Modelling the contribution of genetics and clinical measures to birth weight and risk of large-for-gestational-age in mothers and babies of European and South Asian ancestry

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PhD Thesis

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Abstract

Birthweight is of major interest because of its associations with the baby's survival rate in the first year of life, childhood development and the onset of diseases in later adult life. Maternal characteristics are important in determining a baby's birthweight, and the fetal genotype is a crucial determinant of offspring birthweight. However, the contribution of genetics on top of other determinants of birthweight has not been explored before. The first part of this PhD investigated the contributions of genetics in explaining variation in offspring birthweight on top of routinely available clinical features.

An important feature associated with a baby's survival rate and its adult health, birthweight, varies across ethnic groups. It has been found that South Asian babies are on average of lesser weight than European babies despite a higher maternal fasting glucose level, a higher parity, and a lower rate of smoking in the mothers. Genetic associations with birthweight within South Asians and Europeans have been compared before but comparison between the contributions of genetics on top of other clinical features in the two ethnic groups has not been done before and this is what the second part of this thesis aimed at investigating.

Finally, to cater for antenatal and postnatal care, it is important to be able to predict a baby's birthweight. Current clinical practice uses the mother's fasting glucose level as a means of assessing the risk of a baby being born large-for-gestational age (LGA), but this determinant of birthweight has not been used in a prediction of LGA before in healthy pregnancies. The final part of this PhD aims at building a clinical prediction model, using maternal fasting glucose and other routinely available clinical features for estimating the risk of LGA in babies in a European and South Asian population. In conclusion, this thesis has investigated the associations of genetics with birthweight, on top of other routinely available clinical features in European and South Asian babies as well as the prediction of risk of delivering a baby with LGA in these two ethnicities.

Contents

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Author's Declaration

I was involved in the design, implementation, analysis, and manuscript preparation for all of the studies that form this thesis, as were Rachel Freathy and Beverley Shields.

The specific roles of other co-authors are as follows:

Chapter 1: Maneka Haulder wrote the first draft of the chapter and Rachel Freathy and Beverley Shields provided advice on making critical revisions to the chapter.

Chapter 2: Maneka Haulder wrote the first draft of the chapter and Rachel Freathy provided advice on the genotyping and Beverley Shields provided advice on the methodology.

Chapter 3: Maneka Haulder, Andrew T Hattersley, Beverley Shields and Rachel Freathy conceived and designed this study. Bridget Knight, Beverley Shields and Andrew Hattersley contributed to the collection and management of cohort data. Maneka Haulder undertook the analyses, with support from Alice Hughes and Robin Beaumont. Maneka Haulder, Alice Hughes, Andrew Hattersley, Beverley Shields and Rachel Freathy discussed and interpreted the data. Maneka Haulder wrote the first draft of the paper, with support from Beverley Shields and Rachel Freathy. All authors read and made critical revisions to the manuscript. All authors read and made critical revisions to the manuscript, and all authors read and approved the final version of the manuscript.

Chapter 4: Maneka Haulder, Andrew Hattersley, Beverley Shields and Rachel Freathy conceived and designed this study. Brandon Lim, Daniel Leirer, Alice Hughes and Robin Beaumont contributed to the management of cohort data.

Maneka Haulder undertook the analyses, with support from Brandon Lim, Daniel Leirer, Alice Hughes and Robin Beaumont. Maneka Haulder, Andrew Hattersley, Beverley Shields and Rachel Freathy discussed and interpreted the data. Maneka Haulder wrote the first draft of the paper. Beverley Shields and Rachel Freathy read and suggested critical revisions to the manuscript.

Chapter 5: Maneka Haulder, Beverley Shields and Rachel Freathy conceived and designed this study. Bridget Knight, Beverley Shields and Andrew Hattersley contributed to the collection and management of EFSOCH data. Brandon Lim, Daniel Leirer, Alice Hughes and Robin Beaumont contributed to the management of Born-in-Bradford data. Maneka Haulder undertook the analyses, with support from Beverley Shields. Maneka Haulder, Andrew Hattersley, Beverley Shields and Rachel Freathy discussed and interpreted the data. Maneka Haulder wrote the first draft of the paper, with support from Beverley Shields and Rachel Freathy. All authors read and made critical revisions to the manuscript.

Chapter 6: Maneka Haulder wrote the first draft of the chapter and Rachel Freathy and Beverley Shields provided advice on making critical revisions to the chapter.

Definitions

AC	**Abdominal Circumference**
BPD	**Biparietal Diameter**
BiB	**Born in Bradford**
BMA	**Bayesian Model Averaging**
BMI	**Body Mass Index**
CRL	**Crown Rump Length**
DOHaD and	**Developmental Origins of Health Disease**
EGG	**Early Growth Genetics Consortium**
EFSOCH	**Exeter Family Study of Childhood Health**
EFW	**Estimated Fetal Weight**
FL	**Fermur Length**
GDM	**Gestational Diabetes Mellitus**
GRS	**Genetic Risk Score**
GWAS	**Genome Wide Association Studies**
HC	**Head Circumference**
HIV	**Human immunodeficiency virus**
IUGR	**Intrauterine growth restriction**
LGA	**Large-for-gestational age**
NHS	**National Health Service**
OGTT	**Oral Fasting Glucose Test**
SD	**Standard Deviation**
SDS	**Standard Deviation Score**
SE	**Standard Error**
SGA	**Small-for-gestational age**
T1D	**Type 1 Diabetes**
T2D	**Type 2 Diabetes**
UKBB	**UK Biobank**

Chapter 1
Introduction

1.1 Why is it important to study birthweight?

Birthweight is a key factor in epidemiological studies as this phenotype is freely and widely available, low birthweight has a strong relationship with infant mortality (Wilcox, 2001) and both low and high birthweight are associated with poor perinatal outcomes and poorer later-life health outcomes.

According to a study (Biks et al., 2021), birthweight data is easily and freely available and is mostly collected from household surveys. In the UK, neonatal data is stored on electronic patient records (EPR), on a web-based platform, BadgerNet and managed by an approved NHS (National Health Service) supplier, Clevermed Ltd (Battersby et al., 2018) and overall, birthweight data is easily accessible.

The strong relationship between low birthweight and a baby's survival rate can predict a baby's risk of mortality. Low birth weight is associated with poor perinatal outcomes (for example, stillbirth, pre-term birth and SGA) (Dutton et al., 2012) and poorer later-life health outcomes such as diabetes, cardiovascular diseases, cancers and other problems. High birthweight is associated with type 2 diabetes (T2D) and cardiometabolic risks in the long term. Thus, by studying birthweight, it makes it easier to understand the causes of these adverse health outcomes and work towards prevention.

Complications with birthweight can occur due to both a lower and higher birth weight than average and expand to health problems in childhood and adulthood. Common complications at birth with both low and high birthweight include shoulder dystocia, risk of c-section for the mother, still birth and eventually, there is the risk of developing type 2 diabetes, cardiometabolic diseases, and other diseases such as musculoskeletal traits and cancers. Overall, using birthweight which is freely available to study the associated risks of diseases and health complications and use those findings for prevention is what makes this phenotype so important.

The following section addresses the complications associated with variations in birthweight.

Small-for-gestational age (SGA)

A baby is termed as small-for-gestational age (SGA) if its birthweight is less than or equal to the 10th centile in a group of other babies of the same gestational age(Saenger et al., 2007). An alternative way of defining SGA is birthweight being less than or equal to the 5th centile in a group of babies of the same gestational age(Saenger et al., 2007). Babies can be termed "constitutionally normal" infants who are SGA or those who are SGA because of growth restrictions (Osuchukwu and Reed, 2023). Constitutionally SGA infants are smaller than the 10th centile because of fundamental factors such as mother's height, weight, ethnicity, and parity, whereas infants with SGA due to growth restriction are smaller because of placental factors, infections (such as HIV) or medical conditions (such as hypertension)(Osuchukwu and Reed, 2023). There are several complications related to SGA which include stillbirth, neonatal asphyxia, hypoglycemia at birth and the risk of developing type 2 diabetes in adulthood (Osuchukwu and Reed, 2023).

Large-for-gestational age (LGA)

A baby is termed as large-for-gestational age (LGA) if its birthweight is greater than or equal to the 90th centile in a group of other babies of the same gestational age(Monari et al., 2021). There is another way of defining LGA such as birthweight being greater than or equal to the 95th centile in a group of babies of the same gestational age (Plasencia et al., 2011). This is to focus on the group with the highest risk. Similarly, to SGA, there are associated complications with infants born LGA, for example, higher risk of shoulder dystocia and associated brachial plexus injury, perinatal asphyxia, meconium aspiration, hypoglycemia and fetal death(Weissmann-Brenner et al., 2012). Later in life, babies born with LGA are at higher risk of becoming obese or developing type 2 diabetes(Scifres, 2021). For the mother, the associated risks include prolonged labor, caesarean delivery, and coronary heart disease (Boyd et al., 1983).

Cardiovascular risks in later life

There are increased cardiovascular risks in later life, associated with being born either SGA or LGA. Cardiovascular diseases are a group of common and often preventable diseases which affect the cardiovascular system, that is, the heart and circulation of blood and therefore, metabolic health. While SGA is more related to cardiovascular risk later in life, the risk with LGA is not entirely defined (Dong et al., 2018), (Renom Espineira et al., 2011). SGA infants have more catching up to do while LGA infants have catching down to do in terms of growth in the early phase of life. The intensity of this offsetting growth can cause negative health outcomes as early growth patterns highly influence cardiovascular health(Nordman et al., 2020). The “rapid catch-up” growth hypothesis states that SGA born infants who experience rapid catch-up growth develop higher risk of cardiovascular diseases later in life (Cauzzo et al., 2023). Another study investigated the predictors of catch-up growth (Ong et al., 2000) and showed that infants who showed catch-up growth were smaller and thinner at birth and became heavier than other children at five years of age. The study concluded that factors affecting catch-up growth could, therefore, be related to the underlying mechanisms of the fetal origin’s hypothesis. This hypothesis builds on the fact that undernutrition in the womb results in improper fetal growth and this makes the baby more susceptible to certain diseases (for example, cardiovascular diseases) in adulthood(Morley, 2006). This, therefore, shows a causal relationship between birthweight and the development of cardiovascular diseases. Another study showed that insufficient “catch-down” growth leads to higher chances of being overweight and consequently this increases chances of cardiometabolic problems in both childhood and adulthood(Renom Espineira et al., 2011). Babies born SGA and LGA are at higher risk of being overweight and obese throughout childhood and adulthood, having alterations in glucose metabolism, developing dyslipidemia, hypertension, and low-grade inflammation which all negatively affect cardiometabolic health(Belbasis et al., 2016).

Type 2 Diabetes

Previous research showed that there is an association between low birth weight and an increased risk of type 2 diabetes (T2D)(Barker and Osmond, 1986). The same study has shown a similar association between high birth weight and an increased risk in T2D (Wei et al., 2003). Initially, the “small baby syndrome hypothesis” suggested an inverse linear relationship between birthweight and type 2 diabetes (Harder et al., 2007). Conducted meta-analyses then showed a U-shaped relationship between the risk of developing T2D and birthweight, which implies that it is at higher risk of developing T2D with both low and high birthweight and less likely to develop it within what is assumed to be a normal range for birthweight (2,500–4,000 g)(Pettitt and Jovanovic, 2001). Finally, it was found that the relationship is actually inverse J-Shaped (Mi et al., 2017).

Current research is still trying to pin down the underlying mechanisms between birthweight and the risk of T2D. Theories of malnutrition in the perinatal period have explained neonatal overfeeding for low offspring birthweight which could lead to neonatal weight gain which then, in turn relates to adult overweight and diabetogenic disturbances throughout life (Mi et al., 2017).

In terms of lower birthweight, this could also relate to the fetal insulin hypothesis. The hypothesis states that the relationship between low birthweight and adult insulin resistance is genetically mediated, that is, low birthweight and the onset of adult- T2D are two phenotypes from the same genotype (Hattersley and Tooke, 1999). This then leads to low insulin mediated fetal growth in the utero and insulin resistance in both childhood and adulthood, which causes low birthweight. Insulin resistance refers to the fact that the human pancreas tends to produce more insulin to help the uptake of glucose into the cells when the cells are not able to absorb glucose on their own. Hence, glucose intolerance and diabetes are results of the same insulin-resistant genotype. The decreased ability to secrete and respond to insulin then results in higher risk of adult-onset T2D. Recent genetic and epidemiological studies have found evidence to support this hypothesis: heterozygous mutation in the glucokinase gene (GCK) causing MODY has been able to explore how the fetal genotype determines insulin-mediated growth in the utero, single genes (for example, INS, INSR)

have been found to affect insulin secretion (Garin et al., 2010) (Krook et al., 1993) and loci that are associated with risk of T2D are also associated with lower birthweight and some of the loci that showed very strong associations (for example, ADCY5 and CDKAL1) also affect pancreatic beta cell function (Mahajan et al., 2018).

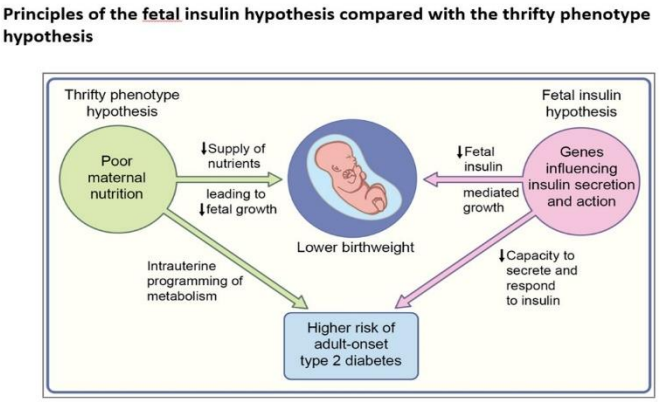

Figure 1: Principles of the fetal insulin hypothesis compared with the thrifty phenotype hypothesis

Another interesting hypothesis is that of the “thrifty genotype hypothesis” (see **Figure 1(Hughes et al., 2021))**, now termed as Developmental Origins of Health and Disease (DOHaD), which states that poor maternal nutrition results in the decrease in supply of nutrients, leading to a decrease in fetal growth. Poor maternal nutrition also contributes to intrauterine programming of a decreased metabolism. The baby is then susceptible to higher risk of T2D. The evidence for this hypothesis has been backed by a study of monozygotic twins in Denmark where the diabetic twin had a lower birthweight compared to the normoglycemic co-twin and assuming the twins are genetically identical, the difference in birthweight is explained then by the maternal intrauterine environment (Martin-Gronert and Ozanne, 2010). Another study compared individuals born before a period of famine in the Netherlands to those born after the famine (who were in utero during the famine) and it showed that poor maternal nutrition during the famine resulted in those born after to have higher glucose levels and became obese in adulthood, supporting the statement that

poor maternal nutrition results in the baby being susceptible to T2D or obesity in adulthood(Martin-Gronert and Ozanne, 2010).

Research has also shown that a high birthweight is strongly associated with risk of developing T2D. This association could be due to perinatal "malprogramming" and the presence of maternal hyperglycemia (which leads to an increase in fetal insulin levels) and fetal hyperinsulinism which are both causes of fetal macrosomia (Harder et al., 2007). Maternal hyperglycemia is caused by the mother not being able to produce enough insulin due to impairments in β-cell function in producing insulin(Brown and Walker, 2016). Since the offspring and mother share the same genotype, the baby is also then unable to produce enough insulin which results in the development of T2D in adulthood. The associations of birthweight with risk of T2D are various and another reason for studying birthweight is to further investigate this relationship and treating T2D.

Other health issues

Other than SGA, LGA and cardiometabolic diseases, there are other health issues that are associated with the baby's birthweight. The maternal intrauterine environment is related to diseases in early life and disease traits in later adult life. The relationship between birthweight and several diseases have been extensively studied: anthropometry and metabolic diseases, cancers, respiratory diseases, perinatal outcomes, and musculoskeletal traits. A comprehensive assessment of 78 associations between birthweight and diverse health outcomes (Belbasis et al., 2016) has shown that there exists convincing evidence between associations of low birthweight and increased risk for all-cause mortality, per 1 kg increase in birthweight.

1.3 What influences birth weight?

Birthweight is influenced by a lot of factors including maternal intrauterine environment, environmental factors, genetics, parity, ethnicity, and race (Wilcox, 2001).

Maternal intrauterine environment and maternal glucose

The maternal intrauterine environment can be seen as a site where the maternal signals and that of the growing fetus converge. This site plays a significant role in maintaining healthy fetal growth as this is where the fetus gets its nutrition from (Masiakwala et al., 2023). To ensure appropriate fetal growth trajectory, the placenta adapts itself to normal or modified intrauterine environments (Hughes et al., 2023, Cetin et al., 2013). The maternal nutritional status influences fetal growth primarily by the transmission of glucose and nutrients to the fetus via the placenta. Although insulin does not cross the placenta, maternal glucose is directly transmitted via the placenta and in cases where the mother is diabetic or has gestational diabetes, a high glucose level incites insulin secretion from the pancreas of the fetus to regulate its blood glucose levels. A recent monogenic study has shown that insulin-mediated fetal growth accounts for half of offspring birthweight at term(Hughes et al., 2023). The intrauterine environment is also important in terms of complications associated with birthweight such as intrauterine growth restrictions. In such cases, the inhibition of pancreatic β-cell replication results in reduced fetal insulin production and affects fetal growth. Although this has not been properly defined before but debates about adverse intrauterine environments causally increasing the risk of future cardiometabolic diseases have also been raised (Masiakwala et al., 2023, Hughes et al., 2023, Cetin et al., 2013).

Environmental factors

Environmental influences have been found to contribute to explaining about 25% of offspring birthweight (Johnston et al., 2002). There are also several environmental factors that can cause adverse pregnancy outcomes; exposure to air pollution, general as well as physical stress. Socioeconomic and demographic factors influence the environment a person is surrounded with, and this is also a risk factor for birthweight (Cramer, 1995). While video display terminals, metals and aesthetic gases are associated with the risk of congenital defects, childhood cancer, spontaneous abortion, and infertility, respectively, the most influential environmental factor has been found to be the mother's smoking status during pregnancy (Triche and Hossain, 2007). Previous studies

(Kataoka et al., 2018) have shown an increasing risk of low birthweight with smoking during pregnancy, the possibility of intrauterine growth restriction, delivery of preterm infants, the need for instantaneous abortion and problems with the placenta. Smoking during pregnancy affects placental function and affects fetal development by crossing the placenta to act as neuroteratogen.

Genetics

Genetics plays a crucial role in determining a child's birthweight. A baby inherits half of its genes from its mother and the other half from its father to build its own genetic makeup. To identify associations between a trait and variants, genome wide association studies (GWAS) have been devised. Recent GWAS of own birthweight and offspring birthweight have identified 243 links existing between the human genetic code and birthweight (Warrington et al., 2019) where the fetal genotype has a direct effect on a child's birthweight and the maternal genotype affects a baby's birthweight via the maternal intrauterine environment.

Genome Wide Association Studies (GWAS)

A genome-wide association study (GWAS) aims at identifying associations between a trait and specific loci on the genome (Uffelmann et al., 2023); it is a way of finding how genetics is linked to specific phenotypes. A meta-analysis of a multi-ancestry GWAS for birthweight, identified 60 fetal genotype loci related to birthweight ($P < 5 \times 10^{-8}$). The threshold for genome-wide significance was determined as $P < 5 \times 10^{-8}$ previously but more recent studies use $P < 6.6 \times 10^{-9}$ as a new threshold(Warrington et al., 2019). The study comprised of the following ancestries from UK Biobank: European, African American, Chinese, Filipino, Surinamese, Turkish and Moroccan from six studies. The study also found that there is a genetic association between fetal growth and later-life diseases which influences variation on both the maternal and fetal genomes. The study found these birthweight related loci to be associated with SBP, cardiometabolic traits and T2D (Juliusdottir et al., 2021).

Maternal genetics

The maternal genome contributes to half a baby's birthweight via the maternal intrauterine environment and the rest is passed on directly to the fetus. A study has found that approximately 7.6% of offspring birthweight variation can be explained by tagged maternal genetic variation (Warrington et al., 2019). Previous research has shown that there is a strong, significant association between the maternal genetic variant and offspring birthweight (after taking the fetal genotype into consideration)(Juliusdottir et al., 2021) and this adds to the proof that maternal exposure is causally related to birthweight. Mendelian randomization analyses also show that indirect maternal effects of height-raising genotypes contribute to higher offspring birthweight (Warrington et al., 2019). Also, the maternal genome positively adds to birthweight through variants related to glycemic traits such as T2D, HbA1C and glucose (Juliusdottir et al., 2021, Haulder et al., 2022). In conclusion, maternal genetics has a significant contribution to birthweight.

Fetal genetics

Direct effects from the fetal genome have a substantial contribution to birthweight; height-raising alleles contribute to higher offspring birthweight and blood-pressure raising alleles contribute to lowering birthweight through the fetal genome (Warrington et al., 2019). Fetal genetics has also been found to explain variation in birthweight on top of clinical features and about 146 direct links have been established (Warrington et al., 2019) between the fetal genome and offspring birthweight in a study. Other direct links exist between the fetal genome and birthweight, for example in rare fetal mutations which cause neonatal diabetes (Rubio-Cabezas and Ellard, 2013). A study estimating SNP heritability called maternal-genome wide complex trait analysis (M-GCTA) and divides SNP heritability distinctively into maternal and fetal components has estimated that about 28.5 % of the variance in birthweight can be explained by tagged fetal genetic variation(Warrington et al., 2019).

Fetal Insulin

Another particularly important determinant of birthweight remains fetal insulin. The fetal insulin hypothesis was put forward in 1999 (Hattersley and Tooke, 1999) and it stated that lower birthweight and adult-onset type 2 diabetes are two phenotypes of the same genotype. Research following this hypothesis has now shown that there are several genes affecting insulin secretion that induce insulin-mediated growth (Hattersley et al., 1998, Flanagan et al., 2007, Garin et al., 2010). This is important because the production of fetal insulin regulates the uptake of maternal glucose from the placenta by the fetus in the maternal intrauterine environment. Variants of certain genes such as glucokinase gene (GCK-MODY) and HNF4A-MODY result in impaired pancreatic beta cell function causing either reduced insulin secretion which in turn leads to lower birthweight or increased insulin secretion which results in increased birthweight (neonatal hyperinsulinism and macrosomia) (Hughes et al., 2021). Results have shown that GCK mutations in beta-cells could alter insulin response to glucose and thus, influence intrauterine growth as well as glucose metabolism after birth(Terauchi et al., 2000). In the case of individuals with the HNF4A-MODY, a higher inherited birthweight suggests the greater capacity of the individual to secrete insulin. Overall, fetal insulin plays a crucial role in fetal growth in the maternal intrauterine environment and therefore primarily influences birthweight.

