic performance, including 1) major congenital anomaly, genetic, or chromosomal difference; 2) neurological or cerebrovascular disease; 3) childhood malignancy; 4) inborn errors of metabolism or immunodeficiency; 5) congenital hearing impairment or visual loss; early onset of severe organ dysfunction; 6) dependency on medical machines, functional implants, artificial feeding or breathing.
    - Children without HES data during the time frame (1 year) are assumed to not have any major medical morbidities. There may be a small number of children who were being managed entirely via the private health care system or not have required any hospital management at all over the course of a year, however given the medical complexity of the exclusion criteria, this is unlikely and would only apply to a very small number of children.

All data sets are identified only by anonymised study ID

5. Defining exposures

- Maternal Hb levels measured at booking (12 weeks of gestation) and at the third trimester (28 weeks of gestation) and delta Hb as continuous variables
- Maternal anaemia as categorical variable (yes/no) [definition according to BSH guidelines: Hb <110 g/L in first trimester, <105 g/L in second and third trimesters[12]]

6. Defining outcomes: educational attainment at age 5-7 years

Early years foundation stage (EYFS), representing educational attainment at the age of 5 with 7 domains of assessment: 3 prime areas consisting of communication and language (CL), physical development (PHY), personal-social-emotional development (PSE), and 4 specific areas consisting of literacy (LIT), mathematics (MAT), understanding the world (UTW), expressive arts and design (EXP). There are in total 17 early learning goals (ELG) of those 7 domains of EYFS. Children were assessed at the end of reception year to be either at an 'emerging' (=1), 'expected' (=2), or 'exceeding' (=3) level of development in the 17 ELG, making 17 as minimum and 51 as maximum scores, respectively. Good level of development (GLD) is achieved if children have *at least* the expected level for the ELG in CL, PHY, PSE, LIT, and MAT.

Phonics assessment, representing educational attainment at the age of 6 (end of year 1), is measured by using 40-word test with national threshold for pass is ≥32. Children who did not pass would repeat this assessment at the end of year 2.

Key stage 1 (KS1), representing educational attainment at the age of 7 (end of year 2), consisting of reading, writing, mathematics, and science tests. Corresponding teachers would judge the test results compared to standard: 'below', 'at', 'above expected'.

Supplementary Appendix 1 - Table 1 Type of each outcome variable analysis plan

Dependent variables/Outcomes: School outcomes at age 5-7 years	
Variable	**Type**
Early years	Binary categories: achieved GLD vs non-GLD
Phonics assessment	Binary categories: first-time passed vs failed
Key stage 1	Binary categories: below vs at/above, per domain being assessed: reading, writing, mathematics, science

7. Defining maternal occupation data handling

Maternal occupations were coded into 9 groups based on ISCO-08 - ONS SOC Hierarchy 2010 (onsdigital.github.io):

1 = Managers, Directors, and Senior Officials; 2 = Professional occupations; 3 = Associate Professional and Technical Occupations; 4 = Administrative and Secretarial Occupations, 5 = Skilled Trades Occupations; 6 = Caring, Leisure, and Other Service Occupations; 7 = Sales and Customer Service Occupations; 8 = Process, Plant, and Machine Operatives; 9 = Elementary occupations

and 2 categories were additionally derived:

0 = refers to all occupations with no stable regular income, including unemployed, housewife or homemaker, student, voluntary work; and 10 = self-employed without any other information

These 10 occupation groups were then modified into 4 categories:

M = managerial occupations or group 1; P = professional occupations or group 2; A = Associate and administrative occupations or group 3 and 4; E = occupations that require

elementary-level educations or group 5, 6, 7, 8, and 9; U = refers to un-, freelance, or self-employment (excluding business owner)

8. Defining covariates

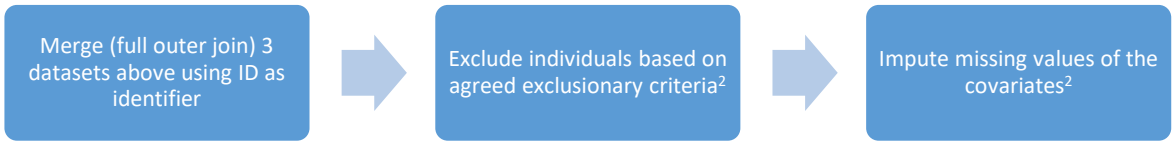
Supplementary Appendix 1 - Table 2 List of covariates used in the analyses