Parity

In the UK, parity refers to the number of times a mother has previously given birth to a fetus with a gestational age of 24 weeks or more, regardless of whether the child was born alive or stillborn (Opara and Zaidi, 2007)and nulliparity refers to the condition in which a mother has never given birth before (Miranda et al., 2011). Birthweight and parity are inversely related with nulliparous women delivering babies of lower birthweight and birthweight increasing in subsequent pregnancies with multiparous women. A study (Hinkle et al., 2014) has assessed the relationship between parity and birthweight reporting selection bias as an explanation for low birthweight in nulliparous women who do not have any other children. There is also the possible explanation in physiological conditions associated with the body where the first

pregnancy prepares the body for any subsequent ones and hence, the mother's body adapts itself for successive pregnancies and birthweight changes or first offspring have an immune intrauterine environment which restricted growth, and this changed with subsequent pregnancies and consistent paternity (Hinkle et al., 2014).

Ethnicity and race

There is no defined pattern between birthweight and ethnicity, but this trait varies across various ethnicities where the lowest birthweights were recorded in Africa, India and the Far East and the highest birthweights were recorded across Europeans and White Americans (Barron, 1983). In the UK, birthweights vary across the different ethnicities: South Asian and Black mothers deliver babies whose birthweights are on average 300g less than those of White mothers. These differences in birthweights could firstly be explained by the differences in maternal characteristics such as height, weight, and parity between mothers of different ancestries. South Asian and Black women are on average shorter and of lower weight as compared to women of White ancestry. As such, the maternal intrauterine environment in the South Asian and Black ethnicity is adapted to reproduce babies that are smaller than those from a White ancestry. A study found that differences in birthweight in the ethnic groups of Black Caribbean, Black African, Bangladeshi, and Pakistani infants could be explained by socioeconomic differences and in Indian and Bangladeshi groups, maternal and infant characteristics were more prevalent (Kelly et al., 2009). Ethnic groups such as Black Carribean, Black African, Bangladeshi, and Pakistani are often subjected to socioeconomic disparities such as lower income. Since income is directly relevant to determining nutrition and prenatal care, it then has a significant influence on birthweight (Cramer, 1995).

1.4 Why do we want to predict birthweight?

Epidemiological research has established birthweight as an important phenotype and in current clinical practice, fetal growth is monitored by ultrasound scans, which while providing valuable information, does not give us

the most accurate estimate of birthweight due to the lack of accuracy in calculating certain variables such as the estimated fetal weight (EFW)(Milner and Arezina, 2018). Thus, to get a more precise estimate of a baby's birthweight, we want to predict it and thus cater for antenatal and postnatal care.

1.5 What do we know about clinical prediction models for birthweight and LGA?

Several clinical prediction models have been developed to predict LGA (Meertens et al.). The use of a clinical prediction model ensures that a combination of predictors is used together to model risk of LGA or birthweight. If used on their own, the amount of information provided by the birthweight predictors would provide limited information or not account for correlations among the predictors. In previous research, clinical prediction models for LGA have used maternal characteristics (maternal height, weight, age and smoking status) as their main predictors and in addition to these, other predictors such as medical history, biomarkers, ultrasound predictors and ethnicity have been used. While a lot of these models are useful in providing estimates of the risk of a baby being born LGA, none of these models are currently used in clinical practice to predict the risk of LGA(Meertens et al., 2019).

1.6 What are the individual predictors of birthweight and LGA?

There are many predictors of birthweight, the main ones being maternal characteristics such as maternal pre-pregnancy BMI and maternal BMI, maternal weight, height, age, ethnicity, parity and smoking status during the pregnancy. Other external factors include father's height (Zhang et al., 2015), for example. Maternal fasting glucose is also a main factor in predicting birthweight (Mi et al. 2023). There are several causes of LGA including weight gain during pregnancy, maternal diabetes, race and ethnicity. In addition, other markers such as parity, biomarkers (PAPP-A and β-hCG), fetal biometry (Amark et al., 2019) and maternal and paternal height(Takagi et al., 2019) are all

predictors of LGA.

Mother's height and Father's height

Maternal height is directly related to offspring birthweight where an increase in maternal height results in an increase in birthweight(Zhang et al., 2015). The underlying mechanisms that relate maternal height to birthweight are defined by fetal genetics. Data that includes paternal height is very limited and it has been found that paternal height is a significant and independent predictor of low birthweight in offspring (Magnus et al., 2001).

Mother's weight (pre-pregnancy weight and weight at 28 week's gestation)

Pre-pregnancy weight is defined as the mother's weight before conception. Mother's pre-pregnancy weight and weight at 28 weeks' gestation are both strongly associated with birthweight (Metgud et al., 2012).

Mother's BMI (pre-pregnancy and at 28 weeks' gestation)

BMI (Body Mass Index) is a metric that is based on a person's height and weight and is used to measure body fat (Rothman, 2008). Similarly, to mother's weight and pre-pregnancy weight, mother's BMI and pre-pregnancy BMI are strongly and positively associated with baby's birthweight. (Gul et al., 2020, Bahrami Taghanaki et al., 2016)

Fetal Sex

Fetal sex is an important component in pregnancy that influences intrauterine growth, placentation and perinatal outcomes. Studies have reported a difference in birthweight based on fetal sex with average birthweight in males being 124 g (95% CI 122-126) higher than in females (3525 vs 3401 g; $P < 0.001$) (Voskamp et al., 2020). Fetal sex effects, for example, maternal glucose metabolism and insulin sensitivity as well as differential placentation probably due to imprinting of placental genes during pregnancy may be a factor in the observed differences in birthweight (Poyrazoglu et al., 2017, Hattersley and Tooke, 1999, Hughes et al., 2021, Terauchi et al., 2000).

What is missing from current clinical prediction models?

Clinical prediction models have used maternal characteristics, ultrasound scans, medical history and biomarkers as predictors of birthweight(Meertens et al., 2019). Maternal fasting glucose has not been used before as a predictor for LGA, when modeling healthy pregnant women but it has been used as a predictor in women with gestational diabetes (GDM)(Wang et al., 2023, Cooray et al., 2022). Maternal fasting glucose is an important factor that contributes to offspring birthweight and implementing this predictor in future models would help reproduce models of better quality.

Also, while it has been established that, genetics plays a crucial role in explaining offspring birthweight, the variance explained by genetics in addition to clinical risk factors has not been investigated. With GWAS' becoming easily available and hence, the creation of genetic scores becoming easy to implement, genetics and more accurately genetic scores can also be added to multivariable models explaining variation in birth weight -- to assess their potential for future use in clinical prediction. Prediction models for LGA have been built in all three trimesters with different predictors (Frick et al., 2016, Monari et al., 2021, Wang et al., 2023). In terms of a model for predicting LGA, it would be preferable to have a model that can predict LGA earlier rather than later. If a baby is estimated to be born with LGA, altering the maternal fasting glucose (Tennant et al., 2022)can help reduce the risk of LGA.

1.8 What are the aims of this PhD?

a. To compare the performance of multivariable prediction models for birthweight that include genetic scores from mother and child alongside easily available clinical features with those models that contain only easily available clinical features in a European cohort.

b. To compare the performance of multivariable prediction models for birthweight that include genetic scores from mother and child alongside easily available clinical features with those models that contain only easily available clinical features between South Asians and Europeans.

c. To develop a risk prediction tool for large-for-gestational age (LGA) babies in healthy pregnancies in a European cohort with readily available clinical features, including glucose in the normal range and to externally validate this model in a South Asian cohort.

Chapter 2
Methodology

2 Methodology

We needed to build statistical models based on data from mothers and their babies at birth and hence, determine the utility of clinical features and genetics for predicting birthweight. In this chapter, I describe the datasets we use, the methods for developing the genetic scores for birthweight, and the statistical approaches we used to develop the prediction models.

Datasets

Exeter Family Study of Childhood Health (EFSOCH)

The Exeter Family Study of Childhood Health (EFSOCH) was used as a primary dataset for building multivariable linear regression models for birthweight (Chapter 3) and then multivariable logistic regression models for prediction of LGA (Chapter 5). This dataset is a cohort of 1017 families in Exeter with homogeneous, non-diabetic participants of White European ancestry. The participants were from central Exeter. The dataset comprised of detailed anthropometric measurements for both parents and children at birth, 12 weeks, 1 year and 2 years of age. Insulin and other biochemical analysis were measured in fasting parental samples at 28 weeks' gestation and an umbilical cord blood sample was taken at delivery (Knight et al., 2006). The phenotypes to be measured were identified prior to the commencement of the study and protocols were written in terms of how to best collect data. Three part-time midwives were responsible for data collection and repeat measures were taken for the phenotypes to ensure reproducibility. Some of the key phenotypes available in the original dataset that we used in this research project were parental heights, which were measured three times, and an average value was then computed for use in analysis. Similarly, other phenotypes were measured thrice and an average was computed: mother's weight was measured to the nearest 100g, offspring birthweight was measured to the nearest 10g, and parental birthweights were self-reported. Mother's fasting glucose was recorded from fasting blood samples taken at 28 weeks' gestation, early morning, at the parent's house. Parity (recorded as first pregnancy vs not first pregnancy), mother's age, and smoking status (recorded as mother smoked during pregnancy or did not smoke during pregnancy) were also self-reported.

Gestational age was calculated based on the mother's last menstrual period and the date of birth. The difference between the two dates gave an accurate measurement of the gestational period(Knight et al., 2006).

Collection and cleaning of additional data from medical records

We wanted to explore the contributions of blood pressure and fetal biometry to LGA (Chapter 5), and at the start of this PhD, the raw data was only available in EFSOCH medical records. To collect the data, I worked with a team of other researchers to manually extract the systolic blood pressure (SBP) and diastolic blood pressure (DBP) measurements including timings when these were recorded, from patient notes for EFSOCH and stored in an online database created in Access. Data was collected for 931 unique mothers with an average of 20 readings per observation.

The blood pressure measurements had been noted at different time points when the mother visited the clinic during her pregnancy (Minium 5 observations and Maximum 20 observations). Missing blood pressure values were removed if not eligible (any measurements higher than 120 for the upper limit and any measurements less than 50 for the lower limit). Wrongly written EFSOCH IDs, which are the main identifiers in this database, were corrected. These were IDs with a missing digit. Similarly, wrongly written dates were also corrected by referring to the original patient notes and noting them again. These were identified by a negative gestational age or a gestational age that would be realistically too long. The data was also checked for outliers by plotting two phenotypes against each other and then removing any extreme values. Other checks included generation of box plots to identify outliers.

Scan data from maternity records for EFSOCH were recorded for biparietal diameter (BPD), Femur Length (FL), Crown Rump Length (CRL), Head Circumference (HC), Abdominal Circumference (AC) and Estimated Fetal Weight (EFW). The scan measure for EFW was at 20 weeks' gestation and the rest were at 12 weeks' gestation. Similar to blood pressure measurements, wrongly written dates and EFSOCH IDs were corrected for the observations

that had a missing or additional digit and the data was checked for outliers and those were then removed.

BiB (Born in Bradford)

The Born in Bradford (BiB) dataset is a deprived multi-ethnic cohort study comprising of White Europeans and South Asians based in Bradford in the UK. This dataset was used to make comparisons between birthweights, other phenotypes, and genetic scores between the two different ancestries (Chapter 4) and in the external validation of the prediction model for LGA (Chapter 5). The dataset consists of 13776 pregnancies which were followed up after these were recruited when they were between 26- and 28-weeks' gestation. The maternal fasting glucose was recorded at 26-28 weeks' gestation after an OGTT test and mother's height, and weight were measured after their interview which was offered alongside the OGTT test. Other anthropometric measurements such as mother's arm circumference and triceps skinfold were also taken from the mother by a trained project worker at recruitment. Anthropometric measurements such as head, arm and abdominal circumference measured, along with subscapular and triceps skinfold thickness were taken from infants at birth and followed up to 2 years of age. To ensure reliability of the measurements, the same phenotype was measured twice by the same trained worker or two separate measurements were taken for the same phenotype by two different trained workers and an average computed. About 20% of the population of parents in Bradford are of South Asian ancestry (Wright et al., 2013). Birthweight was measured at birth in g and ethnicity, smoking status as well as parity were all self-reported measures (West et al., 2013).

Growth charts

In this section, we want to address how the centiles were used to create the LGA variable in Chapter 5. A growth chart is built on the idea that assessing a

child's height or weight against the distribution of heights or weights of a reference sample provides evidence of the normality or otherwise of the process of growth (Cameron and Hawley, 2010).

In the UK, the most used growth charts are the UK 1990's and the UKWHOterm and these were used in this thesis. Initially, the UK1990's growth chart was used to create the LGA variable in the EFSOCH dataset and the UKWHOterm growth chart was used to create the LGA variable in the BiB dataset.

However, because the baseline risk was different when these two growth charts were being used, external validation was not giving us a good mean predictive probability close to the actual observed mean probability. Thus, we decided to use the UKWHOterm growth chart uniformly. This growth chart was then used to create the LGA variable in both EFSOCH and BiB. The package "zanthro" was used to generate the z-scores for birthweight by using the UKWHOterm growth charts in EFSOCH and LGA was defined as that group with birthweight >90th centile(Vidmar et al., 2013).

The UKWHOterm growth chart is constructed on WHO Child Growth Standards which is based on the breastfed infant. One of the disadvantages of using this growth standard to define LGA is the assumption that all assessed infants have been breastfed, which is not always the case, especially in UK-born infants. Also, defining LGA by a single international standard can result in inaccuracies as what is considered as LGA in one ethnicity, for example in South Asians, is not necessarily LGA in another ethnicity, for example in Europeans. Also, by defining LGA based on a specific growth chart, it should be taken into consideration that several changes could have occurred from the year the growth chart was construed to when it is being used. The UK 1990's growth chart was devised in 1990 and people's eating habits have considerably changed since then. Hence, defining LGA based on a similar growth standard for a cohort of babies born outside this time period might not accurately reflect the real percentage of LGA for that cohort.

Creating genetic scores

An important part of this research investigates the contribution of genetics in explaining variation to birthweight. To include genetics in multivariable linear regression models or multivariable logistic regression models, genetic scores were created for both the mother and the child.

Genetic Scores (GS) were created using a weighted formula which averages all the SNP's that have crossed the threshold for genome wide significance ($P<6.6x10^{-9}$)(Warrington et al., 2019) (Fadista et al., 2016)over a weighted average. This can be represented by the following formula (Collister et al., 2022):

$$GS=\sum_i w_i g_i$$

where GS is the genetic score, wi is the weight for SNP i and gi is the genotype dosage at SNP i. The weights in this case were the effect estimates for each SNP from the GWAS.

There were two sets of data from which genetic scores were created and these were the genetic data from EFSOCH and BiB. In EFSOCH, genotyping had been carried out using the Illumina HumanCoreExome array. This is a tool for assessing the genotypes of approximately 500,000 variants across the genome from a single DNA sample. Samples with low call rate, kinship errors, sex mismatches, or ancestry outliers were removed. The included samples were of European ancestry assessed using flashPCA. flashPCA (Abraham and Inouye, 2014)is a tool used to conduct principal component analysis (PCA) for identification of genome-wide single-nucleotide polymorphism (SNP) data for detecting population structure and potential outliers. Genotype call rates were > 98% and phenotypic sex and kinship were validated using genotype data (Hughes et al., 2018) assessed by KING (Manichaikul et al., 2010)software.

The included genotyped SNPs had call rates > 95%, Hardy-Weinberg p $> 1 \times 10^{-6,}$ and minor allele frequency (MAF) > 1%. The Haplotype Reference Consortium (HRC) version r1.1 reference panel (Michigan Imputation Server) was used to impute additional genotypes in all samples. We extracted genotypes for a total of 209 SNPs (Warrington et al., 2019)from the genome-wide genotype data to construct genetic scores for our analyses. A total of 98%

of the SNPs included in the scores had an imputation quality > 0.4 and a Minor Allele Frequency > 0.001.

In BiB, two separate microarrays, the Illumina HumanCoreExome array and the Infinium Global Screening array (GSA), were used to obtain maternal and fetal genomic data. Genotype data was imputed against the Haplotype Reference Consortium panel, version r1.1 using Minimac4, after quality control (minor allele frequency > 1% and Hardy-Weinberg Equilibrium P value > $1 \times 10^{-6)}$.(Fadista et al., 2016).

Prediction Models

Prediction models are extremely useful for estimating the probability of an outcome by using a set of predictor variables. In clinical practice, prediction models are used to assess the risk of an outcome (in this case, occurrence of a disease) given a set of patient characteristics (in this case, the predictors). These are an easy and affordable way of assessing risk and hence, inform diagnosis or prognosis in healthcare (Steyerberg, 2010). Common clinical prediction models include models for predicting SGA (Kalafat et al., 2019), LGA(Monari et al., 2021), the Framingham risk functions for cardiovascular disease (Menotti et al., 2000), spontaneous pregnancy chances(Smeenk et al., 2007), probability of renal artery stenosis (Lee et al., 2014) or deep venous thrombosis. Prediction models have also been developed for the risk of breast cancer and to inform surgical decision-making such as replacement of risky heart valves (Ambler et al., 2005). There are more uses of prediction models, including in public health (to target preventive interventions) and in research (such as development of RCTs). A project carried out for part of this thesis will focus on clinical prediction model for predicting risk of LGA.

Model Building

There are several steps in building a clinical prediction model and these are: consideration of the research question and initial data inspection, coding of predictors, model specification, model estimation, evaluation of model

performance, internal validation and external validation. Finally, to report clinical prediction models, TRIPOD (Transparent reporting of a multivariable prediction model for individual prognosis or diagnosis) reporting guidelines are used (Collins et al., 2015).

Consideration of the research question and initial data inspection

The research question is devised by considering what message we want to put across, and then the predictors are coded if these are not continuous. Smoking was a binary variable not a past/present; primiparity was first pregnancy vs not first baby.

Model Specification

Model specification includes choosing the type of model to be used. In this thesis, we used two main types of regression analyses to build our prediction models: linear regression and logistic regression. Other methods that are also used to build clinical prediction models are ordinal regression analysis (that is, the outcome variable consists of an ordinal response variable), survival analysis, (that is, the modelling of time to event data) (Harrell et al., 1984) and machine learning approaches(Sun et al., 2022). Regression analysis is used to assess the relationship between variables. This statistical tool can be used as a simple regression model or a multivariable regression model. In linear regression, the outcome variable is a continuous variable, for example, birthweight and in logistic regression, the outcome variable is binary, for example, baby is LGA or not LGA. In a simple linear regression model, the effect of one covariate is being assessed on the continuous outcome variable and in a multivariable regression model, the combined effects of all the covariates are being assessed on the outcome variable(Sykes, 1993). The model returns the intercept and coefficients representing the weightings for the slope of a linear association. For a logistic regression model, because the outcome is binary the coefficients are for predicting the log odds ratio of the outcome instead (because the association with log odds can be linear). The next steps in model building are variable selection, checking of assumptions, developing the model, internal and external validation.

Variable selection

This is an important part of building a clinical prediction model as it assesses which variables to include, and which ones are irrelevant so that a set of variables is chosen to build the model with the best fit. Some of the main methods include backward elimination, forward selection, and stepwise selection. In this case, we used backward selection to select the optimal set of variables. This implies adding all the variables in a model and then removing the one with the highest p-value (that is, the variable which is not contributing to the model). This is repeated until the model includes all the significant variables (Chowdhury and Turin, 2020).

Checking of assumptions

There are several model assumptions that need to be verified depending on the type of outcome variable and the model type. In linear regression as well as logistic regression, there should be no multicollinearity between the variables, and this is ensured by checking that the correlation coefficients between individual predictors are of low magnitude. In linear regression, the QQ plot of residuals can be used to check the normality assumption. If most of the points fall on the reference line, normality can be assumed. Also, to check for homoscedasticity, the residuals can be plotted. If no linear pattern is observed, we conclude that there is no heteroscedasticity. The sample size (Riley et al., 2020) should meet a certain number depending on several factors in the model and this is verified by using the “psampsize” function in R, for example, by specifying the prevalence of the outcome, the number of predictors to be used in the model and a predicted minimum c-statistic. Additionally, we want to ensure that the observations are independent of one another, and this is achieved by removing duplicate observations and any observations from matched data. Finally, to meet the assumption of the outcome variable being binary, we denote it as baby being LGA (1) or not (0).

Assessing model performance

Once the variables have been selected and the model has been built, it is also important to assess how well the model performs. Measures of model performance include model fit, discrimination and calibration (Labarere et al., 2014). Model fit statistics include R-squared and adjusted R-squared statistics, or the Brier score. Usually, the model that explains the most variance will be chosen as the best one (that is, the model with the highest adjusted R-squared statistic). The adjusted R-squared statistic is an estimate built on the natural logarithm likelihood scale and the higher the value, the better is the fit. The Brier score quantifies the average squared difference between the observed outcome and observed probability and the closer the value is to zero, the better the model fit is. Other useful statistics are the Akaike information criterion (AIC) and Bayesian information criterion (BIC). These are log-likelihood based criteria that include a penalty for additional predictors (BIC uses a bigger penalty) and are used to check for overfitting. Therefore, AIC and BIC can be used to compare nested models to find the best model with the smallest predictor set. The lower the value of AIC or BIC, the better the model fit.

Discrimination is the model's ability to categorize individuals with and without the outcome of interest. The c-statistic is a good measure of discrimination and is identical to the area under the receiver operating characteristic (ROC) curve. A value of 0.5 indicates that the model is as good as a random prediction but the closer the value of the c-statistic is to 1, the better is the discrimination between individuals with and without the outcome.

Calibration refers to how close the actual observed probability for the outcome of interest is to the predicted mean probability. This can be assessed by the visual representation of observed probability versus predicted probability (calibration plots). Assessment of calibration includes the assessment of a modelling regression line with intercept (alpha) and slope (beta). For well calibrated models, the intercept is usually zero or close to zero and the slope is 1 or close to 1. The Hosmer-Lemeshow test, while a goodness of fit test, might not be statistically strong enough to assess good calibration (Labarere et al., 2014).

Internal Validation

Internal validation is used to validate the model that has been built in the same data set it was constructed in. Measures of internal validation include split-sample, cross-validation and bootstrapping. In this thesis, we have explored bootstrapping as a measure of internal validation as it has been found to be the best measure of internal validation (Steyerberg et al., 2001). Bootstrapping is a method where new samples are drawn from the main sample, with replacement. In each of these resampled cohorts, the model performance is tested, and these results are pooled to determine internal validation performance(Singh and Xie, 2008). Some statistics such as Emax or the Somer's Dxy can be used to assess optimism due to overfitting. Low levels of optimism indicate good model fit. Overfitting refers to the fact that predictions for new observations might not be valid which then causes optimism about the model's performance. Optimism can, therefore, be described as true performance minus apparent performance, where true performance is the actual population and apparent performance is the estimated performance in the sample(Steyerberg, 2019).

External Validation

External validation is an important step in model building in terms of validating a model in an external data set for generalizability. The ROC curve and calibration curve are the measures of external validation for assessing model performance in an external dataset. Compared to internal validation which assesses the model performance in a similar setting in which the model was built, external validation considers analysis in a completely independent dataset which could include patients having one or more different characteristics for generalizability. For example, patients may be from a different geographical location, from a different type of care setting or have a different underlying disease (Ramspek et al., 2021).

The ROC curve assesses sensitivity versus specificity, that is, true positive rate versus true negative rate. A value close to 1 for area under the curve of an ROC curve shows that the model is performing well in terms of differentiating true positives from false positives (Hajian-Tilaki, 2013). The calibration curve assesses how well the mean probability of the outcome occurring within specific

groups matches the actual observed probability occurring within those specific groups.

The next section of this thesis will expand more into the individual chapters: assessing whether genetic scores explain extra variation in birthweight, when added to clinical and anthropometric measures, the contributions of genetic scores, maternal glycemia, and other maternal characteristics to variation in birth weight in South Asian compared with European babies and developing and validating a multivariable clinical prediction model for babies born LGA in European and South Asian cohorts.

References

ABRAHAM, G. & INOUYE, M. 2014. Fast principal component analysis of large-scale genome-wide data. *PLoS One,* 9**,** e93766.

AMBLER, G., OMAR, R. Z., ROYSTON, P., KINSMAN, R., KEOGH, B. E. & TAYLOR, K. M. 2005. Generic, simple risk stratification model for heart valve surgery. *Circulation,* 112**,** 224-31.

BAHRAMI TAGHANAKI, H., HASHEMIAN, M., LOTFALIZADEH, M. & NORAS, M. 2016. The relationship between Body Mass Index (BMI) and birth weight and some pregnancy outcomes. *The Iranian Journal of Obstetrics, Gynecology and Infertility,* 19**,** 1-8.

CAMERON, N. & HAWLEY, N. L. 2010. Should the UK use WHO growth charts? *Paediatrics and Child Health,* 20**,** 151-156.

CHOWDHURY, M. Z. I. & TURIN, T. C. 2020. Variable selection strategies and its importance in clinical prediction modelling. *Fam Med Community Health,* 8**,** e000262.

COLLINS, G. S., REITSMA, J. B., ALTMAN, D. G. & MOONS, K. G. 2015. Transparent reporting of a multivariable prediction model for individual prognosis or diagnosis (TRIPOD): the TRIPOD statement. *BMJ,* 350**,** g7594.

COLLISTER, J. A., LIU, X. & CLIFTON, L. 2022. Calculating Polygenic Risk Scores (PRS) in UK Biobank: A Practical Guide for Epidemiologists. *Front Genet,* 13**,** 818574.

COORAY, S. D., BOYLE, J. A., SOLDATOS, G., ALLOTEY, J., WANG, H., FERNANDEZ-FELIX, B. M., ZAMORA, J., THANGARATINAM, S. & TEEDE, H. J. 2022. Development, validation and clinical utility of a risk prediction model for adverse pregnancy outcomes in women with gestational diabetes: The PeRSonal GDM model. *EClinicalMedicine,* 52**,** 101637.

FADISTA, J., MANNING, A. K., FLOREZ, J. C. & GROOP, L. 2016. The (in)famous GWAS P-value threshold revisited and updated for low-frequency variants. *Eur J Hum Genet,* 24**,** 1202-5.

FRICK, A. P., SYNGELAKI, A., ZHENG, M., POON, L. C. & NICOLAIDES, K. H. 2016. Prediction of large-for-gestational-age neonates: screening by maternal factors and biomarkers in the three trimesters of pregnancy. *Ultrasound Obstet Gynecol,* 47**,** 332-9.

GUL, R., IQBAL, S., ANWAR, Z., AHDI, S. G., ALI, S. H. & PIRZADA, S. 2020. Pre-pregnancy maternal BMI as predictor of neonatal birth weight. *PLoS One,* 15**,** e0240748.

HAJIAN-TILAKI, K. 2013. Receiver Operating Characteristic (ROC) Curve Analysis for Medical Diagnostic Test Evaluation. *Caspian J Intern Med,* 4**,** 627-35.

HARRELL, F. E., JR., LEE, K. L., CALIFF, R. M., PRYOR, D. B. & ROSATI, R. A. 1984. Regression modelling strategies for improved prognostic prediction. *Stat Med,* 3**,** 143-52.

HATTERSLEY, A. T. & TOOKE, J. E. 1999. The fetal insulin hypothesis: an alternative explanation of the association of low birthweight with diabetes and vascular disease. *Lancet,* 353**,** 1789-92.

HUGHES, A. E., HATTERSLEY, A. T., FLANAGAN, S. E. & FREATHY, R. M. 2021. Two decades since the fetal insulin hypothesis: what have we learned from genetics? *Diabetologia,* 64**,** 717-726.

HUGHES, A. E., NODZENSKI, M., BEAUMONT, R. N., TALBOT, O., SHIELDS, B. M., SCHOLTENS, D. M., KNIGHT, B. A., LOWE, W. L., JR., HATTERSLEY, A. T. & FREATHY, R. M. 2018. Fetal Genotype and Maternal Glucose Have Independent and Additive Effects on Birth Weight. *Diabetes,* 67**,** 1024-1029.

KALAFAT, E., MORALES-ROSELLO, J., THILAGANATHAN, B., DHOTHER, J. & KHALIL, A. 2019. Risk of neonatal care unit admission in small for gestational age fetuses at term: a prediction model and internal validation. *J Matern Fetal Neonatal Med,* 32**,** 2361-2368.

KNIGHT, B., SHIELDS, B. M. & HATTERSLEY, A. T. 2006. The Exeter Family Study of Childhood Health (EFSOCH): study protocol and methodology. *Paediatr Perinat Epidemiol,* 20**,** 172-9.

LABARERE, J., RENAUD, B. & FINE, M. J. 2014. How to derive and validate clinical prediction models for use in intensive care medicine. *Intensive Care Med,* 40**,** 513-27.

LEE, Y., SHIN, J. H., PARK, H. C., KIM, S. G. & CHOI, S. I. 2014. A prediction model for renal artery stenosis using carotid ultrasonography measurements in patients undergoing coronary angiography. *BMC Nephrol,* 15**,** 60.

MANICHAIKUL, A., MYCHALECKYJ, J. C., RICH, S. S., DALY, K., SALE, M. & CHEN, W. M. 2010. Robust relationship inference in genome-wide association studies. *Bioinformatics,* 26**,** 2867-73.

MENOTTI, A., PUDDU, P. E. & LANTI, M. 2000. Comparison of the Framingham risk function-based coronary chart with risk function from an Italian population study. *Eur Heart J,* 21**,** 365-70.

MONARI, F., MENICHINI, D., SPANO' BASCIO, L., GRANDI, G., BANCHELLI, F., NERI, I., D'AMICO, R. & FACCHINETTI, F. 2021. A first trimester prediction model for large for gestational age infants: a preliminary study. *BMC Pregnancy Childbirth,* 21**,** 654.

POYRAZOGLU, S., DARENDELILER, F., AHMED, S. F., HUGHES, I., BRYCE, J., JIANG, J., RODIE, M., HIORT, O., HANNEMA, S. E., BERTELLONI, S., LISA, L., GURAN, T., COOLS, M., DESLOOVERE, A., CLAAHSEN-VAN DER GRINTEN, H. L., NORDENSTROM, A., HOLTERHUS, P. M., KOHLER, B., NIEDZIELA, M. & KRONE, N. 2017. Birth Weight in Different Etiologies of Disorders of Sex Development. *J Clin Endocrinol Metab,* 102**,** 1044-1050.

RAMSPEK, C. L., JAGER, K. J., DEKKER, F. W., ZOCCALI, C. & VAN DIEPEN, M. 2021. External validation of prognostic models: what, why, how, when and where? *Clin Kidney J,* 14**,** 49-58.

RILEY, R. D., ENSOR, J., SNELL, K. I. E., HARRELL, F. E., JR., MARTIN, G. P., REITSMA, J. B., MOONS, K. G. M., COLLINS, G. & VAN SMEDEN, M. 2020. Calculating the sample size required for developing a clinical prediction model. *BMJ,* 368**,** m441.

ROTHMAN, K. J. 2008. BMI-related errors in the measurement of obesity. *Int J Obes (Lond),* 32 Suppl 3**,** S56-9.

SINGH, K. & XIE, M. 2008. Bootstrap: a statistical method. *Unpublished manuscript, Rutgers University, USA. Retrieved from http://www. stat. rutgers. edu/home/mxie/RCPapers/bootstrap. pdf***,** 1-14.

SMEENK, J. M., BRAAT, D. D., STOLWIJK, A. M. & KREMER, J. A. 2007. Pregnancy is predictable: a large-scale prospective external validation of the prediction of spontaneous pregnancy in subfertile couples. *Hum Reprod,* 22**,** 2344-5; author reply 2345-6.

STEYERBERG, E. W. 2010. *Clinical prediction models : a practical approach to development, validation and updating,* New York, Springer.

STEYERBERG, E. W. 2019. Overfitting and Optimism in Prediction Models. *Clinical Prediction Models: A Practical Approach to Development, Validation, and Updating.* Cham: Springer International Publishing.

STEYERBERG, E. W., HARRELL, F. E., JR., BORSBOOM, G. J., EIJKEMANS, M. J., VERGOUWE, Y. & HABBEMA, J. D. 2001. Internal validation of predictive models: efficiency of some procedures for logistic regression analysis. *J Clin Epidemiol,* 54**,** 774-81.

SUN, H., DEPRAETERE, K., MEESSEMAN, L., CABANILLAS SILVA, P., SZYMANOWSKY, R., FLIEGENSCHMIDT, J., HULDE, N., VON DOSSOW, V., VANBIERVLIET, M., DE BAERDEMAEKER, J., ROCCARO-WALDMEYER, D. M., STIEG, J., DOMINGUEZ HIDALGO, M. & DAHLWEID, F. M. 2022. Machine Learning-Based Prediction Models for Different Clinical Risks in Different Hospitals: Evaluation of Live Performance. *J Med Internet Res,* 24**,** e34295.

SYKES, A. O. 1993. An introduction to regression analysis.

TENNANT, P., DOXFORD-HOOK, E., FLYNN, L., KERSHAW, K., GODDARD, J. & STACEY, T. 2022. Fasting plasma glucose, diagnosis of gestational diabetes and the risk of large for gestational age: a regression discontinuity analysis of routine data. *BJOG,* 129**,** 82-89.

TERAUCHI, Y., KUBOTA, N., TAMEMOTO, H., SAKURA, H., NAGAI, R., AKANUMA, Y., KIMURA, S. & KADOWAKI, T. 2000. Insulin effect during embryogenesis determines fetal growth: a possible molecular link between birth weight and susceptibility to type 2 diabetes. *Diabetes,* 49**,** 82-6.

VIDMAR, S. I., COLE, T. J. & PAN, H. 2013. Standardizing Anthropometric Measures in Children and Adolescents with Functions for Egen: Update. *The Stata Journal,* 13**,** 366-378.

VOSKAMP, B. J., PEELEN, M., RAVELLI, A. C. J., VAN DER LEE, R., MOL, B. W. J., PAJKRT, E., GANZEVOORT, W. & KAZEMIER, B. M. 2020. Association between fetal sex, birthweight percentile and adverse pregnancy outcome. *Acta Obstet Gynecol Scand,* 99**,** 48-58.

WANG, N., GUO, H., JING, Y., ZHANG, Y., SUN, B., PAN, X., CHEN, H., XU, J., WANG, M., CHEN, X., SONG, L. & CUI, W. 2023. Development and validation of risk prediction models for large for gestational age infants using logistic regression and two machine learning algorithms. *J Diabetes,* 15**,** 338-348.

WARRINGTON, N. M., BEAUMONT, R. N., HORIKOSHI, M., DAY, F. R., HELGELAND, O., LAURIN, C., BACELIS, J., PENG, S., HAO, K., FEENSTRA, B., WOOD, A. R., MAHAJAN, A., TYRRELL, J., ROBERTSON, N. R., RAYNER, N. W., QIAO, Z., MOEN, G. H., VAUDEL, M., MARSIT, C. J., CHEN, J., NODZENSKI, M., SCHNURR, T. M., ZAFARMAND, M. H., BRADFIELD, J. P., GRARUP, N., KOOIJMAN, M. N., LI-GAO, R., GELLER, F., AHLUWALIA, T. S., PATERNOSTER, L., RUEEDI, R., HUIKARI, V., HOTTENGA, J. J., LYYTIKAINEN, L. P., CAVADINO, A., METRUSTRY, S., COUSMINER, D. L., WU, Y., THIERING, E., WANG, C. A., HAVE, C. T., VILOR-TEJEDOR, N., JOSHI, P. K., PAINTER, J. N., NTALLA, I., MYHRE, R., PITKANEN, N., VAN LEEUWEN, E. M., JORO, R., LAGOU, V., RICHMOND, R. C., ESPINOSA, A., BARTON, S. J., INSKIP, H. M., HOLLOWAY, J. W., SANTA-MARINA, L., ESTIVILL, X., ANG, W., MARSH, J. A., REICHETZEDER, C., MARULLO, L., HOCHER, B., LUNETTA, K. L., MURABITO, J. M., RELTON, C. L., KOGEVINAS, M., CHATZI, L., ALLARD, C., BOUCHARD, L., HIVERT, M. F., ZHANG, G., MUGLIA, L. J., HEIKKINEN, J., CONSORTIUM, E. G. G., MORGEN, C. S., VAN KAMPEN, A. H. C., VAN SCHAIK, B. D. C., MENTCH, F. D., LANGENBERG, C., LUAN, J., SCOTT, R. A., ZHAO, J. H., HEMANI, G., RING, S. M., BENNETT, A. J., GAULTON, K. J., FERNANDEZ-TAJES, J., VAN ZUYDAM, N. R., MEDINA-GOMEZ, C., DE HAAN, H. G., ROSENDAAL, F. R., KUTALIK, Z., MARQUES-VIDAL, P., DAS, S., WILLEMSEN, G., MBAREK, H., MULLER-NURASYID, M., STANDL, M., APPEL, E. V. R., FONVIG, C. E., et al. 2019. Maternal and fetal genetic effects on birth weight and their relevance to cardio-metabolic risk factors. *Nat Genet,* 51**,** 804-814.

WEST, J., LAWLOR, D. A., FAIRLEY, L., BHOPAL, R., CAMERON, N., MCKINNEY, P. A., SATTAR, N. & WRIGHT, J. 2013. UK-born Pakistani-origin infants are relatively more adipose than white British infants: findings from 8704 mother-offspring pairs in the Born-in-Bradford prospective birth cohort. *J Epidemiol Community Health,* 67**,** 544-51.

WRIGHT, J., SMALL, N., RAYNOR, P., TUFFNELL, D., BHOPAL, R., CAMERON, N., FAIRLEY, L., LAWLOR, D. A., PARSLOW, R., PETHERICK, E. S., PICKETT, K. E., WAIBLINGER, D., WEST, J. & BORN IN BRADFORD SCIENTIFIC COLLABORATORS, G. 2013. Cohort Profile: the Born in Bradford multi-ethnic family cohort study. *Int J Epidemiol,* 42**,** 978-91.

Chapter 3

Assessing whether genetic scores explain extra variation in birthweight, when added to clinical and anthropometric measures

Assessing whether genetic scores explain extra variation in birthweight, when added to clinical and anthropometric measures

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Abstract

Background: Human birthweight is a complex, multifactorial trait. Maternal characteristics contribute to birthweight variation by influencing the intrauterine environment. Variation explained by genetic effects is also important, but their contributions have not been assessed alongside other key determinants. We aimed to investigate variance in birthweight explained by genetic scores in addition to easily measurable clinical and anthropometric variables.

Methods: We analysed 549 European-ancestry parent-offspring trios from a UK community-based birth cohort. We investigated variance explained in birthweight (adjusted for sex and gestational age) in multivariable linear regression models including genetic scores, routinely measured maternal characteristics, and parental anthropometric variables. We used R-Squared (R^2) to estimate variance explained, adjusted R-squared (Adj-R^2) to assess improvement in model fit from added predictors, and F-tests to compare nested models.

Results: Maternal and fetal genetic scores together explained 6.0% variance in birthweight. A model containing maternal age, weight, smoking, parity, and 28-week fasting glucose explained 21.7% variance. Maternal genetic score explained additional variance when added to maternal characteristics (Adj-R^2 = 0.233 vs Adj-R^2 = 0.210, $p < 0.001$). Fetal genetic score improved variance explained (Adj-R^2 = 0.264 vs 0.248, $p < 0.001$) when added to maternal characteristics and parental heights.

Conclusions: Genetic scores account for variance explained in birthweight in addition to easily measurable clinical variables. Parental heights partially capture fetal genotype and its contribution to birthweight, but genetic scores explain additional variance. While the genetic contribution is modest, it is comparable to that of individual clinical characteristics such as parity, which suggests that genetics could be included in tools aiming to predict risk of high or low birthweights.

Introduction

Birthweight is a complex trait with considerable variability. It is important to understand what contributes to this variability because babies born large for gestational age (LGA) or small for gestational age (SGA) are at a higher risk for adverse pregnancy and perinatal outcomes (Kc et al., 2015). There are also well replicated associations between variation in birthweight and risks of later life cardio-metabolic disease (Barker, 1995, Barker et al., 1993, Knop et al., 2018) .

Previous research has shown that factors associated with the maternal intrauterine environment, for example, maternal glycaemia, age, parity, weight and smoking, account for some variation in birthweight, once fetal sex and gestational duration have been accounted for (Catalano et al., 1995). Maternal smoking during pregnancy is associated with lower birthweight (Kataoka et al., 2018). Parity is also associated with birthweight (Seidman et al., 1988, Shah and births, 2010), with babies of later birth order having higher birthweight, on average. A low pre-pregnancy BMI increases the risk of SGA and a high pre-pregnancy BMI has been found to increase the risk of LGA (Yu et al., 2013). There is a positive continuous association between maternal fasting glucose and birthweight(Breschi et al., 1993). However, each of these variables contributes only modestly to birthweight variation. For example, maternal fasting glucose levels have been reported to explain only a small fraction (10%) of variation in birthweight(Breschi et al., 1993), and most LGA babies are not born to mothers with glucose levels that are high enough to be classified as diabetes(Sacks et al., 2006).

Fetal genetic variation contributes to variation in birthweight independently of the intrauterine environment and is therefore important to consider. Some of the fetal genetic contribution to birthweight can be captured by measuring paternal or maternal height. Height is a highly heritable trait, and the correlation between birthweight and paternal height in particular, via fetal skeletal growth (Knight et al., 2005)occurs due to genetic inheritance.

A recent genome-wide-association study (GWAS) identified 190 regions of the genome where common single nucleotide polymorphisms (SNPs) are associated with birthweight variation (Warrington et al., 2019). The associated genetic variants at three-quarters of the 190 identified loci exert their effects directly from the fetal genotype, with a small proportion of those showing additional maternal effects. Associated variants at the other quarter of identified loci originated from the mother's genome and showed indirect effects, via the maternal environment. A fetal genetic score consisting of 58 variants was shown to make a significant contribution to birthweight independently of maternal glucose levels (Hughes et al., 2018), suggesting measurements of fetal genetics could add to the variance in birthweight explained by other factors. However, the contribution of genetic variation to birthweight has not been assessed directly alongside other clinical variables. We therefore aimed to assess the contributions of genetic scores to variation in offspring birthweight, in addition to easily obtained clinical and anthropometric variables, in a UK community-based study of mothers, fathers and children.

Methods

All methods were carried out in accordance with relevant guidelines and regulations.

Study population

We used data from the Exeter Family Study of Childhood Health (EFSOCH) (Knight et al., 2006). EFSOCH is a study based on children born between 2000 and 2004 in postcodes EX1-4 in central Exeter, UK.

Inclusion criteria for the current analyses consisted of only those parent-offspring trios where the offspring was born at term (≥37 and <42 weeks gestation (Knight et al., 2006)) and complete clinical, anthropometric and maternal, paternal and fetal genetic data were available. Most trios had complete phenotype data, but following genotype quality control, and owing mainly to lower availability of fetal DNA from cord blood compared with parental DNA, complete genotype data was available for both parents and offspring in 60% of the trios. The final dataset consisted of 549 parent-offspring trios. The selection of variables is illustrated by the flowchart in Figure 1. To check for any differences between excluded and included participants, we used t-tests to compare means of continuous variables of the excluded with the included (maternal height, maternal weight, gestational duration, birthweight and maternal age), and chi-square tests to compare the excluded categorical variables with the included categorical variables (maternal smoking status, parity and sex of the baby).