	Variable	Type	Notes
Maternal factors	Age	Numeric (rounded to one decimal place)	Derived from date of POPS of recruitment – date of birth (in years)
	Pre-pregnancy maternal BMI	Numeric (rounded to one decimal place)	Derived from maternal weight at booking divided by the square of measured height (in kg/m^2)
	Ethnicity	Factor (binary categories)	White (Caucasian) vs non-white
	Occupation	Factor	Group 1, 2, 3, 4 (see point d – maternal occupation data handling)
	Partner status	Factor (binary categories)	Yes/no
	Smoking history	Factor	"never smoked", "quit during pregnancy", "quit before pregnancy", and "currently smoking during pregnancy" (could further be grouped into binary variables: smoking vs not smoking during pregnancy)
Infant factors	Gestational age (GA)	Numeric (integer)	In weeks, taking into account remaining days as fraction of corresponding number of weeks, e.g. 37 weeks and 3 days = 37.4 weeks
	Sex	Factor (binary categories)	Male/female
	Birth seasonality	Factor	1. born between 1st of December at 00:00 to 28th (or 29th during a leap year) of February at 23:59 (representing winter) 2. born between 1st of March at 00:00 to 31st of May at 23:59 (representing spring) 3. born between 1st of June at 00:00 to 31st of August at 23:59 (representing summer) 4. born between 1st of September at 00:00 to 30th of November at 23:59 (representing autumn)

Potential modifiers			
	SGA	Factor	Yes/No SGA is defined as BW <10th percentile based on relevant growth reference (UK 1990 growth reference is used in this study), adjusted for sex and GA
	PET	Factor	Yes/No Pre-eclampsia as defined in previous POPS publication[14]
	GDM status	Factor	Yes/No GDM status as defined by contemporaneous local testing protocol
Socioeconomic factors	Index of multiple deprivation (IMD)	Numeric (integer	Based on the English indices of deprivation 2007 (postcode-based)
	School funding type	Factor	Academy (state-funded), community, voluntary
	Year	Factor	Year when the assessment being conducted (all outcomes will be adjusted for year, ***except*** numerical total EY z-scores)
	Maternal occupation	Factor	Group 1, 2, 3, 4 (see point d – maternal occupation data handling)
Childhood physical health	Relevant childhood comorbidities	Factor (binary categories)	Any evidence of these diseases, recorded from hospital episode statistics (HES)-NHS digital from either A&E attendances, outpatient appointments, or inpatient admissions at NHS hospitals in England: 1) major congenital anomaly, genetic, or chromosomal difference; 2) neurological or cerebrovascular disease; 3) childhood malignancy; 4) inborn errors of metabolism or immunodeficiency; 5) congenital hearing impairment or visual loss; early onset of severe organ dysfunction; 6) dependency on medical machines, functional implants, artificial feeding or breathing

9. Defining general analytical plan

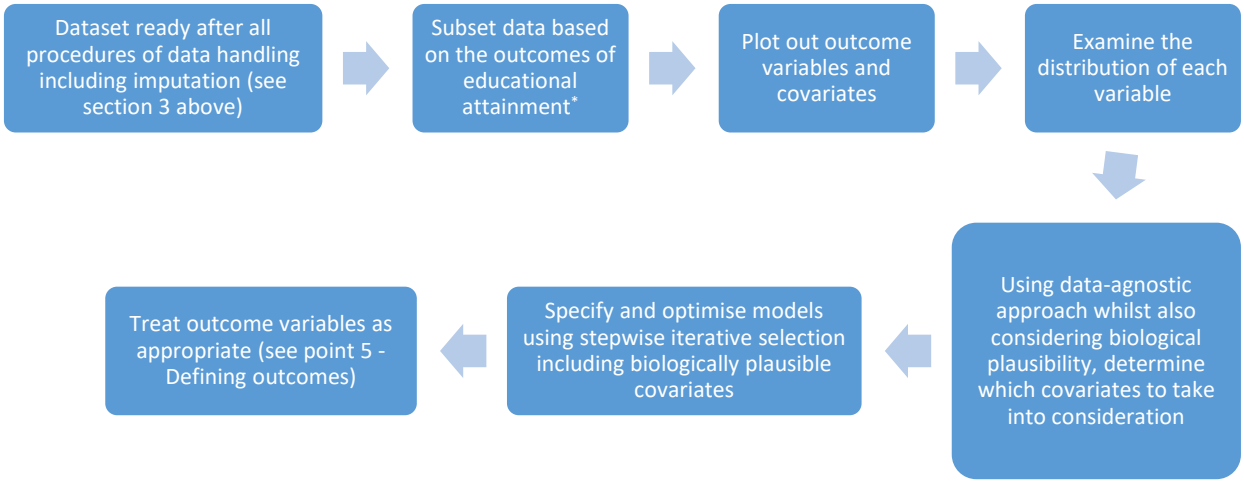
Supplementary Appendix 1 - Figure 1 Pre-analysis flowchart



Notes:

1. Full outer join/merge will return all rows/subjects from each dataset and match up rows where possible
- Subjects without any educational data in the NPD will also be omitted from the analyses.
2. Multiple imputations by chained equations (MICE)[15] will be employed to replace covariates' missing data values under assumption that the data are missing at random (MAR).

Supplementary Appendix 1 - Figure 2 Strategy of data analyses



*3 models will be constructed based on NPD academic outcomes: 1) early years, 2) phonics, 3) Key Stage/KS1

Supplementary Appendix 1 – Table 3 Statistical methodology

Methods	**Outcomes**
Descriptive analyses	Descriptive comparisons between groups (whole POPS vs included vs excluded populations; between exposure groups of interest, e.g. anaemic vs non-anaemic mothers) - For numerical variables (normal distribution presented in mean ±standard deviation; non-normal distribution in median (interquartile range)) - For categorical variables (in N and %)
Correlation analyses	Correlation coefficients and *p values* (Pearson's for normal distribution and Spearman's for non-normal distribution)

Linear and non-linear regression models (including generalised additive models)	B coefficients, standard errors, and *p values*
Logistic regression models	Odd ratios, confidence intervals, and *p values*

Analysis strategy:

1. Generalised additive models (GAM) and regression models will be run to assess the associations between continuous exposure variables (maternal Hb at 12 wks, maternal Hb at 28 wks, delta Hb) and binary outcomes (attained v did not attain expected standard) for childhood educational attainment, separately at age 5, 6, and 4x domains of age 7.
2. The associations between maternal anaemia at 12 and 28 wks (BSH definitions; binary yes/no) and childhood educational outcomes (as above) will be tested using generalised linear models (GLM) or logistic regression models.
3. Unadjusted models will be run and compared with those adjusted for all covariates (see point 7 above)
4. Other pregnancy pathologies (e.g. SGA, PET, GDM) will be individually tested as possible effect modifiers and therefore included sequentially as covariates and tested for positive interactive effects with maternal Hb exposure variables (see below)

10. Sensitivity and additional analyses

If interactive effects of other pregnancy pathologies and Hb exposure variables are found then to take account of possible interactions with other co-variates, we will construct further logistic regression models stratifying low/high Hb levels with/without other adverse outcomes, e.g. comparing educational outcomes between children born SGA to mothers with low Hb, children born SGA to mothers without low Hb, children born AGA of mothers without low Hb, children born AGA of mothers without low Hb. In order to ensure that these analyses are adequately powered we will include models using both BSH definition of anaemia (as above), and also lowest/highest 10 & 20% of Hb values within the POPS study.

GAM or logistic regression models will be fitted to interrogate the associations between maternal Hb levels and mid-childhood educational attainment among a 'healthy' population, i.e. children born at term to mothers without any perinatal morbidities or adverse birth outcomes (NICU admission, severe childhood morbidity).

If the change in Hb between 12 -> 28 weeks (deltaHb) is associated with poor educational outcomes, then we will perform additional analyses to interrogate this result further. Using data from other studies of population-level changes of maternal Hb concentrations between the first and last trimesters we will add additional outcomes variables of (i) >5 g/L Hb drop [12] (yes/no) and (ii) >14 g/L or >11% (yes/no)[16]

Unfortunately, additional analyses by taking into account ferritin levels and iron supplementation during pregnancy are not feasible due to data unavailability.

11. Quality control

Regular weekly/fortnightly meetings will be conducted between the data analyst and PI to discuss ongoing data analyses. Data will be discussed with all other POPStar study team members once ready and being released from the SRS.