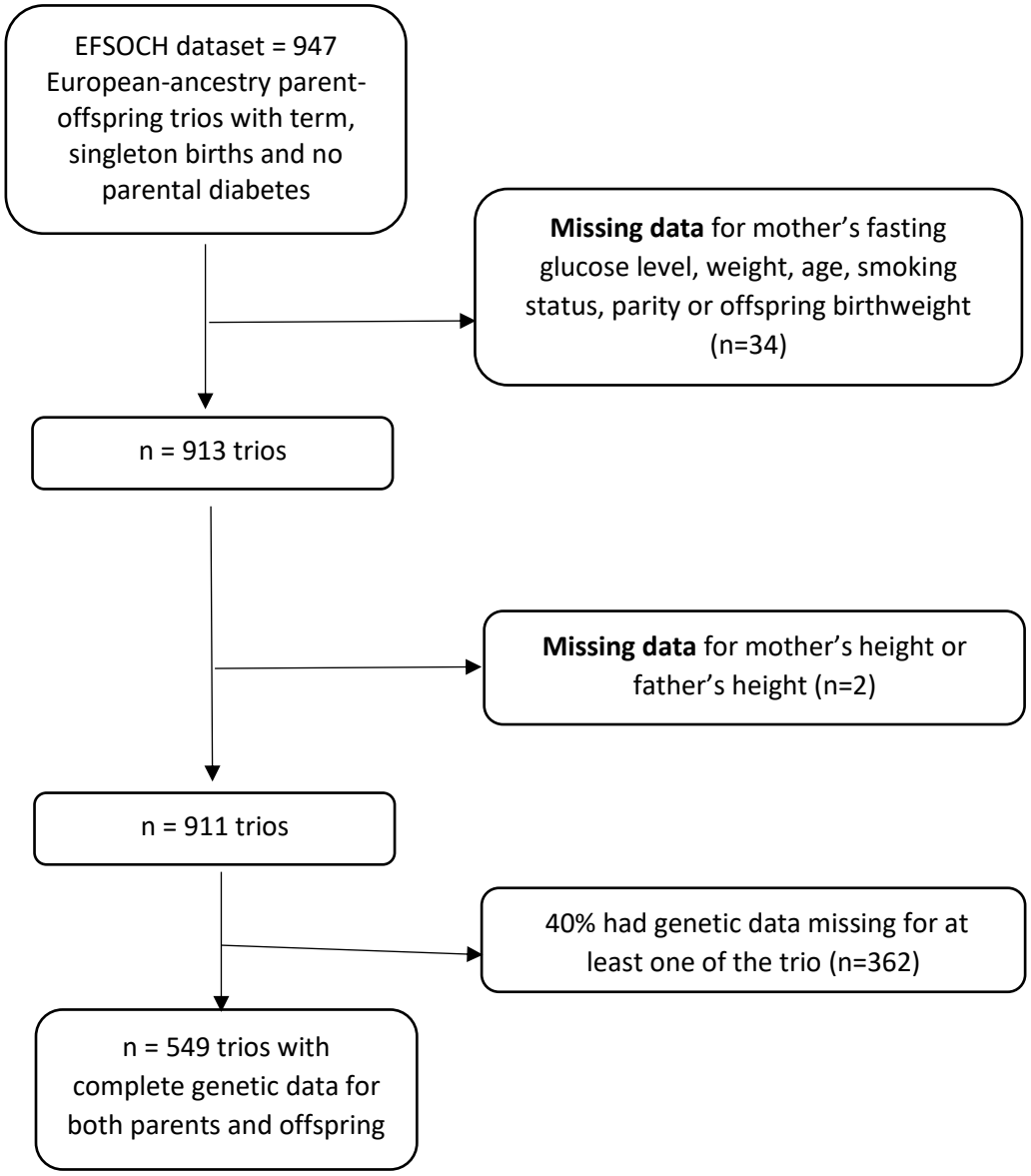

Fig 1: Flowchart illustrating how the data was prepared for analysis

Characteristics of participants

Full details of data collection are found in the EFSOCH study protocol (Knight et al., 2006). Briefly, detailed anthropometric measurements and biochemistry from the parents were taken at 28 weeks' gestation. All measurements were taken three times and an average value was calculated. Maternal and paternal heights were measured to the nearest 0.1 cm. Maternal weight was measured to the nearest 100 g. Birthweights of the parents were self-reported. Offspring birthweight was measured at birth, to the nearest 10 g and adjusted for sex and gestational age, centred around 40 weeks, according to the UK 1990 birthweight standards (Cole et al., 1998).

Maternal glucose was measured in fasting maternal samples (fasting for at least 10h prior to sampling), early morning at the parents' home. Pregnancy details such as parity were obtained from medical records. Information about the mother's smoking status was obtained via a questionnaire completed by the mother at recruitment.

Genotyping

Parental and offspring DNA were extracted to allow molecular genetic analysis of variants implicated in fetal growth. At birth, a sample of cord blood was taken at delivery. DNA was extracted from the spun white cells. The EFSOCH sample (consisting of 2,768 samples: mothers (n=969), fathers (n=937) and offspring (n=862)) genotyping was carried out using the Illumina HumanCoreExome array. A total of 106 samples were excluded due to low call rate, kinship errors, sex mismatches or ancestry outliers. The 2662 included samples were of European ancestry (assessed using flashPCA (Abraham and Inouye, 2014) with genotype call rates >98% and phenotypic sex and kinship were validated using genotype data assessed by KING software(Manichaikul et al., 2010)). The included genotyped SNPs had call rates >95%, Hardy-Weinberg $p > 1 \times 10^{-6}$, and minor allele frequency (MAF) >1%. The Haplotype Reference Consortium (HRC) version r1.1 reference panel (Michigan Imputation Server) was used to impute the genotypes in all samples. A total of 98% of the SNPs included in the scores had an

imputation quality > 0.4 and a Minor Allele Frequency > 0.001 in EFSOCH (see **Table S1; Supplementary Info**).

Statistical Analyses

Genetic scores

We created independent maternal and fetal genetic scores for birthweight, and also a paternal genetic score for father's own birthweight (analogous to a fetal genetic score). We calculated the genetic scores (GS) according to Equation 1, where N_{SNP} is the total number of SNPs, w_i is the weight for SNP i and g_i is the genotype dosage at SNP i.

$$\boldsymbol{GS = \sum_i w_i g_i} \qquad \textbf{(1)}$$

A total of 209 SNPs, identified at 190 loci in the most recent GWAS of birthweight (Warrington et al., 2019), were used to calculate the maternal, paternal and fetal genetic scores (see **Table S1; Supplementary Info**).Effect estimates for each SNP were used as weights, and for the maternal score, these had been adjusted to represent the maternal effects independent of fetal genotype effects using a structural equation model (Warrington et al., 2019). For the fetal score, fetal effect estimates independent of maternal genotype effects were used as weights, and for the paternal score for father's own birthweight, the fetal GWAS weights were unadjusted so as to capture maximum information. Each genetic score variable was then standardized to a mean of 0 and SD of 1. To validate the genetic scores, we tested the associations between each standardized genetic score and its respective phenotype using simple linear regression models.

Linear regression models to estimate variance in adjusted birthweight by genetic and other factors

We used multivariable linear regression models to model the variance in birthweight explained by several clinical, anthropometric and genetic factors. We ensured that the regression model assumptions were met and the model assumptions were checked using diagnostic plots of residuals and fitted values. To determine the additional variability explained by genetics, we examined the following models, with birthweight (adjusted for sex and gestational age) as the outcome variable:

Model 1: Genetic scores model: maternal and fetal genetic scores were included as predictors to investigate their contribution to birthweight.

$$Birth\ weight \sim Maternal\ genetic\ score + Fetal\ genetic\ score$$

Model 2: Maternal clinical features (intrauterine environment) model: maternal fasting glucose, age, weight, parity and the mother's smoking status were used in this model.

$$Birth\ weight \sim Maternal\ fasting\ glucose + Age + Weight + Parity + Mother'smoking\ status$$

Model 3: Maternal genetic score + maternal clinical (intrauterine environment) features: The maternal genetic score was added to Model 2 to investigate the additional contribution of maternal genetics to variance explained in birthweight.

$$Birth\ weight \sim Maternal\ fasting\ glucose + Age + Weight + Parity + Mother'smoking\ status + Maternal\ genetic\ score$$

Model 4: Maternal clinical features + Parental anthropometric traits (genetics) model: Maternal and paternal height are variables that capture the effects of fetal

genetics and are easily measurable; these were added as predictors to Model 2 to create Model 4.

$$Birth\ weight \sim Maternal\ fasting\ glucose + Age + Weight + Parity + Mother's moking\ status + \ Maternal\ height + Paternal\ height$$

Model 5: Fetal genetic score + maternal clinical features + parental anthropometric (genetic) traits: The fetal genetic score was added to Model 4 to further investigate the contributions of the fetal genetic score in addition to parental heights and clinical features.

$$Birth\ weight \sim Maternal\ fasting\ glucose + Age + Weight + Parity + Mother's moking\ status + \ Maternal\ height + Paternal\ height + Fetal\ genetic\ score$$

Model 6: Parental genetic scores + maternal clinical features + parental anthropometric (genetic) traits: Given that the fetal genetic score for birthweight is not available prior to delivery, we analysed the contribution of the maternal genetic score for offspring birthweight and the paternal genetic score for father's own birthweight in Model 6 in addition to clinical features and parental heights.

$$Birth\ weight \sim Maternal\ fasting\ glucose + Age + Weight + Parity + Mother's moking\ status + \ Maternal\ height + Paternal\ height + Maternal\ genetic\ score + Paternal\ genetic\ score$$

Model 7: Fetal genetic score + maternal genetic score + maternal clinical features + parental anthropometric (genetic) traits:

The maternal genetic score was added to Model 5 to further investigate the contributions of the maternal genetic score in addition to parental heights and clinical features.

$$Birth\ weight \sim Maternal\ fasting\ glucose + Age + Weight + Parity + Mother's smoking\ status + Maternal\ height + Paternal\ height + Fetal\ genetic\ score + Maternal\ genetic\ score$$

Additional models: Parents' own birthweights: We additionally investigated the contribution of maternal and paternal self-reported birthweights because these may also capture information about fetal genetics. These were available in a smaller sample (n=425 trios).

We used the Adj-R^2 statistic to assess improvement in model fit based on any added predictors. An F-test was used to compare nested models and check for any improvements in the explanation of variance in birthweight. The R^2 statistic and its 95% confidence intervals were used to assess the overall explanation of variance in birthweight by the predictors in the model. Confidence intervals were calculated by bootstrapping. Multicollinearity between predictor variables in the models was checked by using the Variance Inflation Factor (VIF).

As a sensitivity analysis to check for any potential impact of poor-quality SNP genotype data, we repeated models containing genetic scores with only those SNPs that had minor allele frequency > 0.1% and imputation quality $r^2 > 0.4$. We used the statistical software R (version 3.5.2) to develop the multiple linear regression models and to calculate the F-tests between nested models.

Results

Descriptive characteristics for the 549 parent-offspring trios are shown in Table 1. There was no strong evidence that individuals excluded from the analysis differed in their basic characteristics from those included (see **Table S2**)

n = 549 trios	
Phenotype	**Mean or % (SD)**
Maternal Height (cm)	165.0 (6.4)
Maternal Weight (kg)	76.3 (12.6)
Gestational Duration (weeks)	40.1 (1.2)
Birthweight (g)	3570 (444)
Maternal Age (years)	30 (5)
Maternal smoking status (%Yes)	14.6
Parity (%1st pregnancy)	44.8
Sex of the baby (% Male)	52.2
Maternal Fasting Glucose(mmol/L)	4.4(0.4)
Father's Height (cm)	178(5.1)

Table 1: Key characteristics of study population

The genetic scores all showed strong associations with their respective phenotypes (**Table S3**).

Maternal and fetal genetic scores contribute additively to offspring birthweight variation

Variable	Change in birthweight (g) per 1 SD change in independent variable	95% Confidence Interval	t value	p-value
Intercept	3672	3530,3813	50.9	<0.001
Maternal genetic score for offspring birthweight (adjusted for fetal effects)	81	45,116	4.4	<0.001
Fetal genetic score for offspring birthweight (adjusted for maternal effects)	69	33,105	3.7	<0.001

Table 2: Model 1**-** Results of a multivariable linear regression model testing the association between birthweight (adjusted for sex and gestational age), maternal genetic score and fetal genetic score (n=549 parent-offspring trios).
R^2 = 0.060; Adj-R^2=0.053

A multivariable linear regression model (Model 1; **Table 2**) showed that maternal and fetal genetic scores have additive contributions to variance in offspring birthweight**.** On its own, the fetal genetic score explained 2% of variation in adjusted birthweight (R^2 = 0.020) and the maternal genetic score explained 3% of variance in birthweight (R^2 = 0.030). For comparison, the variables parity, mother's smoking status and paternal height each explained 3 % of variation.

Maternal genetic score for birthweight explained additional variance in birthweight when added to easily measurable clinical variables

A multivariable linear regression model (Model 2; **Table 3)** including variables that are readily available in the clinical setting (maternal fasting glucose, maternal age, maternal weight, parity and the mother's smoking status), showed that each variable contributed to variance explained in birthweight**.** The total variation in birthweight explained by these maternal characteristics (R^2 = 0.217) was higher than that explained by genetic scores alone (R^2 = 0.06; Model 1).

Variable	Change in birthweight (g) per 1 SD change in independent variable	95% Confidence Interval	t value	p-value
Model 2: maternal clinical characteristics (n=549 parent-offspring trios). R^2 = 0.217; Adj-R^2=0.210				
Intercept	3691	3643, 3740	149.1	<2e-16
Maternal age	-38	-73, -3	-2.1	0.04
Maternal weight	125	89, 161	6.8	2.09 e-11
Mother's smoking status*	-280	-377, -182	-5.6	3.03 e-08
Parity*	-187	-255, -118	-5.3	1.55e-07
Mother's fasting glucose at 28 weeks' gestation	87	51, 123	4.7	2.83e-06
Model 3: maternal clinical characteristics and maternal genetics (n=549 parent-offspring trios). R^2 = 0.244; Adj-R^2=0.233				
Intercept	3790	3655, 3925	55.3	<2e-16
Maternal age	-42	-77, -8	-2.4	0.026
Maternal weight	121	85, 156	6.7	1.18e-10
Mother's smoking status	-273	-369, -176	-5.6	8.45e-08
Parity	-194	-262, -126	-5.6	3.45e-08
Mother's fasting glucose at 28 weeks' gestation	85	49, 120	4.1	8.93e-07
Maternal genetic score for offspring birthweight (adjusted for fetal effects)	68	36, 101	4.7	0.00023

* indicates a binary variable

Table 3: Results of multivariable linear regression models testing the association between birthweight (adjusted for sex and gestational age) and maternal clinical characteristics, with and without the maternal genetic score

The addition of the maternal genetic score for offspring birthweight to Model 2 as a predictor (Model 3; **Table 3**) made little change to the coefficients of the maternal clinical variables, which were very similar to Model 2, but there was an improvement in the Adj-R^2 statistic when comparing the nested models (Adj-R^2 =0.233 vs 0.210, p<0.001), indicating that the maternal genetic score captured additional variance in birthweight.

Maternal and paternal height explained additional variance in birthweight when added to maternal clinical variables

The addition of maternal and paternal height variables, that can capture the effects of fetal genetics, to Model 2 (routinely available clinical features only) showed that the additional variables can further explain variance in birthweight (adjusted for sex and gestational age) (Model 4; **Table 4**) with Adj-R^2 increasing from 0.210 to 0.248 ($p<0.001$).

Variable	Change in birthweight (g) per 1 SD change in independent variable	95% Confidence Interval	t value	p-value
Model 4: maternal clinical characteristics and parental heights (n=549 parent-offspring trios). R^2 = 0.258; Adj-R^2=0.248				
Intercept	3697	3650, 3744	152.7	<2e-16
Maternal age	-49	-84, -15	-2.8	0.00547
Maternal weight	101	64, 139	5.3	1.47e-07
Mother's smoking status	-251	-348, -155	-5.1	3.85e-07
Parity	-210	-278, -142	-6.1	2.47e-09
Mother's fasting glucose at 28 weeks' gestation	104	68, 140	5.7	2.03e-08
Maternal height	52	16, 87	2.9	0.00431
Paternal height	69	35, 102	4.0	6.88e-05
Model 5: maternal clinical characteristics (n=549 parent-offspring trios), parental heights, and fetal genetic score. R^2 = 0.277; Adj-R^2=0.264				
Intercept	3799	3667, 3931	56.5	<2e-16
Maternal age	-50	-84, -16	-2.9	0.00427
Maternal weight	97	60, 134	5.2	3.64e-07
Mother's smoking status	-241	-336, -146	-5.0	8.65e-07
Parity	-216	-284, -149	-6.3	6.43e-10
Mother's fasting glucose at 28 weeks' gestation	106	71, 142	5.9	7.42e-09
Maternal height	49	14, 84	2.7	0.00681
Paternal height	66	33, 99	3.9	0.000121
Fetal genetic score for offspring birthweight (adjusted for maternal effects)	56	23, 88	3.4	0.000746

Table 4: Results of multivariable linear regression models testing the association between birthweight (adjusted for sex and gestational age), maternal clinical characteristics and parental heights, with and without the fetal genetic score (n=549 parent-offspring trios).

In a subsample of n=425 available trios, we found that mother's and father's own self-reported birthweights explained additional variance in offspring birthweight

when added to a model that included parental heights (**Table S4,** Adj-R^2=0.302 vs 0.258 without parent birthweights, p<0.001)**.**

Fetal genetic score for birthweight explained additional variance in birthweight when added to easily-measured anthropometric variables that capture fetal genotype

With the addition of the fetal genetic score for offspring birthweight to Model 4 as a predictor (Model 5; **Table 4**), there was little change in the coefficients of the maternal clinical variables, or of the maternal and paternal heights, which were very similar to Model 4, but there was an improvement in the Adj-R^2 statistic when comparing the nested models (Adj-R^2 =0.264 vs. 0.248, p<0.001), indicating that the fetal genetic score captured additional variance in birthweight. The fetal genetic score also improved variance explained in the model containing parental birthweights in a subsample of 425 trios (**Table S5**; P=0.09 comparing Adj-R^2=0.302 for the model with no fetal genetic score with Adj-R^2= 0.310 for the model with the fetal genetic score).

Maternal and paternal genetic scores further improved variance explained in birthweight when added to clinical and anthropometric variables

When we added the maternal and paternal genetic scores to Model 4, (Model 6**;** **Table 5**), both parental genetic scores explained variation in birthweight on top of the basic clinical and anthropometric variables (Adj-R^2=0.271 vs Adj-R^2=0.248, p<0.001).

Variable	Change in birthweight (g) per 1 SD change in independent variable	95% Confidence Interval	t value	p-value
Model 2: maternal clinical characteristics (n=549 parent-offspring trios). R^2 = 0.217; Adj-R^2=0.210				
Intercept	3691	3643, 3740	149.1	<2e-16
Maternal age	-38	-73, -3	-2.1	0.04
Maternal weight	125	89, 161	6.8	2.09 e-11
Mother's smoking status*	-280	-377, -182	-5.6	3.03 e-08
Parity*	-187	-255, -118	-5.3	1.55e-07
Mother's fasting glucose at 28 weeks' gestation	87	51, 123	4.7	2.83e-06
Model 3: maternal clinical characteristics and maternal genetics (n=549 parent-offspring trios). R^2 = 0.244; Adj-R^2=0.233				
Intercept	3790	3655, 3925	55.3	<2e-16
Maternal age	-42	-77, -8	-2.4	0.026
Maternal weight	121	85, 156	6.7	1.18e-10
Mother's smoking status	-273	-369, -176	-5.6	8.45e-08
Parity	-194	-262, -126	-5.6	3.45e-08
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Maternal genetic score for offspring birthweight (adjusted for fetal effects)	68	36, 101	4.7	0.00023

* indicates a binary variable

Table 3: Results of multivariable linear regression models testing the association between birthweight (adjusted for sex and gestational age) and maternal clinical characteristics, with and without the maternal genetic score

The addition of the maternal genetic score for offspring birthweight to Model 2 as a predictor (Model 3; **Table 3**) made little change to the coefficients of the maternal clinical variables, which were very similar to Model 2, but there was an improvement in the Adj-R^2 statistic when comparing the nested models (Adj-R^2 =0.233 vs 0.210, p<0.001), indicating that the maternal genetic score captured additional variance in birthweight.

Maternal and paternal height explained additional variance in birthweight when added to maternal clinical variables

Variable	Change in birthweight (g) per 1 SD change in independent variable	95% Confidence Interval	t value	p-value
Model 4: maternal clinical characteristics and parental heights (n=549 parent-offspring trios). R^2 = 0.258; Adj-R^2=0.248				
Intercept	3697	3650, 3744	152.7	<2e-16
Maternal age	-49	-84, -15	-2.8	0.00547
Maternal weight	101	64, 139	5.3	1.47e-07
Mother's smoking status	-251	-348, -155	-5.1	3.85e-07
Parity	-210	-278, -142	-6.1	2.47e-09
Mother's fasting glucose at 28 weeks' gestation	104	68, 140	5.7	2.03e-08
Maternal height	52	16, 87	2.9	0.00431
Paternal height	69	35, 102	4.0	6.88e-05
Model 5: maternal clinical characteristics (n=549 parent-offspring trios), parental heights, and fetal genetic score. R^2 = 0.277; Adj-R^2=0.264				
Intercept	3799	3667, 3931	56.5	<2e-16
Maternal age	-50	-84, -16	-2.9	0.00427
Maternal weight	97	60, 134	5.2	3.64e-07
Mother's smoking status	-241	-336, -146	-5.0	8.65e-07
Parity	-216	-284, -149	-6.3	6.43e-10
Mother's fasting glucose at 28 weeks' gestation	106	71, 142	5.9	7.42e-09
Maternal height	49	14, 84	2.7	0.00681
Paternal height	66	33, 99	3.9	0.000121
Fetal genetic score for offspring birthweight (adjusted for maternal effects)	56	23, 88	3.4	0.000746

Table 4: Results of multivariable linear regression models testing the association between birthweight (adjusted for sex and gestational age), maternal clinical characteristics and parental heights, with and without the fetal genetic score (n=549 parent-offspring trios).

The addition of maternal and paternal height variables, that can capture the effects of fetal genetics, to Model 2 (routinely available clinical features only) showed that the additional variables can further explain variance in birthweight (adjusted for sex and gestational age) (Model 4; **Table 4**) with Adj-R^2 increasing from 0.210 to 0.248 ($p<0.001$).

In a subsample of n=425 available trios, we found that mother's and father's own self-reported birthweights explained additional variance in offspring birthweight when added to a model that included parental heights (**Table S4,** Adj-R^2=0.302 vs 0.258 without parent birthweights, $p<0.001$)**.**

Fetal genetic score for birthweight explained additional variance in birthweight when added to easily measured anthropometric variables that capture fetal genotype

With the addition of the fetal genetic score for offspring birthweight to Model 4 as a predictor (Model 5; **Table 4**), there was little change in the coefficients of the maternal clinical variables, or of the maternal and paternal heights, which were very similar to Model 4, but there was an improvement in the Adj-R^2 statistic when comparing the nested models (Adj-R^2 =0.264 vs. 0.248, $p<0.001$), indicating that the fetal genetic score captured additional variance in birthweight. The fetal genetic score also improved variance explained in the model containing parental birthweights in a subsample of 425 trios (**Table S5**; P=0.09 comparing Adj-R^2=0.302 for the model with no fetal genetic score with Adj-R^2= 0.310 for the model with the fetal genetic score).