12. Timeline

Supplementary Appendix 1 – Table 4 Study Timeline

Month/date (2023)	Action
March-15 April	• Data analysis plan • Data ingest request to SRS (maternal haemoglobin levels)
15-30 April	Executing analyses as planned, manuscript draft writing
1-15 May	Circulation of preliminary results and manuscript draft to the whole POPStar team
15-30 May	• Manuscript revision • Clearance of figures/tables/supplementary materials from SRS
1 June	• Manuscript submission

13. References

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Supplementary Appendix 2. Assessment of educational attainment aged 5, 6, and 7 years in the UK

Educational outcomes were obtained from the National Pupil Database, a national record-level data resource curated by the UK Department for Education (DfE). All fully/partially state-funded schools in England have a mandatory reporting requirement to return individual-level pupil data on an annual basis.

In the UK, children typically start school/reception (also called Year 0 of primary school) in September after their 4th year birthday. They must have started full-time education by the start of the next term following their 5th birthday. Therefore, the vast majority of children will be aged 5, 6, and 7 respectively in the first 3 school years.

Assessment aged 5

At the end of the first school year (Year 0/Reception), educational attainment is assessed in 7 key areas, divided into 17 early learning goals (ELG) (see table below). For each ELG, children are assessed as 'emerging' (=1), 'expected' (=2), or 'exceeding' (=3) level of development. The minimum possible score is therefore 17 and the maximum is 51. Scores are assigned on the basis of ongoing observations by the class teacher.

The standardised binary outcome reported by the Department for Education is 'Good level of development' (GLD). This is achieved if children attain the expected level or above for the ELGs in the areas of (i) communication and language, (ii) physical development, (iii) personal, social, and emotional development, (iv) literacy, and (v) mathematics.

Supplementary Appendix 2 – Table 1 Age 5 Key areas and early learning goals

No	Key areas	Early learning goals	Included as parameters of 'Good Learning Development'
1	**Communication and language**	1. Listening & attention 2. Understanding 3. Speaking	V
2	**Physical development**	1. Moving and handling 2. Health and self-care	V
3	**Personal, social, and emotional development**	1. Self-confidence and self-awareness 2. Managing feelings and behaviour 3. Making relationships	V
4	**Literacy**	1. Reading 2. Writing	V
5	**Mathematics**	1. Numbers 2. Shape, space, and measures	V
6	**Understanding the world**	1. People and communities 2. The world 3. Technology	X
7	**Expressive arts and design**	1. Exploring and using media and materials 2. Being imaginative	X

Assessment aged 6

Towards the end of the second school year (Year 1), pupils are assessed using a standardised 40-word test, administered by a trained education professional known to the child. This assessment results in a numerical global score (1-40) for each child, which is then converted into a binary outcome by comparing their performance to a national threshold (set on a yearly basis, usually ~32). Children who do meet the national threshold aged 6 are given the opportunity to repeat this test aged 7, however in our analysis we used only the scores from each child's first attempt aged 6.

Assessment aged 7 (Key Stage 1 or KS1)

At the end of the third school year (Year 2), children are assessed by a teacher who has had contact with the pupil over a period of time. Four key domains (reading, writing, mathematics, and science) are assessed as either 'below', 'at', or 'above' expected standards. A binary outcome (below *versus* at or above standard) was generated for each KS1 domain.

Further details of assessment criteria, standardisation of testing, and audit/validation methods can be found in the following documents:

Department for Education. *National Pupil Database.* https://www.gov.uk/government/collections/national-pupil-database. Published 2018. Accessed November 20, 2021.

Department for Education. *Early years foundation stage profile results: 2018 to 2019*; https://www.gov.uk/government/statistics/early-years-foundation-stage-profile-results-2018-to-2019. Published 17 October 2019, Last updated 7 April 2020. Accessed 4 May, 2022.

Department for Education. *National curriculum assessments at key stage 1 and phonics screening checks in England, 2018;* https://www.gov.uk/government/statistics/phonics-screening-check-and-key-stage-1-assessments-england-2018/national-curriculum-assessments-at-key-stage-1-and-phonics-screening-checks-in-england-2018. Published 27 September 2018, Updated 25 October 2019. Accessed 4 May 2022.

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Office of Qualifications and Examinations Regulation (Ofqual). *Regulatory framework for national assessments, National curriculum and early years foundation stage assessments;* https://assets.publishing.service.gov.uk/government/uploads/system/uploads/attachment_data/file/685610/National_assessment_regulatory_framework_-_March_2018.pdf . Published March 2018. Accessed 4 May 2022.

Supplementary Appendix 3. Childhood medical conditions for model adjustment

• Major congenital anomaly, genetic, or chromosomal difference
• Neurological or cerebrovascular disease
• Childhood malignancy
• Inborn errors of metabolism or immunodeficiency
• Congenital hearing impairment or visual loss
• Early onset of severe organ dysfunction
• Dependency on medical machines, functional implants, artificial feeding/breathing

These conditions could confound the association between disrupted intrauterine environment and reduced educational performance and therefore were adjusted in primary analyses. This pre-specified morbidity list was defined in consultation with a paediatric consultant (HW). A full year of HES was obtained for each child, as it is highly likely that any child with a significant excludable health condition would have at least one HES-recorded appointment within a year. Children without any HES data recorded during the time frame (1 year) are assumed to not have any of the pre-specified morbidities. There may be a small number of children who were being managed entirely via the private health care system or not have required any hospital management at all over the course of a year, however given the medical complexity of the pre-specified conditions, this is unlikely and would only apply to a very small number of children.

Supplementary Table 1. Baseline characteristics of all eligible POPS participants and the analytic sample

Characteristic	Analytic sample (N=3285)	All POPS participants (N= 4164)
Age at recruitment, yrs, mean (SD)	29.7 (5.1)	29.9 (5.1)
Height, cm, mean (SD)	165.1 (6.4)	165.2 (6.4)
BMI, kg/m^2	25.3 (4.8)	25.1 (4.7)
Ethnicity, No./total (%) of White/Europeans	3133/3285 (95.3)	3861/4094 (94.3)
Smoking history, No. (%) of 'never smoked'	1895 (57.7)	2475 (59.4)
Alcohol consumption, No. (%) of 'does not drink'	3132 (95.3)	3975 (95.5)
Partner status, No./total (%) of 'with partner'	3225 (98.2)	4011 (96.4)
Missing, No. (%)	<10	<10
IMD score, mean (SD)	10.2 (6.5)	10.3 (6.5)
Missing, No. (%)	137 (4.2)	171 (4.1)
Occupation, No. (%) of 'management level/professional'	1598 (48.6)	*Not coded*
Gestational age, weeks, mean (SD)	39.9 (1.7)	40 (1.7)
Missing, No. (%)	<10	<10
Sex, No. (%) of female	1637 (49.8)	2065 (49.6)
Birth seasonality, No. (%) of 'born in autumn'	891 (27.1)	1123 (27)
Mode of delivery, No. (%) of 'vaginal delivery'	2367 (72)	3008 (72.4)
Missing, No. (%)	<10	<10
Birth weight, centile, mean (SD)	45.8 (26.2)	45.6 (26)

Data are obtained from the original dataset before imputation. For fields where there is no "missing" row, data were 100% complete. Maternal age was defined as age at recruitment. Maternal BMI was derived from weight measured at recruitment divided by the square of height (kg/m^2). All other maternal characteristics were either self-reported at the 20 week-gestational age visit, from examination of the clinical record, or linkage to the hospital's electronic databases. IMD score 2007 was calculate based on census data from the area of the mother's postcode. Birth weight percentiles were calculated using UK 1990 growth reference. BMI indicates body mass index; IMD, index of multiple deprivation; N, sample size. There are no differences between the analytic sample and the entire POPS cohort, with the exception of partner status; this is a small but statistically significant difference amounting to an additional 1.6% of mothers who reported having a partner in the analytic sample.

Supplementary Table 2. Association between maternal haemoglobin concentration at 12 weeks and pregnancy complications

Pregnancy complication		Hb at 12 weeks			Anaemia at 12 weeks		
		aOR	95% CI	p	aOR	95% CI	p
GDM	N=154	0.98	0.79-1.22	0.87	1.11	0.77-4.84	0.11
Pre-eclampsia	N=233	1.20	1.00-1.42	0.05	0.36	0.06-1.21	0.17
SGA	N=296	0.99	0.85-1.16	0.97	0.93	0.36-2.03	0.87
Pre-term birth	N=142	1.03	0.84-1.29	0.74	NA		

Hb: maternal haemoglobin in g/L (n=3162). Anaemia at 12 weeks defined as Hb <110g/L. For anaemia at 12 weeks, children whose mothers were not anaemic were the referent group (n=3089/3162). GDM: gestational diabetes mellitus; SGA: small-for-gestational age, defined as <10th centile for gestational age and sex on British 1990 growth charts; N: sample size.