Maternal and paternal genetic scores further improved variance explained in birthweight when added to clinical and anthropometric variables

When we added the maternal and paternal genetic scores to Model 4, (Model 6**;** **Table 5**), both parental genetic scores explained variation in birthweight on top of the basic clinical and anthropometric variables (Adj-R^2=0.271 vs Adj-R^2=0.248, p<0.001).

Variable	Change in birthweight (g) per 1 SD change in independent variable	95% Confidence Interval	t-value	p-value
Intercept	3796	3665, 3927	56.7	<2e-16
Maternal age	-54	-88, -20	-3.1	0.00215
Maternal weight	97	60, 134	5.1	3.78e-07
Mother's smoking status	-241	-335, -146	-5.0	8.45e-07
Parity	-211	--278, -144	-6.2	1.25e-09
Mother's fasting glucose at 28 weeks' gestation	104	69, 140	5.8	1.50e-08
Maternal height	44	9, 80	2.5	0.0138
Paternal height	61	28, 95	3.6	0.000349
Maternal genetic score for offspring birthweight (adjusted for fetal effects)	57	25, 90	3.5	0.000567
Paternal genetic score for father's own birthweight	39	6, 71	2.3	0.0192

Table 5: Model 6-Results of a multivariable linear regression model testing the association between birthweight (adjusted for sex and gestational age), maternal clinical characteristics (n=549 parent-offspring trios), and parental heights and genetic scores. R^2 = 0.285; Adj-R^2=0.271

Maternal genetic score further improved variance explained in birthweight when added to fetal genetic score

When the maternal genetic score and the fetal genetic scores were added on top of clinical variables (Model 7; **Table 6**), in Model 4 there was additional improvement in explanation of variance in birthweight (Adj-R^2=0.280 vs Adj-R^2=0.248, p<0.001).

Variable	Change in birthweight (g) per 1 SD change in independent variable	95% Confidence Interval	t-value	p-value
Intercept	3799	3669, 3929	57.1	<2e-16
Maternal age	-53	-87, -19	-3.1	0.00231
Maternal weight	98	62, 135	5.3	2.11e-07
Mother's smoking status	-241	-335, -147	-5.0	6.98e-07
Parity	-217	-283, -150	-6.4	3.94e-10
Mother's fasting glucose at 28 weeks' gestation	101	66, 136	5.7	2.59e-08
Maternal height	39	4, 74	2.2	0.0288
Paternal height	64	31, 96	3.7	0.000178
Maternal genetic score for offspring birthweight (adjusted for fetal effects)	60	27, 92	3.6	0.000314
Fetal genetic score for offspring birthweight(adjusted for maternal effects)	57	25, 89	3.5	0.000449

Table 6: Model 7-Results of a multivariable linear regression model testing the association between birthweight (adjusted for sex and gestational age), maternal clinical characteristics (n=549 parent-offspring trios), parental heights, and maternal and fetal genetic scores. R^2 = 0.294; Adj-R^2=0.280

A summary of the R^2 values across all the main models is shown in **Figure 2.** This indicates the improvement in R^2 with added successive variables.

There was a negligible difference between the models that contained genetic scores with only those SNPs that had minor allele frequency > 0.1% and imputation quality r^2 > 0.4 and the models that contained genetic scores with 98% of the SNPs having minor allele frequency > 0.001 and imputation quality > 0.4.

Discussion

We have shown that maternal, paternal and fetal genetic scores contribute to variation in sex- and gestational age-adjusted birthweight, in addition to variables easily obtained in a clinical setting. We also showed that maternal and paternal heights, which are easily measured and capture some of the genetic contribution to fetal growth, explain variance in birthweight independently of routinely measured maternal clinical variables. However, the maternal and fetal (or paternal) genetic scores made additional, independent contributions to birthweight variance. GWAS have established that fetal and maternal genetic variants are associated with birthweight (Warrington et al., 2019), but as many of the underlying causal genes are likely associated with clinical or anthropometric traits, such as height, weight, and glucose, it has been important to quantify the added value.

Maternal and fetal genetics are known to be important determinants of fetal growth but the contribution of genetic scores to variance explained in birthweight has not been investigated previously using multivariable regression models containing other clinical and parental anthropometric characteristics. We showed, consistent with other epidemiological studies (Nahum et al., 1999, Makgoba et al., 2012), that clinical variables, both routinely measured (glucose, weight, smoking), but also parental height, can explain approximately 26% variation in birthweight that has already been adjusted for sex and gestational age. The addition of the fetal genetic score to the models explained a further 2% of variation in birthweight. For comparison, the variables parity, mother's smoking status and paternal height each explained 3 % of variation individually, in sex-and gestational age-adjusted

birthweight. The precise mechanisms through which most of the genetic variants in the fetal score influence growth are not known, but evidence to date suggests they are likely to capture variation in growth factors such as fetal insulin, as well as variation in placental growth and function (Warrington et al., 2019).

Fetal genetic scores are not available before birth, so they are not informative for predicting birthweight at present. However, we showed that maternal and paternal genetic scores can also explain variation in birthweight. The parental genetic effects are mediated both through direct effects of genes inherited by the fetus and indirect maternal genetic effects on the intra-uterine environment. Some of these effects will have been captured by clinical features. Previous research has shown that associations between maternal height and offspring birthweight is predominantly defined by fetal genetics (Zhang et al., 2015). Paternal height has also been shown to influence offspring birthweight through fetal genetics (Knight et al., 2006). We have shown that the parental heights explain further variation in birthweight and that parental genetic scores for birthweight are contributing to variation in birthweight independently of parental heights. The independent and additive associations of the parental genetic scores with birthweight show that these scores are offering additional predictive value. The fetal genetic score also added information on top of self-reported parent birthweights.

It was unexpected that the R^2 value for the maternal GS was larger than that of the fetal GS because previous work (Warrington et al., 2019) has shown that fetal genetic variants explain more birthweight variation than maternal genetic variants. However, further investigation showed that the R^2 values for maternal and fetal genetic scores were not precise enough in this relatively small sample to be able to infer confidently whether one was bigger than the other (as reflected in the 95% confidence intervals), and point estimate values of R^2 fluctuated so that the fetal estimate appeared larger than the maternal estimate when the models were re-run in wider samples that did not require all family members to be genotyped (see **Table S6**).

This study has benefited from the use of a well-phenotyped and genotyped sample of parents and children. However, there are some limitations. Firstly, in the

EFSOCH dataset, some clinical features known to contribute to variance explained in birthweight in other studies (e.g. blood pressure (Catalano et al., 1995)) were not available, so studies in additional samples would be needed to enable assessment of the contribution of genetic scores in relation to those variables. In addition, although we aimed to assess the contribution of parents' own birthweights as anthropometric variables in addition to parental heights, the parental birthweights were self-reported and were not available in the full sample (they were available in only 425 complete trios). However, when we created models using the dataset containing 425 trios (**Tables S4-5**), the coefficients of the explanatory variables were similar to those in the models created with the larger dataset of 549 observations, so the limited availability of self-reported birthweights did not impact materially on the results.

Another limitation of this study is that we conducted the analyses in a UK-based, northern European-ancestry population and it is likely that the associations between birthweight and both genetics and parental clinical features will differ in samples of other ancestries and in other settings. Further studies will be necessary to investigate the contribution of genetic scores and other variables to birthweight in other populations.

Since the EFSOCH study was part of the maternal GWAS study that identified SNPs associated with birthweight (Warrington et al., 2019), there is a small risk of overfitting in our models. However, we expect the risk of this to be minimal because EFSOCH only made up 0.4% of the maternal GWAS meta-analysis sample and was not included in the fetal GWAS.

We have shown that maternal and fetal genetic scores explain variation in birthweight in healthy pregnancies, in addition to clinical and anthropometric variables that are routinely or easily collected. While the individual contribution of each genetic score is not large (e.g. 2% for fetal genetic score), it is comparable to the individual contributions of variables such as parity or maternal smoking status. This raises the possibility that genetic scores might be useful alongside clinical characteristics in prediction models, for example, those aiming to predict risk of LGA in pregnancies affected by gestational diabetes. Further work is needed to

determine whether genetic information could improve a full clinical prediction model over and above what is currently done routinely in clinical practice.

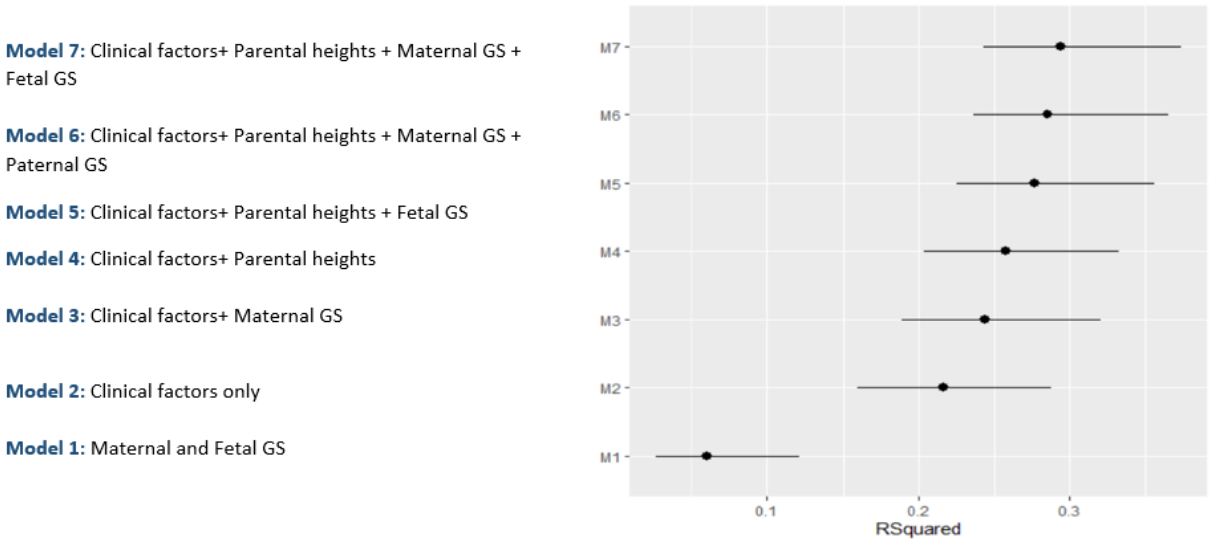

Fig 2: Plot showing R-squared values for each model with 95% confidence intervals.

References

ABRAHAM, G. & INOUYE, M. 2014. Fast principal component analysis of large-scale genome-wide data. *PLoS One,* 9**,** e93766.

BARKER, D. J. 1995. Fetal origins of coronary heart disease. *BMJ,* 311**,** 171-4.

BARKER, D. J., HALES, C. N., FALL, C. H., OSMOND, C., PHIPPS, K. & CLARK, P. M. 1993. Type 2 (non-insulin-dependent) diabetes mellitus, hypertension and hyperlipidaemia (syndrome X): relation to reduced fetal growth. *Diabetologia,* 36**,** 62-7.

BRESCHI, M. C., SEGHIERI, G., BARTOLOMEI, G., GIRONI, A., BALDI, S. & FERRANNINI, E. 1993. Relation of birthweight to maternal plasma glucose and insulin concentrations during normal pregnancy. *Diabetologia,* 36**,** 1315-21.

CATALANO, P. M., DRAGO, N. M. & AMINI, S. B. 1995. Factors affecting fetal growth and body composition. *Am J Obstet Gynecol,* 172**,** 1459-63.

COLE, T. J., FREEMAN, J. V. & PREECE, M. A. 1998. British 1990 growth reference centiles for weight, height, body mass index and head circumference fitted by maximum penalized likelihood. *Stat Med,* 17**,** 407-29.

HUGHES, A. E., NODZENSKI, M., BEAUMONT, R. N., TALBOT, O., SHIELDS, B. M., SCHOLTENS, D. M., KNIGHT, B. A., LOWE, W. L., JR., HATTERSLEY, A. T. & FREATHY, R. M. 2018. Fetal Genotype and Maternal Glucose Have Independent and Additive Effects on Birth Weight. *Diabetes,* 67**,** 1024-1029.

KATAOKA, M. C., CARVALHEIRA, A. P. P., FERRARI, A. P., MALTA, M. B., DE BARROS LEITE CARVALHAES, M. A. & DE LIMA PARADA, C. M. G. 2018. Smoking during pregnancy and harm reduction in birth weight: a cross-sectional study. *BMC Pregnancy Childbirth,* 18**,** 67.

KC, K., SHAKYA, S. & ZHANG, H. 2015. Gestational diabetes mellitus and macrosomia: a literature review. *Ann Nutr Metab,* 66 Suppl 2**,** 14-20.

KNIGHT, B., SHIELDS, B. M. & HATTERSLEY, A. T. 2006. The Exeter Family Study of Childhood Health (EFSOCH): study protocol and methodology. *Paediatr Perinat Epidemiol,* 20**,** 172-9.

KNIGHT, B., SHIELDS, B. M., TURNER, M., POWELL, R. J., YAJNIK, C. S. & HATTERSLEY, A. T. 2005. Evidence of genetic regulation of fetal longitudinal growth. *Early Hum Dev,* 81**,** 823-31.

KNOP, M. R., GENG, T. T., GORNY, A. W., DING, R., LI, C., LEY, S. H. & HUANG, T. 2018. Birth weight and risk of type 2 diabetes mellitus, cardiovascular disease, and hypertension in adults: a meta-analysis of 7 646 267 participants from 135 studies. *Journal of the American Heart Association,* 7**,** e008870.

MAKGOBA, M., SAVVIDOU, M. D. & STEER, P. J. 2012. The effect of maternal characteristics and gestational diabetes on birthweight. *BJOG,* 119**,** 1091-7.

MANICHAIKUL, A., MYCHALECKYJ, J. C., RICH, S. S., DALY, K., SALE, M. & CHEN, W. M. 2010. Robust relationship inference in genome-wide association studies. *Bioinformatics,* 26**,** 2867-73.

NAHUM, G. G., STANISLAW, H. & HUFFAKER, B. J. 1999. Accurate prediction of term birth weight from prospectively measurable maternal characteristics. *J Reprod Med,* 44**,** 705-12.

SACKS, D. A., LIU, A. I., WOLDE-TSADIK, G., AMINI, S. B., HUSTON-PRESLEY, L. & CATALANO, P. M. 2006. What proportion of birth weight is attributable to maternal glucose among infants of diabetic women? *Am J Obstet Gynecol,* 194**,** 501-7.

SEIDMAN, D. S., EVER-HADANI, P., STEVENSON, D. K., SLATER, P. E., HARLAP, S. & GALE, R. 1988. Birth order and birth weight reexamined. *Obstetrics and gynecology,* 72**,** 158-162.

SHAH, P. S. & BIRTHS, K. S. G. O. D. O. L. P. 2010. Parity and low birth weight and preterm birth: a systematic review and meta-analyses. *Acta obstetricia et gynecologica Scandinavica,* 89**,** 862-875.

WARRINGTON, N. M., BEAUMONT, R. N., HORIKOSHI, M., DAY, F. R., HELGELAND, O., LAURIN, C., BACELIS, J., PENG, S., HAO, K., FEENSTRA, B., WOOD, A. R., MAHAJAN, A., TYRRELL, J.,

ROBERTSON, N. R., RAYNER, N. W., QIAO, Z., MOEN, G. H., VAUDEL, M., MARSIT, C. J., CHEN, J., NODZENSKI, M., SCHNURR, T. M., ZAFARMAND, M. H., BRADFIELD, J. P., GRARUP, N., KOOIJMAN, M. N., LI-GAO, R., GELLER, F., AHLUWALIA, T. S., PATERNOSTER, L., RUEEDI, R., HUIKARI, V., HOTTENGA, J. J., LYYTIKAINEN, L. P., CAVADINO, A., METRUSTRY, S., COUSMINER, D. L., WU, Y., THIERING, E., WANG, C. A., HAVE, C. T., VILOR-TEJEDOR, N., JOSHI, P. K., PAINTER, J. N., NTALLA, I., MYHRE, R., PITKANEN, N., VAN LEEUWEN, E. M., JORO, R., LAGOU, V., RICHMOND, R. C., ESPINOSA, A., BARTON, S. J., INSKIP, H. M., HOLLOWAY, J. W., SANTA-MARINA, L., ESTIVILL, X., ANG, W., MARSH, J. A., REICHETZEDER, C., MARULLO, L., HOCHER, B., LUNETTA, K. L., MURABITO, J. M., RELTON, C. L., KOGEVINAS, M., CHATZI, L., ALLARD, C., BOUCHARD, L., HIVERT, M. F., ZHANG, G., MUGLIA, L. J., HEIKKINEN, J., CONSORTIUM, E. G. G., MORGEN, C. S., VAN KAMPEN, A. H. C., VAN SCHAIK, B. D. C., MENTCH, F. D., LANGENBERG, C., LUAN, J., SCOTT, R. A., ZHAO, J. H., HEMANI, G., RING, S. M., BENNETT, A. J., GAULTON, K. J., FERNANDEZ-TAJES, J., VAN ZUYDAM, N. R., MEDINA-GOMEZ, C., DE HAAN, H. G., ROSENDAAL, F. R., KUTALIK, Z., MARQUES-VIDAL, P., DAS, S., WILLEMSEN, G., MBAREK, H., MULLER-NURASYID, M., STANDL, M., APPEL, E. V. R., FONVIG, C. E., et al. 2019. Maternal and fetal genetic effects on birth weight and their relevance to cardio-metabolic risk factors. *Nat Genet,* 51**,** 804-814.

YU, Z., HAN, S., ZHU, J., SUN, X., JI, C. & GUO, X. 2013. Pre-pregnancy body mass index in relation to infant birth weight and offspring overweight/obesity: a systematic review and meta-analysis. *PLoS One,* 8**,** e61627.

ZHANG, G., BACELIS, J., LENGYEL, C., TERAMO, K., HALLMAN, M., HELGELAND, O., JOHANSSON, S., MYHRE, R., SENGPIEL, V., NJOLSTAD, P. R., JACOBSSON, B. & MUGLIA, L. 2015. Assessing the Causal Relationship of Maternal Height on Birth Size and Gestational Age at Birth: A Mendelian Randomization Analysis. *PLoS Med,* 12**,** e1001865.

Supplementary tables

Phenotype	Analysed Sample (n=549 trios)	Excluded Sample (n=398 trios)	p-value
Maternal Height (cm)	165.0 (6.4)	165.1 (6.2)	0.776
Maternal Weight (kg)	76.3 (12.6)	75.0 (15.1)	0.169
Gestational Duration (weeks)	40.1 (1.2)	40.1 (1.2)	0.793
Birthweight (g)	3570 (444)	3537 (436)	0.228
Maternal Age (years)	30.3 (5.3)	30.6 (5.1)	0.509
Maternal smoking status (%Yes)*	14.6	10.8	0.133
Parity (%1st pregnancy)*	44.8	46.6	0.507
Sex of the baby (% Male)*	52.2	51.1	0.932

Table S2: Key characteristics of excluded participants and results of t-tests and chi-square tests for comparison between the analysed sample and the excluded sample.

* indicates the p-value is for a Chi Square test

Outcome Variable	Genetic score	Change in trait per 1 SD higher genetic score	95% CI	Pearson's Correlation coefficient, r	p-value	n
Offspring birthweight adjusted for sex and gestational age	Maternal genetic score for offspring birthweight (g), with weights adjusted for fetal genotype effects	80.4g	43.7,117.1	0.18	2.03e-05	549
Offspring birthweight adjusted for sex and gestational age	Fetal genetic score for birthweight (g), with weights adjusted for maternal genotype effects	68.0 g	31.3,104.9	0.15	0.000303	549
Father's own self-reported birthweight	Paternal genetic score for father's own birthweight (generated with unadjusted weights)	59.2g	27.8, 90.7	0.17	0.00024	504

Table S3: Associations of maternal, fetal and paternal genetic scores with birthweight

Variable	Change in birthweight (g) per 1 SD change in X variable	95% Confidence Interval	t value	p-value
Intercept	3727	3654, 3778	142.9	<0.001
Maternal age	-44	-81, -7	-2.3	0.02
Maternal weight	82	41, 122	4.0	<0.001
Mother's smoking status	-237	-350, -123	-4.1	<0.001
Parity	-219	-291, -147	-6.0	<0.001
Mother's fasting glucose at 28 weeks' gestation	105	67, 144	5.3	<0.001
Mother's own birthweight	103	66, 140	5.5	<0.001
Father's own birthweight	47	10, 83	2.5	0.01
Maternal height	23	-16, 62	1.2	0.25
Paternal height	60	23, 98	3.2	0.002

Table S4: Results of a multivariable linear regression model testing the association between birthweight (adjusted for sex and gestational age), maternal clinical characteristics (n=425 parent-offspring trios), and additional parental anthropometric features that capture fetal genetics. R^2=0.317; Adj-R^2=0.302

Variable	Change in birthweight (g) per 1 SD change in X variable	95% Confidence Interval	t value	p-value
Intercept	3870	3724, 4016	51.9	<0.001
Maternal age	-43	-80, -6	-2.3	0.02
Maternal weight	81	41, 122	4.0	<0.001
Mother's smoking status	-230	-343, -117	-4.0	<0.001
Parity	-226	-298, -154	-6.1	<0.001
Mother's fasting glucose at 28 weeks' gestation	105	66, 143	5.3	<0.001
Mother's own birthweight	94	57, 132	4.9	<0.001
Father 's own birthweight	46	9, 82	2.5	0.01
Maternal height	23	-16, 61	1.1	0.25
Paternal height	60	23, 97	3.2	0.002
Fetal genetic score for offspring birthweight (adjusted for maternal effects)	32	-4, 67	1.8	0.080

Table S5: Results of a multivariable linear regression model testing the association between birthweight (adjusted for sex and gestational age), maternal clinical characteristics (n=425 parent-offspring trios), additional features that capture fetal genetics, and fetal genetic score. R^2 = 0.329; Adj-R^2=0.310, (p=0.06 when compared to Model in table S4)

Included sample (of those with birthweight and complete phenotype data available)	Total N	Description of model	Change in BW in g per 1 SD higher X variable (95%CI)	R^2 (95%CI)
Max sample with maternal genetic score	820	Outcome: birthweight (adjusted for sex and gestational age) X variable: maternal genetic score	66 (32-100)	0.02432 (0.00513, 0.0502)
Max sample with fetal genetic score	646	Outcome: birthweight (adjusted for sex and gestational age) X variable: fetal genetic score	84 (50-117)	0.03634 (0.0131, 0.0710)
Max sample with maternal and fetal genetic scores	595	Outcome: birthweight (adjusted for sex and gestational age) X variable: maternal genetic score	73 (38-108)	0.02935 (0.00733, 0.0595)
Max sample with maternal and fetal genetic scores	595	Outcome: birthweight (adjusted for sex and gestational age) X variable: fetal genetic score	78(43-113)	0.03307 (0.00941, 0.0654)
Max sample with maternal, fetal and paternal genetic scores	549	Outcome: birthweight (adjusted for sex and gestational age) X variable: maternal genetic score	80 (44-117)	0.036 (0.00966, 0.0697)
Max sample with maternal, fetal and paternal genetic scores	549	Outcome: birthweight (adjusted for sex and gestational age) X variable: fetal genetic score	68 (31-105)	0.0269 (0.00497, 0.0557)

Table S6: Summary of models describing the contribution of the genetic scores to variation in offspring birthweight in different sample sizes.

Chapter 4

The contributions of genetic scores, maternal glycemia, and other maternal characteristics to variation in birth weight in South Asian compared with European babies

The contributions of genetic scores, maternal glycemia, and other maternal characteristics to variation in birth weight in South Asian compared with European babies

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Abstract

Aim: Babies of South Asian (SA) ancestry have lower birth weights compared with those of European ancestry, despite higher mean maternal glucose levels. We aimed to compare the associations with birthweight of fetal (fetal_GS) and maternal (maternal_GS) genetic scores (GS) and routinely available clinical features between these two populations, as well as the contributions of the different predictors to birthweight variance explained.

Methods: We analysed mother-child pairs of SA (n=1177) and European (n=1259) ancestry from the Born in Bradford study, excluding type1 and type2 diabetes, preterm and multiple births. We compared multivariable linear regression models to explain variance in birthweight adjusted for sex and gestational age: Model1 (maternal 28-week fasting plasma glucose [FPG], age, weight, smoking, parity, Systolic Blood Pressure [SBP], Diastolic Blood Pressure [DBP], height); Model2 (Model1+maternal_GS+fetal_GS). We used the R-squared statistic to assess the contributions of different predictors in the two samples.

Results: Mean (SD) birthweight was lower in SAs compared with Europeans (3193 (445) g vs. 3475 (483) g; p<0.001), while mean FPG was higher (4.6 (0.5) vs. 4.4 (0.4) mmol/L, p<0.001). Associations between maternal FPG and adjusted birthweight were similar in both populations (62g [95%CI: 35, 88] higher birthweight per 1 SD higher FPG in SA vs. 81g [40, 123] higher birthweight per 1 SD higher FPG in European; variance explained 2.2% v 3.3%, respectively). Other

characteristics showed similar associations with birthweight in both populations, though the distributions of smoking and parity were quite different. GS contributed additional information in both populations, but the full model explained less overall variation in birthweight in SA pregnancies compared with European (Adj-R^2 = 0.175 vs 0.223).

Conclusions: Genetic scores and maternal characteristics show similar associations with birthweight in babies of SA and European ancestry. Further work is needed to understand what contributes to birthweight differences between SA and Europeans as this may ultimately inform antenatal care.

Introduction

Birth weight varies across different ethnicities; African, Indian and East Asian populations note a lower birthweight on average compared to white Americans and Europeans (Barron, 1983). In the UK, healthy babies of White ethnicity weigh about 3500 g on average and those of Indian, Pakistani and Bangladeshi ethnicity weigh between 280-350g lighter, those of Black Caribbean ancestry are 150 g lighter and those of Black African ancestry are 70 g lighter (Kelly et al., 2009).

A previous study (West et al., 2013) has shown that babies of South Asian (SA) ancestry have lower birth weight than babies of European (Eur) ancestry. It has been shown that some of this difference in birth weight can be explained by the mother's height and weight as South Asian women are shorter and weigh lighter on average. When further comparisons were made between the two ethnicities, fewer South Asian mothers smoked, their fasting glucose level was higher on average, and there was a higher proportion of SA women who had delivered multiple babies.

These differences did not fully account for the difference in birth weight between these two groups. To further investigate the differences in birthweight between these two ethnicities, birthweight, skinfold thickness and cord leptin (West et al., 2013) were compared. Results showed that similar skinfold thickness was observed in both groups but SA babies had greater total fat mass. While maternal height, BMI and gestational age were able to explain some of the differences, glucose, parity, smoking and living with a partner masked the difference in a way that the overall effect on birth weight was negligible (West et al.).

Genetic variation from both mother and fetus contributes to variation in birth weight. Genome-wide association studies have now identified more than 240 regions of the genome where common variants are robustly associated with birth weight(Juliusdottir et al., 2021). The vast majority of the data contributing to GWAS of birth weight to date has been from people of European ancestry. However, a recent large meta-analysis of individuals of Indian and Bangladeshi ancestry from both the UK and Indian subcontinent showed that a fetal genetic score was

strongly associated with birth weight with a similar effect to that in Europeans, despite large differences in mean maternal BMI and mean birth weight. In that study, there was, however, a weaker effect of a maternal genetic score on birth weight in the South Asian studies compared with European, which may indicate different intrauterine exposures (Nongmaithem et al., 2022).

Our previous work has demonstrated that both maternal and fetal genetic scores explained further variation in offspring birth weight on top of clinical variables (Haulder et al.) in a UK study of participants with European ancestry. Since genetic scores identified in Europeans show similar associations with birth weight in samples of European and South Asian ancestry, we would not expect them to explain the significant differences in birth weight that are seen between these two groups. However, it is not known whether these genetic scores explain a similar amount of variation in birth weight when added to maternal characteristics in South Asians, as they do in Europeans.

In this paper, we explored the associations with offspring birthweight and clinical features in two groups of different ethnicities and further explored variables that have not been investigated previously. We used the Born in Bradford (BiB) study (Wright et al., 2013) which comprises of two different ethnic groups living in the same area: White Europeans and South Asians (Bangladeshis and Pakistanis) based in the city of Bradford in the UK, with data collected at the same time. This was a replication of our first study in a different European cohort and, in addition this time, to a South Asian group. We then included a maternal and fetal genetic score on top of clinical features to investigate the contributions of genetics in explaining variability in birthweight in the two groups.

Methods

Study population

We used data from the Born in Bradford Study (BiB). The Born in Bradford cohort represents the obstetric population in Bradford, which is a city in North of England and contains data collected from 12453 women (with 13776 pregnancies) between 2007 and 2010. About 20% of the current generation of parents is of South Asian origin (90% are from Mirpur in Pakistan)(Wright et al., 2013).

Inclusion criteria for the current analyses consisted of only those mother-child pairs where complete clinical, anthropometric, and maternal and fetal genetic data were available. Most pairs had complete phenotype data, but following genotype quality control, and owing to lower availability of fetal DNA from cord blood compared with parental DNA, complete genotype data was available for mothers and offspring in 50% of the pairs in both ancestries. We excluded participants with preterm and multiple births as well as pre-existing type 1 or type 2 diabetes.

We created two separate datasets for the Europeans (UK) ancestry and the South Asians (Pakistani) ancestry. The final datasets consisted of 1259 mother-child pairs for the Europeans and 1177 mother-child pairs for the South Asians, respectively. The selection of variables is illustrated by the flowchart in Figure 1 and Figure 2. To check for any differences between excluded and included participants, we used unpaired t-tests to compare means of continuous variables (maternal height, maternal weight, gestational duration, birth weight and maternal age), and Chi-square tests to compare categorical variables (maternal smoking status, parity, and sex of the baby).

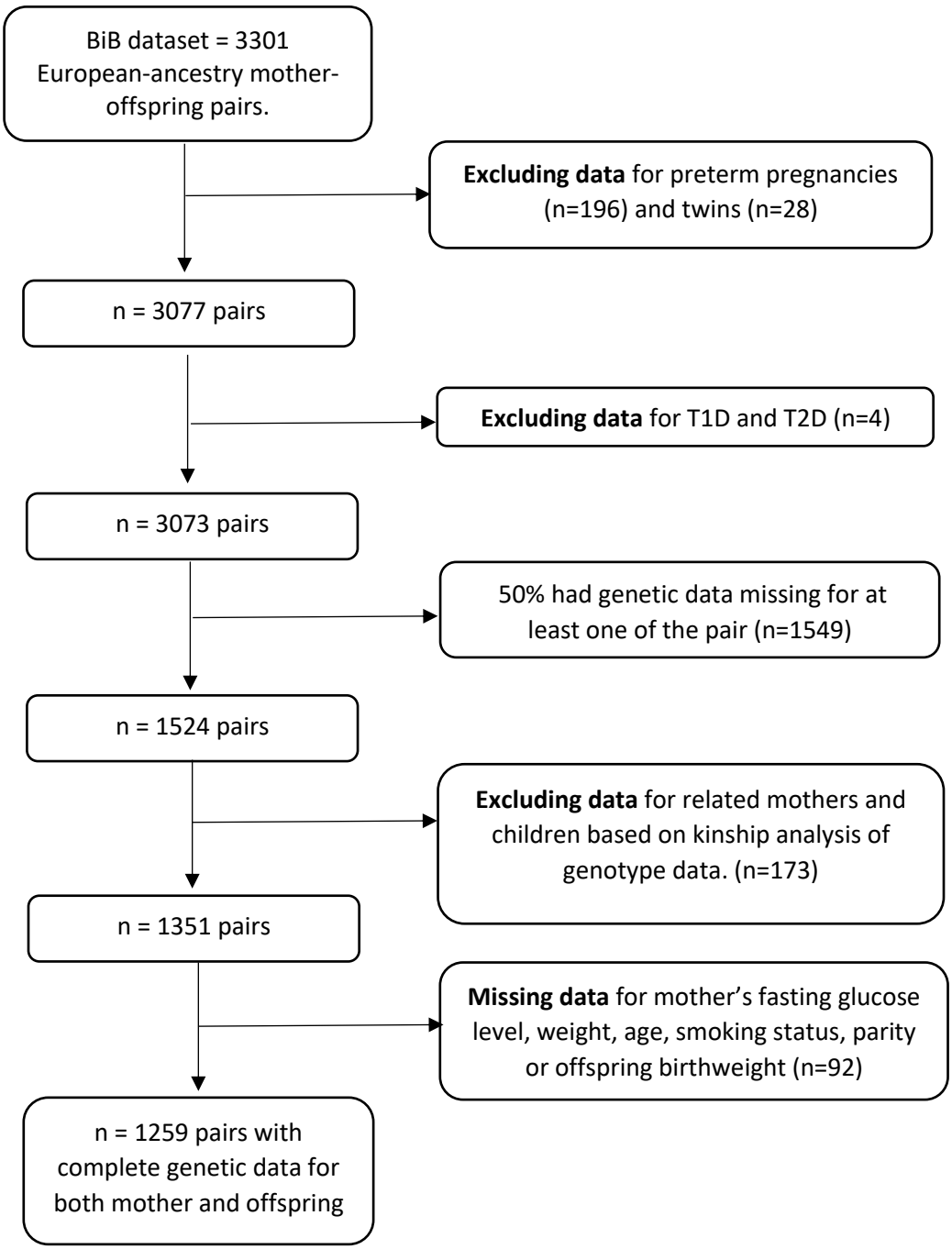


Fig 1: Flowchart illustrating how the European data was prepared for analysis

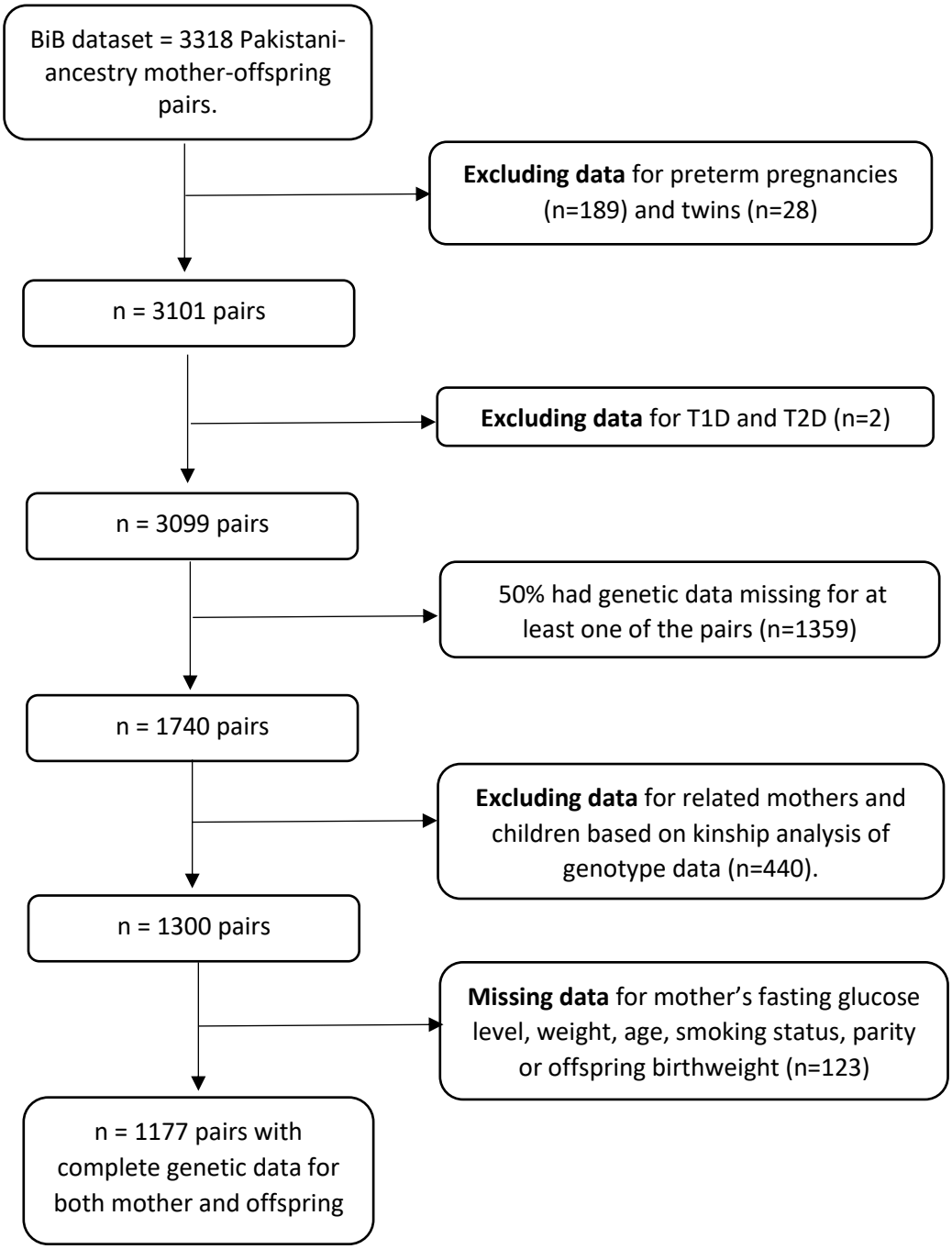


Fig 2: Flowchart illustrating how the Pakistani data was prepared for analysis

Characteristics of participants

Full details of data collection are found in the BiB study protocol(Wright et al., 2013). Participants were recruited at their oral glucose tolerance test (OGTT) appointment between 26 to 28 weeks' gestation. Briefly, detailed anthropometric measurements and biochemistry from the mothers were taken at recruitment. Maternal height was measured to the nearest 0.1 cm. Maternal weight was measured to the nearest 100g. Offspring birth weight was measured at birth, to the nearest 10g. For the current analyses, birth weight was adjusted for sex and gestational age by saving residuals on a regression model of birthweight against sex and gestational age.

Maternal glucose was measured in fasting maternal samples offered at the OGTT appointment. Information about the mother's smoking status and parity was self-reported. The ethnicity of the parents was determined by genetic similarity for the analysis. The systolic blood pressure (SBP) and diastolic blood pressure (DBP) measurements were taken closest to 28 weeks' gestation and if that measurement was not available, the median value between 27 and 29 weeks of gestation was recorded as the blood pressure measurement.

Genotyping

Two separate microarrays, the Illumina HumanCoreExome array and the Infinium Global Screening array (GSA), were used to obtain maternal and fetal genomic data. Genotype data had been already imputed against the Haplotype Reference Consortium panel, version r1.1 using Minimac4, after quality control (minor allele frequency > 1% and Hardy-Weinberg Equilibrium P value > 1×10^{-6}).

Statistical Analyses

Genetic scores

We created independent maternal and fetal genetic scores for birth weight. We calculated the genetic scores (GS) according to Equation 1, where w_i is the weight for SNP i and g_i is the genotype at SNP i.

$$\boldsymbol{GS = \sum_i w_i g_i} \quad \textbf{(1)}$$

A total of 209 SNPs, identified at 190 loci in a recent GWAS of birth weight (Warrington et al., 2019), were used to calculate the maternal and fetal genetic scores (see **Table S1)**. Effect estimates for each SNP were used as weights, and for the maternal score, these had been adjusted to represent the maternal effects adjusted for fetal genotype effects using a structural equation model (Warrington et al., 2019). For the fetal score, fetal effect estimates adjusted for maternal genotype effects were used as weights. Each genetic score variable was then standardized using its mean and standard deviation. To validate the genetic scores, we tested the associations between each standardized genetic score and its respective phenotype using simple linear regression models.

Linear regression models to estimate variance in corrected birth weight explained by clinical, anthropometric, and genetic factors

To compare associations of birthweight with maternal characteristics and the genetic scores between the European and South Asian samples of mothers and babies from the Born in Bradford study, we examined the following multivariable linear regression models, with corrected birth weight (i.e., adjusted for sex and gestational age) as the outcome variable.

Models 1a and 1b: Maternal clinical and anthropometric characteristics: maternal fasting glucose, age, weight, height, parity, SBP, DBP and the mother's smoking status were used in the first model.

Models 2a and 2b: Fetal genetic score + maternal genetic score + maternal clinical and anthropometric characteristics: the maternal and fetal genetic scores were added to Models 1a and 1b to further investigate the contributions of the genetic scores in addition to maternal height and clinical features.

We used the adjusted R-squared statistic to assess improvement in model fit based on any added predictors. We used the R-squared statistic for assessing individual contributions of each variable to birthweight in univariate regression models. An F-test was used to compare nested models and check for any improvements in the explanation of variance in birth weight upon adding the genetic scores. The R-squared statistic and its 95% confidence intervals were used to assess the overall explanation of variance in birth weight by the predictors in the model. Multicollinearity between predictor variables in the models was checked by using the Variance Inflation Factor (VIF).

Results

Descriptive characteristics for the mother-child pairs in the European (UK) ancestry and South Asian (Pakistani) ancestry are shown in Table 1. There was no robust evidence that individuals excluded from the analysis differed in their basic characteristics from those included (see **Table S2 and S3**).

	European (UK) n=1259 mother-child pairs	South Asian (Pakistani) n=1177 mother-child pairs	p-value
Phenotype	**Mean or % (SD)**	**Mean or % (SD)**	
Birth weight (g)	3475(483)	3193(445)	<0.001
Maternal Height (cm)	164.6 (6.2)	159.5(5.6)	<0.001
Maternal Weight (kg)	79.0(16.4)	70.6(13.1)	<0.001
Gestational Duration (weeks)	39.6(1.2)	39.4(1.2)	<0.001
Maternal fasting glucose (mmol/L)	4.4(0.4)	4.6(0.5)	<0.001
Maternal Systolic BP(mmHg)	113.1(11.4)	107.1(11.1)	<0.001
Maternal Diastolic BP(mmHg)	66.6(8.6)	63.7(8.1)	<0.001
Maternal Age (years)	27.2(6.0)	28.0(5.1)	<0.001
Maternal smoking status (%Yes)	31.3	4.0	<0.001
Parity (%1st pregnancy)	49.6	33.9	<0.001
Parity (%2nd pregnancy)	32.5	24.9	
Parity (%More than two pregnancies)	17.9	41.2	
Sex of the baby (% Male)	51.9	52.4	0.837

Table 1: Key characteristics of the study populations

Most characteristics showed differences, on average, between the two samples. For example, in the South Asian group, maternal fasting glucose was higher (4.6 mmol/L) as compared to the Europeans group (4.4 mmol/L). We also observed that in the South Asian group, fewer mothers smoked during the pregnancy (4%) when compared to the Europeans (31.3%). We observed that there were more women from the South Asian ancestry who had given birth more than once (41.2%) than in the European group (17.9%). The genetic scores showed strong associations with birthweight (**Table 2**).

Ethnicity	Genetic score	Change in offspring birth weight adjusted for sex and gestational age per 1 SD higher genetic score (95% CI)	Pearson's Correlation coefficient, r	p-value
European (n=1259)	Maternal genetic score for offspring birthweight (g), with weights adjusted for fetal genotype effects	36 (9, 63)	0.074	0.008
	Fetal genetic score for birthweight (g), with weights adjusted for maternal genotype effects	96 (69,122)	0.20	<0.001
Pakistani (n=1177)	Maternal genetic score for offspring birthweight (g), with weights adjusted for fetal genotype effects	25 (-1,51)	0.069	0.06
	Fetal genetic score for birthweight (g), with weights adjusted for maternal genotype effects	79 (54,105)	0.19	<0.001

Table 2: Associations of the genetic scores with offspring birthweight.

Similar associations were observed between maternal clinical characteristics and offspring birthweight in Europeans and South Asians

Multivariable linear regression models (**Model 1a & 1b; Table 3)** including clinical and anthropometric variables (maternal fasting glucose, maternal age, maternal weight, parity, mother's systolic blood pressure, mother's diastolic blood pressure, mother's height and the mother's smoking status), showed that each variable except for SBP, DBP and maternal age, contributed significantly to variance explained in birthweight in the European and Pakistani samples. In the Pakistani sample, the mother's smoking status was also found to not contribute to explaining variation in birthweight: there was no strong association between smoking status and birthweight.

Similar associations were observed in both ancestries (the coefficient of one sample is within the 95% CI of the coefficient for the other sample) except for the maternal smoking status, which had less influence on birthweight in the South Asian group as compared with the Europeans. This can be explained by the fact that fewer mothers smoked in the South Asian group. The contributions to variation of birthweight by maternal fasting glucose is similar in both populations.