Models covariates included in all models: maternal factors (age at pregnancy, BMI at recruitment, ethnicity, partner status, smoking history), infant factors (gestational age, sex), socio-economic factors (IMD, maternal occupation).

NA denotes insufficient observations for algorithm convergence

No adjustments for multiple hypothesis testing have been made, as no p value met the threshold for significance

Supplementary Table 3. Association between maternal haemoglobin (Hb) during pregnancy and educational attainment aged 5-7 years

Age/domain of assessment		**Maternal Hb at 12 weeks**		**Maternal Hb at 28 weeks**	
		OR (95% CI)	P	OR (95% CI)	p
Age 5	*Unadjusted*	0.97 (0.87-1.08)	0.60	0.85 (0.75-0.96)	0.01
	Interaction with SGA	1.07 (0.68-1.67)	0.78	1.34 (0.81-2.21)	0.25
	Interaction with PET	1.21 (0.76-1.94)	0.43	0.94 (0.55-1.56)	0.81
	Interaction with GDM	0.71 (0.45-1.14)	0.16	0.72 (0.4-1.28)	0.25
Age 6	*Unadjusted*	1.04 (0.82-1.11)	0.55	1.08 (0.92-1.28)	0.35
	Interaction with SGA	1.04 (0.62-1.75)	0.89	0.88 (0.5-1.55)	0.67
	Interaction with PET	1.07 (0.62-1.86)	0.80	0.75 (0.4-1.36)	0.35
	Interaction with GDM	0.71 (0.4-1.26)	0.24	0.51 (0.27-0.96)	0.04
Age 7 - Reading	*Unadjusted*	1.02 (0.9-1.16)	0.76	0.94 (0.82-1.09)	0.42
	Interaction with SGA	1.46 (0.87-2.48)	0.16	1.3 (0.75-2.29)	0.35
	Interaction with PET	1.12 (0.66-1.94)	0.68	0.65 (0.36-1.16)	0.15
	Interaction with GDM	0.99 (0.59-1.68)	0.97	0.66 (0.39-1.23)	0.20
Age 7 - Writing	*Unadjusted*	1.09 (0.98-1.22)	0.12	1.01 (0.89-1.14)	0.90
	Interaction with SGA	1.3 (0.8-1.25)	0.29	1.07 (0.63-1.81)	0.80
	Interaction with PET	0.97 (0.61-1.57)	0.91	0.77 (0.45-1.27)	0.31
	Interaction with GDM	1.13 (0.7-1.87)	0.63	0.85 (0.48-1.48)	0.57
Age 7 - Maths	*Unadjusted*	1.1 (0.97-1.24)	0.15	0.94 (0.81-1.08)	0.35
	Interaction with SGA	1.2 (0.73-1.99)	0.48	1.12 (0.65-1.96)	0.67
	Interaction with PET	1.26 (0.75-2.14)	0.39	0.84 (0.47-1.47)	0.54
	Interaction with GDM	1.41 (0.84-2.44)	0.2	1.07 (0.58-1.96)	0.83
Age 7 - Science	*Unadjusted*	1.03 (0.88-1.2)	0.73	0.84 (0.7-1.0)	0.05
	Interaction with SGA	1.23 (0.66-2.36)	0.51	1.38 (0.67-2.89)	0.39
	Interaction with PET	1.13 (0.59-2.2)	0.71	0.62 (0.29-1.25)	0.20
	Interaction with GDM	1.86 (0.98-3.7)	0.06	0.97 (0.44-2.13)	0.94

Odds ratios (OR) with 95% confidence intervals are displayed for unadjusted models alongside the interaction terms for each relevant pregnancy complication from generalized linear models adjusted for the presence or absence of each complication. N=3089 for 12 weeks, N=2257 for 28 weeks. N= sample size.