	Europeans (UK)		South Asian (Pakistani)	
Variable	**Change in birthweight (g) per 1 SD change in independent continuous variable**	**95% CI**	**Change in birthweight (g) per 1 SD change in independent continuous variable**	**95% CI**
Maternal age(years)	-25	-53, 4	-21	-51, 9
Maternal weight(kg)	106	76, 135	96	67, 124
Mother's smoking status*(smoker vs nonsmoker)	-259	-314, -203	-23	-150, 105
Parity(1^{st} pregnancy vs 3+ pregnancies)**	-169	-242, -96	-173	-240, -106
Parity(2^{nd} pregnancy vs 3+ pregnancies)**	-23	-96, 50	-77	-141, -13
Mother's fasting glucose at 28 weeks' gestation (mmol/L)	62	35, 88	81	40, 123
Maternal Systolic Blood Pressure (mmHg)	-7	-36, 22	12	-19, 43
Maternal Diastolic Blood Pressure(mmHg)	-15	-44, 13	-20	-52, 11
Maternal height(cm)	56	30, 83	74	48, 100

* indicates a binary variable

**indicates a multilevel variable

Table 3: Results of multivariable linear regression models testing the association between birthweight (adjusted for sex and gestational age), maternal characteristics Model 1a: Eur (n=1259 mother-child pairs) R^2 = 0.1877; Adj-R^2= 0.1818

Model 1b: SA (n=1177 mother-child pairs). R^2 =0.1443; Adj-R^2= 0.1374

Maternal genetic score and fetal genetic score further improved variance explained in birthweight when added to maternal clinical and anthropometric variables

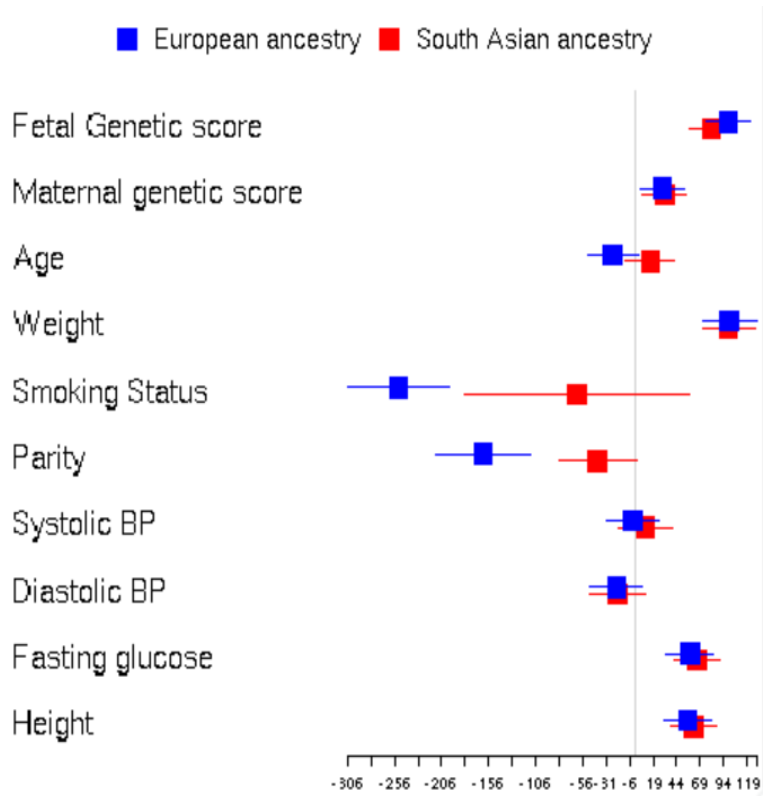
When the maternal genetic score and the fetal genetic scores were added on top of clinical variables and maternal height (**Model 2a & 2b; Table 4**), there were additional improvements in explanation of variance in birthweight in both ancestries (Eur: Adj-R^2= 0.2233 vs 0.1818 p<0.001; SA: Adj-R^2=0.1665 vs 0.1374, p<0.001 **Table 5**)

	Europeans (UK)		South Asian (Pakistani)	
Variable	**Change in birthweight (g) per 1 SD change in independent variable**	**95% CI**	**Change in birthweight (g) per 1 SD change in independent variable**	**95% CI**
Maternal age(years)	-25	-52, 3	-19	-49, 10
Maternal weight(kg)	101	72, 130	92	64, 121
Mother's smoking status*(smoker vs nonsmoker)	-254	-308,-200	-25	-150, 101
Parity(1st pregnancy vs 2+ pregnancies)**	-164	-235, -93	-172	16, 141
Parity(2nd pregnancy vs 2+ pregnancies)**	-6	-78, 65	-83	-157, -31
Mother's fasting glucose at 28 weeks' gestation(mmol/L)	65	39, 91	103	50, 132
Maternal height (cm)	56	31, 82	70	45, 96
Maternal Systolic Blood Pressure(mmHg)	-5	-33, 23	8	-23, 39
Maternal Diastolic Blood Pressure(mmHg)	-18	-46, 10	-20	-51, 11
Fetal Genetic Score for Offspring birth weight (per unit GS)	98	74, 122	75	51, 100
Maternal Genetic Score for Offspring birth weight (per unit GS)	30	6, 54	25	1, 49

Table 4: Results of a multivariable linear regression model testing the association between birthweight (adjusted for sex and gestational age), maternal clinical characteristics, maternal height, and maternal and fetal genetic scores.

Model 2a: Eur (n=1259 mother-child pairs) R^2 = 0.2301; Adj-R^2=0.2233
Model 2b: SA (n=1177 mother-child pairs) R^2 =0.1747; Adj-R^2=0.1665

The addition of a maternal genetic score and a fetal genetic score results in an increase in the R-Squared statistic in both the European (R-Squared: 0.2301 vs 0.1877) and Pakistani (R-Squared: 0.1747 vs 0.1443) cohort. The associations of the genetic scores were similar in both groups (the coefficient of one sample is within the 95% CI of the coefficient for the other sample).



Change in birthweight (g) per 1 SD higher continuous trait. For binary traits: change in birthweight (g) associated with smoking vs. non-smoking, or first birth vs. subsequent (parity).

Figure 2: Forest plot illustrating the associations between maternal characteristics and adjusted birthweight in Eur vs SA

Note: The parity variable is Primiparity compared with delivering 2 or more babies**
Error bars represent 95% CI

Associations between most maternal characteristics and adjusted birthweight were similar in both samples, though smoking status and parity were less strongly associated in South Asians than in Europeans **(Figure 2).**

The genetic scores and maternal characteristics together explained less variation in birthweight in the SA pregnancies compared with the European pregnancies

	Clinical Features (Adj-R^2)	Clinical Features + Genetics (Adj-R^2)	p-value
South Asian Ancestry	0.137	0.175	2×10^{-15}
European Ancestry	0.182	0.223	2×10^{-11}

Table 5: Summary of Adj-R^2 for the models.

The maternal and fetal genetic scores explained further variability in adjusted birthweight in addition to the clinical features. However, in the South Asian group, the genetic scores together with the maternal characteristics explained less variation than in the European group (Adj-R^2 = 0.175 vs 0.223). A summary of the associations of maternal characteristics, maternal and fetal genetics with adjusted birthweight across the two populations is shown in **Figure 2.**

The R-Squared statistic for each variable was similar in Europeans and Pakistanis with the only notable difference being in the smoking status

R-Squared	European	Pakistani
Smoking Status	0.0667	9.24e-06
Primiparity	0.0240	0.0297
Maternal fasting glucose	0.0330	0.0223
Maternal Weight	0.0904	0.0927
Maternal Height	0.0361	0.0482
Mother's Systolic Blood Pressure	0.0104	0.00897
Mother's Diastolic Blood Pressure	0.00775	0.00399
Mother's genetic score	0.00546	0.00311
Fetal genetic score	0.0391	0.0320

Table 6: Summary of R-squared values from univariate linear regression for each variable.

The results show that the R-Squared statistic for univariate analysis between birthweight (corrected for sex and gestational age) is similar for all the predictors used in the multivariable regression models. The smoking status notes a considerable difference in explaining variation in birthweight between the two groups with a higher contribution noted among Europeans.

Discussion

We wanted to explore the contribution to offspring birthweight of routinely available maternal characteristics alongside the maternal and fetal genetic scores in these two groups despite the difference in mean birthweights.

Although maternal FPG levels were, on average, higher in SA than in Europeans, the associations between maternal FPG and adjusted birthweight were similar in both populations (SA v European regression coefficient 81g [95%CI: 40, 123] per 1 SD higher FPG vs. 62g [35, 88] per 1 SD higher FPG; variance explained 2.2% v 3.3%). Most other characteristics showed similar associations with birthweight in both populations. We used the R-Squared statistic to assess the contribution of each predictor to birthweight and we observed that most variables had similar level of contribution in both ethnicities to variation in offspring birth weight, but the smoking status had little contribution in the South Asian group, due to its low prevalence (Table 5). We also observed that the addition of a maternal genetic score and a fetal genetic score for offspring birth weight to routinely available clinical features explained additional variation in birth weight (SA: Adj-R^2 = 0.137 vs 0.167; Eur: Adj-R^2 = 0.182 vs 0.223) in both groups.

Our results are similar to those of our previous study on a European ancestry cohort in the southwest of the UK, a more affluent setting, where we showed that adding maternal and fetal genetic scores to readily available clinical features resulted in the improvement of explanation of variation in offspring birth weight(Haulder et al., 2022).

Our current results show that findings are also consistent in a sample of SA ancestry based in the UK. Previously, the fetal genetic score for birth weight was shown to have similar associations to offspring birth weight in both SA and Europeans(Nongmaithem et al., 2022), but in that study, the maternal genetic score from a meta-analysis, was more weakly associated with birthweight in the South Asians when compared to the Europeans. The reason suggested for this was exposures to different intrauterine environments in the Europeans and SAs. Contrary to this study, we found similar associations between Europeans and South Asians for the maternal genetic score when it was considered in a model

along with other maternal characteristics in two groups found in the same environment. This result matches the point made before that when subjected to the same maternal intrauterine environment, maternal genetic score does in fact contribute to birthweight in SA to the same extent as in Europeans. The sample sizes in the SA groups for the meta-analyses were greater than in this study and therefore, had more power. The difference we observed could also be because the sample used in this study was in the UK and the others were sometimes non-UK. Previous research has shown that the difference in birthweight between these two populations is not explained by the variables that have different associations (mother's smoking status, parity and maternal fasting glucose level) (West et al., 2013). While the height and weight of SA mothers are less than those of European mothers, the study also showed that the difference in birthweight was not fully explained by the differences in height and weight either.

We can observe that although the genetic scores were created using genetic information from GWAS of European ancestry, the contribution of the genetic scores to birth weight in the SA group was similarly informative as in the European group (mGS R-Squared: Eur vs SA; 0.00546 vs 0.00311; fGS R-Squared: Eur vs SA; 0.0391 vs 0.0320; **Table 6**).

This is further supported by the fact that the genetic scores were similarly associated to birth weight in SA as in Europeans (**Figure 2**) and brings us to the conclusion that the underlying difference in birth weight between these two populations is not due to common variants associated with birth weight from GWAS.(Nongmaithem et al., 2022)

Further work needs to be carried out to study the underlying mechanisms that cause birth weight in South Asians to be less than that in Europeans. For example, the contribution to birth weight of the genetic scores of favorable adiposity could be compared in both ethnicities by using multivariable regression models. Adiposity is defined as body fat, and it is more desirable for it to be stored under the skin than around organs such as the liver and the heart. A study showed that Indians tend to have central adiposity and higher body fat as compared to other ethnic groups and are also at high risk of insulin (Yajnik, 2001)resistance syndrome and more

susceptible to coronary diseases and diabetes Genes dictate where this fat is stored, and this can then be termed as having “favorable adiposity” or “unfavorable adiposity.” A previous study (Thompson et al., 2022)has shown that fetal alleles that have a positive influence on favorable adiposity are associated with higher birth weight.

Previous studies (Thompson et al., 2021)have used two sample-MR with GWAS summary statistics to estimate the causal effects of maternal metabolically favorable adiposity on offspring birthweight and have found higher maternal adiposity to result in lower offspring birthweight if accompanied by a favorable metabolic profile. It has also been shown before(Nightingale et al., 2011) that adiposity patterns in South Asians differ from those in Europeans. This could be useful in exploring if adiposity is linked to the difference in birthweight between South Asians and Europeans.

Conclusion

The results of our study show that genetics help explain additional variability in offspring birth weight on top of clinical features in both Europeans and South Asians. This implies there is a possibility genetic scores could be added to prediction models. Further work needs to be done to investigate what are the underlying mechanisms that contribute to the discrepancy in offspring birth weight for these two groups.

Supplementary Information

	European (UK) mother-child pairs (n=2042)	p-value
Phenotype	**Mean or % (SD)**	
Birth weight (g)	3276(606)	<0.001
Maternal Height (cm)	164.2(6.2)	0.04
Maternal Weight (kg)	77.3(16.0)	0.003
Gestational Duration (weeks)	39.0(2.1)	<0.001
Maternal fasting glucose (mmol/L)	4.4(0.4)	0.0556
Maternal Systolic BP (mmHg)	112.8(11.5)	0.596
Maternal Diastolic BP (mmHg)	66.8(8.3)	0.504
Maternal Age (years)	26.5(6.0)	0.0009
Maternal smoking status (%Yes)	34.3	0.061
Parity (%1st pregnancy vs 2+ pregnancy)	18.7	0.094
Parity(%2nd pregnancy vs 2+ pregnancy)	54.0	
Parity (2+ pregnancies)	27.3	
Sex of the baby (% Male)	50.6	0.565

Table S2: Key characteristics of the excluded Europeans

	South Asian (Pakistani) mother-child pairs (n=2201)	p-value
Phenotype	**Mean or % (SD)**	
Birth weight (g)	3082(570)	<0.001
Maternal Height (cm)	160.0(5.8)	0.0364
Maternal Weight (kg)	71.8(14.1)	0.0124
Gestational Duration (weeks)	38.9(2.0)	<0.001
Maternal fasting glucose (mmol/L)	4.6(0.7)	<0.001
Maternal Systolic BP (mmHg)	108.1(11.3)	0.0224
Maternal Diastolic BP (mmHg)	64.6(8.5)	0.0858
Maternal Age (years)	27.9(5.3)	0.3441
Maternal smoking status (%Yes)	3.4	
Parity (%1st pregnancy vs 2+ pregnancy)	36.7	0.075
Parity(%2nd pregnancy vs 2+ pregnancy)	25.1	
Parity (2+ pregnancies)	38.1	
Sex of the baby (% Male)	50.2	0.675

Table S3: Key characteristics of the excluded Pakistani

References

ABRAHAM, G. & INOUYE, M. 2014. Fast principal component analysis of large-scale genome-wide data. *PLoS One,* 9**,** e93766.

AHKTER, J., QURESHI, R., RAHIM, F., MOOSVI, S., REHMAN, A., JABBAR, A., ISLAM, N. & KHAN, M. A. 1996. Diabetes in pregnancy in Pakistani women: prevalence and complications in an indigenous south Asian community. *Diabet Med,* 13**,** 189-91.

AMARK, H., WESTGREN, M. & PERSSON, M. 2019. Prediction of large-for-gestational-age infants in pregnancies complicated by obesity: A population-based cohort study. *Acta Obstet Gynecol Scand,* 98**,** 769-776.

AMBLER, G., OMAR, R. Z., ROYSTON, P., KINSMAN, R., KEOGH, B. E. & TAYLOR, K. M. 2005. Generic, simple risk stratification model for heart valve surgery. *Circulation,* 112**,** 224-31.

ANAND, S. S., GUPTA, M. K., SCHULZE, K. M., DESAI, D., ABDALLA, N., WAHI, G., WADE, C., SCHEUFLER, P., MCDONALD, S. D., MORRISON, K. M., VASUDEVAN, A., DWARAKANATH, P., SRINIVASAN, K., KURPAD, A., GERSTEIN, H. C. & TEO, K. K. 2016. What accounts for ethnic differences in newborn skinfold thickness comparing South Asians and White Caucasians? Findings from the START and FAMILY Birth Cohorts. *Int J Obes (Lond),* 40**,** 239-44.

BARKER, D. J. & OSMOND, C. 1986. Infant mortality, childhood nutrition, and ischaemic heart disease in England and Wales. *Lancet,* 1**,** 1077-81.

BARRON, S. L. 1983. Birthweight and ethnicity. *Br J Obstet Gynaecol,* 90**,** 289-90.

BATTERSBY, C., STATNIKOV, Y., SANTHAKUMARAN, S., GRAY, D., MODI, N., COSTELOE, K., COLLABORATIVE, U. K. N. & MEDICINES FOR NEONATES INVESTIGATOR, G. 2018. The United Kingdom National Neonatal Research Database: A validation study. *PLoS One,* 13**,** e0201815.

BEAUMONT, R. N., WARRINGTON, N. M., CAVADINO, A., TYRRELL, J., NODZENSKI, M., HORIKOSHI, M., GELLER, F., MYHRE, R., RICHMOND, R. C., PATERNOSTER, L., BRADFIELD, J. P., KREINER-MOLLER, E., HUIKARI, V., METRUSTRY, S., LUNETTA, K. L., PAINTER, J. N., HOTTENGA, J. J., ALLARD, C., BARTON, S. J., ESPINOSA, A., MARSH, J. A., POTTER, C., ZHANG, G., ANG, W., BERRY, D. J., BOUCHARD, L., DAS, S., EARLY GROWTH GENETICS, C., HAKONARSON, H., HEIKKINEN, J., HELGELAND, O., HOCHER, B., HOFMAN, A., INSKIP, H. M., JONES, S. E., KOGEVINAS, M., LIND, P. A., MARULLO, L., MEDLAND, S. E., MURRAY, A., MURRAY, J. C., NJOLSTAD, P. R., NOHR, E. A., REICHETZEDER, C., RING, S. M., RUTH, K. S., SANTA-MARINA, L., SCHOLTENS, D. M., SEBERT, S., SENGPIEL, V., TUKE, M. A., VAUDEL, M., WEEDON, M. N., WILLEMSEN, G., WOOD, A. R., YAGHOOTKAR, H., MUGLIA, L. J., BARTELS, M., RELTON, C. L., PENNELL, C. E., CHATZI, L., ESTIVILL, X., HOLLOWAY, J. W., BOOMSMA, D. I., MONTGOMERY, G. W., MURABITO, J. M., SPECTOR, T. D., POWER, C., JARVELIN, M. R., BISGAARD, H., GRANT, S. F. A., SORENSEN, T. I. A., JADDOE, V. W., JACOBSSON, B., MELBYE, M., MCCARTHY, M. I., HATTERSLEY, A. T., HAYES, M. G., FRAYLING, T. M., HIVERT, M. F., FELIX, J. F., HYPPONEN, E., LOWE, W. L., JR., EVANS,

D. M., LAWLOR, D. A., FEENSTRA, B. & FREATHY, R. M. 2018. Genome-wide association study of offspring birth weight in 86 577 women identifies five novel loci and highlights maternal genetic effects that are independent of fetal genetics. *Hum Mol Genet,* 27**,** 742-756.

BELBASIS, L., SAVVIDOU, M. D., KANU, C., EVANGELOU, E. & TZOULAKI, I. 2016. Birth weight in relation to health and disease in later life: an umbrella review of systematic reviews and meta-analyses. *BMC Med,* 14**,** 147.

BIAU, D. J., KERNEIS, S. & PORCHER, R. 2008. Statistics in brief: the importance of sample size in the planning and interpretation of medical research. *Clin Orthop Relat Res,* 466**,** 2282-8.

BIKS, G. A., BLENCOWE, H., HARDY, V. P., GEREMEW, B. M., ANGAW, D. A., WAGNEW, A., ABEBE, S. M., GUADU, T., MARTINS, J. S. D., FISKER, A. B., IMAM, M. A., NETTEY, O. E. A., KASASA, S., DI STEFANO, L., AKUZE, J., KWESIGA, D., LAWN, J. E. & EVERY NEWBORN, I. S. C. G. 2021. Birthweight data completeness and quality in population-based surveys: EN-INDEPTH study. *Popul Health Metr,* 19**,** 17.

BOYD, M. E., USHER, R. H. & MCLEAN, F. H. 1983. Fetal macrosomia: prediction, risks, proposed management. *Obstet Gynecol,* 61**,** 715-22.

BROWN, A. E. & WALKER, M. 2016. Genetics of Insulin Resistance and the Metabolic Syndrome. *Curr Cardiol Rep,* 18**,** 75.

CAUZZO, C., CHIAVAROLI, V., DI VALERIO, S. & CHIARELLI, F. 2023. Birth size, growth trajectory and later cardio-metabolic risk. *Front Endocrinol (Lausanne),* 14**,** 1187261.

CETIN, I., MANDO, C. & CALABRESE, S. 2013. Maternal predictors of intrauterine growth restriction. *Curr Opin Clin Nutr Metab Care,* 16**,** 310-9.

CHOWDHURY, M. Z. I. & TURIN, T. C. 2020. Variable selection strategies and its importance in clinical prediction modelling. *Fam Med Community Health,* 8**,** e000262.

COGSWELL, M. E. & YIP, R. 1995. The influence of fetal and maternal factors on the distribution of birthweight. *Semin Perinatol,* 19**,** 222-40.

COLLINS, G. S., REITSMA, J. B., ALTMAN, D. G. & MOONS, K. G. 2015. Transparent reporting of a multivariable prediction model for individual prognosis or diagnosis (TRIPOD): the TRIPOD statement. *BMJ,* 350**,** g7594.

COLLISTER, J. A., LIU, X. & CLIFTON, L. 2022. Calculating Polygenic Risk Scores (PRS) in UK Biobank: A Practical Guide for Epidemiologists. *Front Genet,* 13**,** 818574.

COORAY, S. D., BOYLE, J. A., SOLDATOS, G., ALLOTEY, J., WANG, H., FERNANDEZ-FELIX, B. M., ZAMORA, J., THANGARATINAM, S. & TEEDE, H. J. 2022. Development, validation and clinical utility of a risk prediction model for adverse pregnancy outcomes in women with gestational diabetes: The PeRSonal GDM model. *EClinicalMedicine,* 52**,** 101637.

COUSTAN, D. R., LOWE, L. P., METZGER, B. E., DYER, A. R., INTERNATIONAL ASSOCIATION OF, D. & PREGNANCY STUDY, G. 2010. The Hyperglycemia and Adverse Pregnancy Outcome (HAPO) study: paving the way for new diagnostic criteria for gestational diabetes mellitus. *Am J Obstet Gynecol,* 202**,** 654 e1-6.