No adjustments for multiple hypothesis testing have been made, as no p values for interactions met the threshold for significance

Supplementary Table 4. Association between maternal haemoglobin parameters and not achieving expected educational standard at each age/domain

Education parameter		Delta Hb aOR (95%CI)	p	Highest decile aOR (95%CI)	p
Age 5	*Unadjusted*	1.14 (0.99-1.31)	0.05	0.79 (0.55-1.11)	0.19
	Adjusted	1.11 (0.96-1.30)	0.17	0.75 (0.50-1.09)	0.14
Age 6	*Unadjusted*	1.08 (0.92-1.28)	0.35	0.82 (0.52-1.25)	0.38
	Adjusted	1.00 (0.83-1.22)	0.92	0.81 (0.49-1.26)	0.36
Age 7-Reading	*Unadjusted*	1.02 (0.88-1.19)	0.76	0.71 (0.46-1.06)	0.10
	Adjusted	0.92 (0.78-1.09)	0.34	0.77 (0.49-1.16)	0.23
Age 7-Writing	*Unadjusted*	1.04 (0.91-1.20)	0.55	0.90 (0.63-1.26)	0.56
	Adjusted	1.01 (0.87-1.18)	0.85	0.94 (0.64-1.35)	0.74
Age 7-Maths	*Unadjusted*	1.09 (0.94-1.28)	0.24	0.84 (0.56-1.23)	0.39
	Adjusted	1.02 (0.86-1.20)	0.85	0.87 (0.57-1.30)	0.51
Age 7-Science	*Unadjusted*	1.15 (0.95-1.40)	0.14	0.87 (0.51-1.39)	0.58
	Adjusted	1.05 (0.85-1.29)	0.65	0.93 (0.54-1.52	0.78

Odds ratios (OR) with 95% confidence intervals are displayed. Delta Hb is a continuous variable calculated as the change in maternal haemoglobin (Hb) between 12 and 28 weeks of pregnancy (N=2448).

The highest decile of maternal Hb at 28 weeks was defined as a binary variable using a population-specific cut-off. The referent group are the children of mothers not in the highest decile of maternal Hb at 28 weeks (N=2034/2257). N=sample size.

Covariates included in all adjusted models: maternal factors (age at pregnancy, BMI at recruitment, ethnicity, partner status, smoking history), infant factors (gestational age, sex, birth seasonality, childhood physical health), socio-economic factors (IMD, school funding type, academic year, maternal occupation), potential effect modifiers (infant small-for-gestational age status, maternal pre-eclampsia, maternal gestational diabetes mellitus).

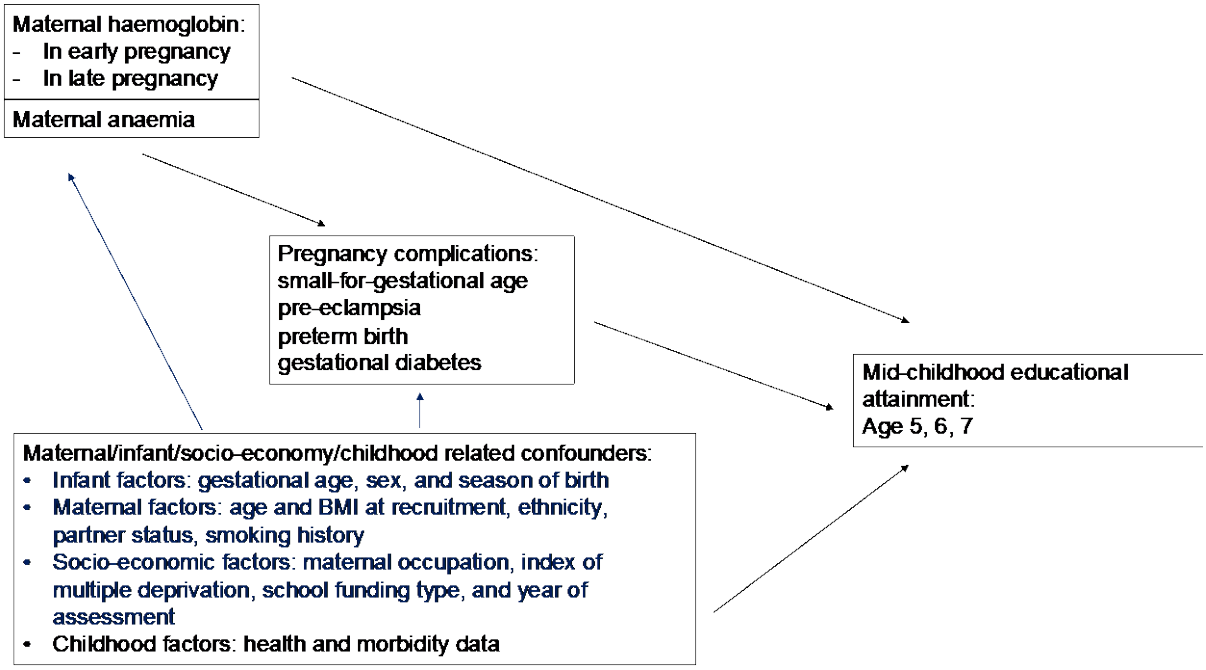
No adjustments for multiple hypothesis testing have been made, as no p value met the threshold for significance

Supplementary Table 5. STROBE Statement—checklist of items that should be included in reports of observational studies

	Item No	Recommendation	Page number
Title and abstract	1	(*a*) Indicate the study's design with a commonly used term in the title or the abstract	1
		(*b*) Provide in the abstract an informative and balanced summary of what was done and what was found	2-4
Introduction			
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported	5-6
Objectives	3	State specific objectives, including any prespecified hypotheses	6
Methods			
Study design	4	Present key elements of study design early in the paper	6-7
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection	6-7
Participants	6	(*a*) *Cohort study*—Give the eligibility criteria, and the sources and methods of selection of participants. Describe methods of follow-up *Case-control study*—Give the eligibility criteria, and the sources and methods of case ascertainment and control selection. Give the rationale for the choice of cases and controls *Cross-sectional study*—Give the eligibility criteria, and the sources and methods of selection of participants	7
		(*b*) *Cohort study*—For matched studies, give matching criteria and number of exposed and unexposed *Case-control study*—For matched studies, give matching criteria and the number of controls per case	NA
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable	7-8
Data sources/ measurement	8*	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group	7-8
Bias	9	Describe any efforts to address potential sources of bias	7-9
Study size	10	Explain how the study size was arrived at	9
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why	8
Statistical methods	12	(*a*) Describe all statistical methods, including those used to control for confounding	7-9
		(*b*) Describe any methods used to examine subgroups and interactions	8-9
		(*c*) Explain how missing data were addressed	8-9
		(*d*) *Cohort study*—If applicable, explain how loss to follow-up was addressed *Case-control study*—If applicable, explain how matching of cases and controls was addressed *Cross-sectional study*—If applicable, describe analytical methods taking account of sampling strategy	NA
		(*e*) Describe any sensitivity analyses	

Results			
Participants	13*	(a) Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed	9 and Fig 1
		(b) Give reasons for non-participation at each stage	Fig 1
		(c) Consider use of a flow diagram	Fig 1
Descriptive data	14*	(a) Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders	9, Suppl Tab 1
		(b) Indicate number of participants with missing data for each variable of interest	Suppl Tab 1
		(c) *Cohort study*—Summarise follow-up time (eg, average and total amount)	NA
Outcome data	15*	*Cohort study*—Report numbers of outcome events or summary measures over time	9-10
		Case-control study—Report numbers in each exposure category, or summary measures of exposure	
		Cross-sectional study—Report numbers of outcome events or summary measures	
Main results	16	(*a*) Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included	9-10, Tab 2, Suppl Tab 3&4
		(*b*) Report category boundaries when continuous variables were categorized	7, Tab 2&3
		(*c*) If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period	NA
Other analyses	17	Report other analyses done—eg analyses of subgroups and interactions, and sensitivity analyses	Supl Tab 3
Discussion			
Key results	18	Summarise key results with reference to study objectives	11
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias	12-13
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence	11-13
Generalisability	21	Discuss the generalisability (external validity) of the study results	12-13
Other information			
Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based	14-15

Supplementary Figure 1. Directed Acyclic Graph



Supplementary Figure 2. Association between maternal Hb at 28 weeks/delta Hb and pregnancy complications

Supplementary Figure 2a – Association between maternal Hb at 28 weeks and birth weight

Supplementary Figure 2b - Association between maternal Hb at 28 weeks and risk of preterm birth

Supplementary Figure 2c - Association between delta Hb and birth weight

Supplementary Figure 3. Sensitivity analyses using a ‘healthy’ population only