CRAMER, J. C. 1995. Racial and ethnic differences in birthweight: the role of income and financial assistance. *Demography,* 32**,** 231-47.
DE GRAAFF, E., SADLER, L., LAKHDHIR, H., SIMON-KUMAR, R., PEIRIS-JOHN, R., BURGESS, W., OKESENE-GAFA, K., CRONIN, R., MCCOWAN, L. & ANDERSON, N. 2023. Grouping women of South Asian ethnicity for pregnancy research in New Zealand. *Aust N Z J Obstet Gynaecol,* 63**,** 499-508.
DHIMAN, P., MA, J., QI, C., BULLOCK, G., SERGEANT, J. C., RILEY, R. D. & COLLINS, G. S. 2023. Sample size requirements are not being considered in studies developing prediction models for binary outcomes: a systematic review. *BMC Med Res Methodol,* 23**,** 188.
DONG, Y., LUO, Z. C., NUYT, A. M., AUDIBERT, F., WEI, S. Q., ABENHAIM, H. A., BUJOLD, E., JULIEN, P., HUANG, H., LEVY, E., FRASER, W. D. & GROUP, D. C. S. 2018. Large-for-Gestational-Age May Be Associated With Lower Fetal Insulin Sensitivity and beta-Cell Function Linked to Leptin. *J Clin Endocrinol Metab,* 103**,** 3837-3844.
DUTTON, P. J., WARRANDER, L. K., ROBERTS, S. A., BERNATAVICIUS, G., BYRD, L. M., GAZE, D., KROLL, J., JONES, R. L., SIBLEY, C. P., FROEN, J. F. & HEAZELL, A. E. 2012. Predictors of poor perinatal outcome following maternal perception of reduced fetal movements--a prospective cohort study. *PLoS One,* 7**,** e39784.
DWI PUTRA, S. E., REICHETZEDER, C., HASAN, A. A., SLOWINSKI, T., CHU, C., KRAMER, B. K., KLEUSER, B. & HOCHER, B. 2020. Being Born Large for Gestational Age is Associated with Increased Global Placental DNA Methylation. *Sci Rep,* 10**,** 927.
EVERETT, T. R. & CHITTY, L. S. 2015. Cell-free fetal DNA: the new tool in fetal medicine. *Ultrasound Obstet Gynecol,* 45**,** 499-507.
FADISTA, J., MANNING, A. K., FLOREZ, J. C. & GROOP, L. 2016. The (in)famous GWAS P-value threshold revisited and updated for low-frequency variants. *Eur J Hum Genet,* 24**,** 1202-5.
FRAGOSO-BARGAS, N., OPSAHL, J. O., KIRYUSHCHENKO, N., BOTTCHER, Y., LEE-ODEGARD, S., QVIGSTAD, E., RICHARDSEN, K. R., WAAGE, C. W., SLETNER, L., JENUM, A. K., PRASAD, R. B., GROOP, L. C., MOEN, G. H., BIRKELAND, K. I. & SOMMER, C. 2021. Cohort profile: Epigenetics in Pregnancy (EPIPREG) - population-based sample of European and South Asian pregnant women with epigenome-wide DNA methylation (850k) in peripheral blood leukocytes. *PLoS One,* 16**,** e0256158.
FRICK, A. P., SYNGELAKI, A., ZHENG, M., POON, L. C. & NICOLAIDES, K. H. 2016. Prediction of large-for-gestational-age neonates: screening by maternal factors and biomarkers in the three trimesters of pregnancy. *Ultrasound Obstet Gynecol,* 47**,** 332-9.
GARIN, I., EDGHILL, E. L., AKERMAN, I., RUBIO-CABEZAS, O., RICA, I., LOCKE, J. M., MAESTRO, M. A., ALSHAIKH, A., BUNDAK, R., DEL CASTILLO, G., DEEB, A., DEISS, D., FERNANDEZ, J. M., GODBOLE, K., HUSSAIN, K., O'CONNELL, M., KLUPA, T., KOLOUSKOVA, S., MOHSIN, F., PERLMAN, K., SUMNIK, Z., RIAL, J. M., UGARTE, E., VASANTHI, T., NEONATAL DIABETES INTERNATIONAL, G., JOHNSTONE, K., FLANAGAN, S. E., MARTINEZ, R., CASTANO, C., PATCH, A. M.,

FERNANDEZ-REBOLLO, E., RAILE, K., MORGAN, N., HARRIES, L. W., CASTANO, L., ELLARD, S., FERRER, J., PEREZ DE NANCLARES, G. & HATTERSLEY, A. T. 2010. Recessive mutations in the INS gene result in neonatal diabetes through reduced insulin biosynthesis. *Proc Natl Acad Sci U S A,* 107**,** 3105-10.

GREENHALGH, T., CLINCH, M., AFSAR, N., CHOUDHURY, Y., SUDRA, R., CAMPBELL-RICHARDS, D., CLAYDON, A., HITMAN, G. A., HANSON, P. & FINER, S. 2015. Socio-cultural influences on the behaviour of South Asian women with diabetes in pregnancy: qualitative study using a multi-level theoretical approach. *BMC Med,* 13**,** 120.

GRIFFITHS, L. J., DEZATEUX, C. & COLE, T. J. 2007. Differential parental weight and height contributions to offspring birthweight and weight gain in infancy. *Int J Epidemiol,* 36**,** 104-7.

GURURAJAN, P., GURUMURTHY, P., NAYAR, P., RAO, G. S., BABU, R. S., SARASABHARATI, A. & CHERIAN, K. M. 2012. Pregnancy associated plasma protein-A (PAPP-A) as an early marker for the diagnosis of acute coronary syndrome. *Indian Heart J,* 64**,** 141-5.

HAJIAN-TILAKI, K. 2013. Receiver Operating Characteristic (ROC) Curve Analysis for Medical Diagnostic Test Evaluation. *Caspian J Intern Med,* 4**,** 627-35.

HARDER, T., PLAGEMANN, A. & HARDER, A. 2008. Birth weight and subsequent risk of childhood primary brain tumors: a meta-analysis. *Am J Epidemiol,* 168**,** 366-73.

HARDER, T., RODEKAMP, E., SCHELLONG, K., DUDENHAUSEN, J. W. & PLAGEMANN, A. 2007. Birth weight and subsequent risk of type 2 diabetes: a meta-analysis. *Am J Epidemiol,* 165**,** 849-57.

HARRELL, F. E., JR., LEE, K. L., CALIFF, R. M., PRYOR, D. B. & ROSATI, R. A. 1984. Regression modelling strategies for improved prognostic prediction. *Stat Med,* 3**,** 143-52.

HATTERSLEY, A. T. & TOOKE, J. E. 1999. The fetal insulin hypothesis: an alternative explanation of the association of low birthweight with diabetes and vascular disease. *Lancet,* 353**,** 1789-92.

HAULDER, M., HUGHES, A. E., BEAUMONT, R. N., KNIGHT, B. A., HATTERSLEY, A. T., SHIELDS, B. M. & FREATHY, R. M. 2022. Assessing whether genetic scores explain extra variation in birthweight, when added to clinical and anthropometric measures. *BMC Pediatr,* 22**,** 504.

HINKLE, S. N., ALBERT, P. S., MENDOLA, P., SJAARDA, L. A., YEUNG, E., BOGHOSSIAN, N. S. & LAUGHON, S. K. 2014. The association between parity and birthweight in a longitudinal consecutive pregnancy cohort. *Paediatr Perinat Epidemiol,* 28**,** 106-15.

HOFFNER, L. & SURTI, U. 2012. The genetics of gestational trophoblastic disease: a rare complication of pregnancy. *Cancer Genet,* 205**,** 63-77.

HONG, Y. H. & LEE, J. E. 2021. Large for Gestational Age and Obesity-Related Comorbidities. *J Obes Metab Syndr,* 30**,** 124-131.

HUGHES, A. E., DE FRANCO, E., FREATHY, R. M., FETAL, I., GROWTH, C., FLANAGAN, S. E. & HATTERSLEY, A. T. 2023. Monogenic disease analysis establishes that fetal insulin accounts for half of human fetal growth. *J Clin Invest,* 133.

HUGHES, A. E., HATTERSLEY, A. T., FLANAGAN, S. E. & FREATHY, R. M. 2021. Two decades since the fetal insulin hypothesis: what have we learned from genetics? *Diabetologia,* 64**,** 717-726.

HUGHES, A. E., NODZENSKI, M., BEAUMONT, R. N., TALBOT, O., SHIELDS, B. M., SCHOLTENS, D. M., KNIGHT, B. A., LOWE, W. L., JR., HATTERSLEY, A. T. & FREATHY, R. M. 2018. Fetal Genotype and Maternal Glucose Have Independent and Additive Effects on Birth Weight. *Diabetes,* 67**,** 1024-1029.

JOHNSTON, L. B., CLARK, A. J. & SAVAGE, M. O. 2002. Genetic factors contributing to birth weight. *Arch Dis Child Fetal Neonatal Ed,* 86**,** F2-3.

JULIUSDOTTIR, T., STEINTHORSDOTTIR, V., STEFANSDOTTIR, L., SVEINBJORNSSON, G., IVARSDOTTIR, E. V., THOROLFSDOTTIR, R. B., SIGURDSSON, J. K., TRAGANTE, V., HJORLEIFSSON, K. E., HELGADOTTIR, A., FRIGGE, M. L., THORGEIRSSON, G., BENEDIKTSSON, R., SIGURDSSON, E. L., ARNAR, D. O., STEINGRIMSDOTTIR, T., JONSDOTTIR, I., HOLM, H., GUDBJARTSSON, D. F., THORLEIFSSON, G., THORSTEINSDOTTIR, U. & STEFANSSON, K. 2021. Distinction between the effects of parental and fetal genomes on fetal growth. *Nat Genet,* 53**,** 1135-1142.

KALAFAT, E., MORALES-ROSELLO, J., THILAGANATHAN, B., DHOTHER, J. & KHALIL, A. 2019. Risk of neonatal care unit admission in small for gestational age fetuses at term: a prediction model and internal validation. *J Matern Fetal Neonatal Med,* 32**,** 2361-2368.

KELLY, Y., PANICO, L., BARTLEY, M., MARMOT, M., NAZROO, J. & SACKER, A. 2009. Why does birthweight vary among ethnic groups in the UK? Findings from the Millennium Cohort Study. *J Public Health (Oxf),* 31**,** 131-7.

KLEIN, R. J., ZEISS, C., CHEW, E. Y., TSAI, J. Y., SACKLER, R. S., HAYNES, C., HENNING, A. K., SANGIOVANNI, J. P., MANE, S. M., MAYNE, S. T., BRACKEN, M. B., FERRIS, F. L., OTT, J., BARNSTABLE, C. & HOH, J. 2005. Complement factor H polymorphism in age-related macular degeneration. *Science,* 308**,** 385-9.

KNIGHT, B., SHIELDS, B. M. & HATTERSLEY, A. T. 2006. The Exeter Family Study of Childhood Health (EFSOCH): study protocol and methodology. *Paediatr Perinat Epidemiol,* 20**,** 172-9.

KNIGHT, B., SHIELDS, B. M., TURNER, M., POWELL, R. J., YAJNIK, C. S. & HATTERSLEY, A. T. 2005. Evidence of genetic regulation of fetal longitudinal growth. *Early Hum Dev,* 81**,** 823-31.

KOUROU, K., EXARCHOS, T. P., EXARCHOS, K. P., KARAMOUZIS, M. V. & FOTIADIS, D. I. 2015. Machine learning applications in cancer prognosis and prediction. *Comput Struct Biotechnol J,* 13**,** 8-17.

KROOK, A., BRUETON, L. & O'RAHILLY, S. 1993. Homozygous nonsense mutation in the insulin receptor gene in infant with leprechaunism. *Lancet,* 342**,** 277-8.

KUHLE, S., MAGUIRE, B., ZHANG, H., HAMILTON, D., ALLEN, A. C., JOSEPH, K. S. & ALLEN, V. M. 2018. Comparison of logistic regression with machine learning methods for the prediction of fetal growth abnormalities: a retrospective cohort study. *BMC Pregnancy Childbirth,* 18**,** 333.

KURPAD, A. V., VARADHARAJAN, K. S. & AEBERLI, I. 2011. The thin-fat phenotype and global metabolic disease risk. *Curr Opin Clin Nutr Metab Care,* 14**,** 542-7.

LABARERE, J., RENAUD, B. & FINE, M. J. 2014. How to derive and validate clinical prediction models for use in intensive care medicine. *Intensive Care Med,* 40**,** 513-27.

LANGE, S., PROBST, C., REHM, J. & POPOVA, S. 2018. National, regional, and global prevalence of smoking during pregnancy in the general population: a systematic review and meta-analysis. *Lancet Glob Health,* 6**,** e769-e776.

LANGER, O., LEVY, J., BRUSTMAN, L., ANYAEGBUNAM, A., MERKATZ, R. & DIVON, M. 1989. Glycemic control in gestational diabetes mellitus--how tight is tight enough: small for gestational age versus large for gestational age? *Am J Obstet Gynecol,* 161**,** 646-53.

LEE, Y., SHIN, J. H., PARK, H. C., KIM, S. G. & CHOI, S. I. 2014. A prediction model for renal artery stenosis using carotid ultrasonography measurements in patients undergoing coronary angiography. *BMC Nephrol,* 15**,** 60.

MAGNUS, P., GJESSING, H. K., SKRONDAL, A. & SKJAERVEN, R. 2001. Paternal contribution to birth weight. *J Epidemiol Community Health,* 55**,** 873-7.

MAHAJAN, A., WESSEL, J., WILLEMS, S. M., ZHAO, W., ROBERTSON, N. R., CHU, A. Y., GAN, W., KITAJIMA, H., TALIUN, D., RAYNER, N. W., GUO, X., LU, Y., LI, M., JENSEN, R. A., HU, Y., HUO, S., LOHMAN, K. K., ZHANG, W., COOK, J. P., PRINS, B. P., FLANNICK, J., GRARUP, N., TRUBETSKOY, V. V., KRAVIC, J., KIM, Y. J., RYBIN, D. V., YAGHOOTKAR, H., MULLER-NURASYID, M., MEIDTNER, K., LI-GAO, R., VARGA, T. V., MARTEN, J., LI, J., SMITH, A. V., AN, P., LIGTHART, S., GUSTAFSSON, S., MALERBA, G., DEMIRKAN, A., TAJES, J. F., STEINTHORSDOTTIR, V., WUTTKE, M., LECOEUR, C., PREUSS, M., BIELAK, L. F., GRAFF, M., HIGHLAND, H. M., JUSTICE, A. E., LIU, D. J., MAROULI, E., PELOSO, G. M., WARREN, H. R., EXOME, B. P. C., CONSORTIUM, M., CONSORTIUM, G., AFAQ, S., AFZAL, S., AHLQVIST, E., ALMGREN, P., AMIN, N., BANG, L. B., BERTONI, A. G., BOMBIERI, C., BORK-JENSEN, J., BRANDSLUND, I., BRODY, J. A., BURTT, N. P., CANOUIL, M., CHEN, Y. I., CHO, Y. S., CHRISTENSEN, C., EASTWOOD, S. V., ECKARDT, K. U., FISCHER, K., GAMBARO, G., GIEDRAITIS, V., GROVE, M. L., DE HAAN, H. G., HACKINGER, S., HAI, Y., HAN, S., TYBJAERG-HANSEN, A., HIVERT, M. F., ISOMAA, B., JAGER, S., JORGENSEN, M. E., JORGENSEN, T., KARAJAMAKI, A., KIM, B. J., KIM, S. S., KOISTINEN, H. A., KOVACS, P., KRIEBEL, J., KRONENBERG, F., LALL, K., LANGE, L. A., LEE, J. J., LEHNE, B., LI, H., LIN, K. H., et al. 2018. Refining the accuracy of validated target identification through coding variant fine-mapping in type 2 diabetes. *Nat Genet,* 50**,** 559-571.

MANICHAIKUL, A., MYCHALECKYJ, J. C., RICH, S. S., DALY, K., SALE, M. & CHEN, W. M. 2010. Robust relationship inference in genome-wide association studies. *Bioinformatics,* 26**,** 2867-73.

MARTIN-GRONERT, M. S. & OZANNE, S. E. 2010. Mechanisms linking suboptimal early nutrition and increased risk of type 2 diabetes and obesity. *J Nutr,* 140**,** 662-6.

MARTIN, S., CULE, M., BASTY, N., TYRRELL, J., BEAUMONT, R. N., WOOD, A. R., FRAYLING, T. M., SOROKIN, E., WHITCHER, B., LIU, Y., BELL, J. D., THOMAS, E. L. & YAGHOOTKAR, H. 2021. Genetic Evidence for Different Adiposity Phenotypes and Their Opposing Influences on Ectopic Fat and Risk of Cardiometabolic Disease. *Diabetes,* 70**,** 1843-1856.

MASIAKWALA, E., NYATI, L. H. & NORRIS, S. A. 2023. The association of intrauterine and postnatal growth patterns and nutritional status with toddler body composition. *BMC Pediatr,* 23**,** 342.

MEERTENS, L., SMITS, L., VAN KUIJK, S., AARDENBURG, R., VAN DOOREN, I., LANGENVELD, J., ZWAAN, I. M., SPAANDERMAN, M. & SCHEEPERS, H. 2019. External validation and clinical usefulness of first-trimester prediction models for small- and large-for-gestational-age infants: a prospective cohort study. *BJOG,* 126**,** 472-484.

MENOTTI, A., PUDDU, P. E. & LANTI, M. 2000. Comparison of the Framingham risk function-based coronary chart with risk function from an Italian population study. *Eur Heart J,* 21**,** 365-70.

METGUD, C. S., NAIK, V. A. & MALLAPUR, M. D. 2012. Factors affecting birth weight of a newborn--a community based study in rural Karnataka, India. *PLoS One,* 7**,** e40040.

MI, D., FANG, H., ZHAO, Y. & ZHONG, L. 2017. Birth weight and type 2 diabetes: A meta-analysis. *Exp Ther Med,* 14**,** 5313-5320.

MILNER, J. & AREZINA, J. 2018. The accuracy of ultrasound estimation of fetal weight in comparison to birth weight: A systematic review. *Ultrasound,* 26**,** 32-41.

MIRANDA, M. L., EDWARDS, S. E. & MYERS, E. R. 2011. Adverse birth outcomes among nulliparous vs. multiparous women. *Public Health Rep,* 126**,** 797-805.

MONARI, F., MENICHINI, D., SPANO' BASCIO, L., GRANDI, G., BANCHELLI, F., NERI, I., D'AMICO, R. & FACCHINETTI, F. 2021. A first trimester prediction model for large for gestational age infants: a preliminary study. *BMC Pregnancy Childbirth,* 21**,** 654.

MORLEY, R. 2006. Fetal origins of adult disease. *Semin Fetal Neonatal Med,* 11**,** 73-8.

NIGHTINGALE, C. M., RUDNICKA, A. R., OWEN, C. G., COOK, D. G. & WHINCUP, P. H. 2011. Patterns of body size and adiposity among UK children of South Asian, black African-Caribbean and white European origin: Child Heart And health Study in England (CHASE Study). *Int J Epidemiol,* 40**,** 33-44.

NONGMAITHEM, S. S., BEAUMONT, R. N., DEDANIYA, A., WOOD, A. R., OGUNKOLADE, B. W., HASSAN, Z., KRISHNAVENI, G. V., KUMARAN, K., POTDAR, R. D., SAHARIAH, S. A., KRISHNA, M., DI GRAVIO, C., MALI, I. D., SANKARESWARAN, A., HUSSAIN, A., BHOWMIK, B. W., KHAN, A. K. A., KNIGHT, B. A., FRAYLING, T. M., FINER, S., FALL, C. H. D., YAJNIK, C. S., FREATHY, R. M., HITMAN, G. A. & CHANDAK, G. R. 2022. Babies of South Asian and European Ancestry Show Similar Associations With Genetic Risk Score for Birth Weight Despite the Smaller Size of South Asian Newborns. *Diabetes,* 71**,** 821-836.

NORDMAN, H., JAASKELAINEN, J. & VOUTILAINEN, R. 2020. Birth Size as a Determinant of Cardiometabolic Risk Factors in Children. *Horm Res Paediatr,* 93**,** 144-153.

ONG, K. K., AHMED, M. L., EMMETT, P. M., PREECE, M. A. & DUNGER, D. B. 2000. Association between postnatal catch-up growth and obesity in childhood: prospective cohort study. *BMJ,* 320**,** 967-71.

OPARA, E. I. & ZAIDI, J. 2007. The interpretation and clinical application of the word 'parity': a survey. *BJOG,* 114**,** 1295-7.

OSUCHUKWU, O. O. & REED, D. J. 2023. Small for Gestational Age. *StatPearls.* Treasure Island (FL).

PAPASTEFANOU, I., SOUKA, A. P., PILALIS, A., ELEFTHERIADES, M., MICHALITSI, V. & KASSANOS, D. 2012. First trimester prediction of small- and large-for-gestation neonates by an integrated model incorporating ultrasound parameters, biochemical indices and maternal characteristics. *Acta Obstet Gynecol Scand,* 91**,** 104-111.

PLASENCIA, W., AKOLEKAR, R., DAGKLIS, T., VEDUTA, A. & NICOLAIDES, K. H. 2011. Placental volume at 11-13 weeks' gestation in the prediction of birth weight percentile. *Fetal Diagn Ther,* 30**,** 23-8.

PLASENCIA, W., GONZALEZ DAVILA, E., TETILLA, V., PADRON PEREZ, E., GARCIA HERNANDEZ, J. A. & GONZALEZ GONZALEZ, N. L. 2012. First-trimester screening for large-for-gestational-age infants. *Ultrasound Obstet Gynecol,* 39**,** 389-95.

POON, L. C., KARAGIANNIS, G., STRATIEVA, V., SYNGELAKI, A. & NICOLAIDES, K. H. 2011. First-trimester prediction of macrosomia. *Fetal Diagn Ther,* 29**,** 139-47.

POYRAZOGLU, S., DARENDELILER, F., AHMED, S. F., HUGHES, I., BRYCE, J., JIANG, J., RODIE, M., HIORT, O., HANNEMA, S. E., BERTELLONI, S., LISA, L., GURAN, T., COOLS, M., DESLOOVERE, A., CLAAHSEN-VAN DER GRINTEN, H. L., NORDENSTROM, A., HOLTERHUS, P. M., KOHLER, B., NIEDZIELA, M. & KRONE, N. 2017. Birth Weight in Different Etiologies of Disorders of Sex Development. *J Clin Endocrinol Metab,* 102**,** 1044-1050.

RAMSPEK, C. L., JAGER, K. J., DEKKER, F. W., ZOCCALI, C. & VAN DIEPEN, M. 2021. External validation of prognostic models: what, why, how, when and where? *Clin Kidney J,* 14**,** 49-58.

RANI, P. R. & BEGUM, J. 2016. Screening and Diagnosis of Gestational Diabetes Mellitus, Where Do We Stand. *J Clin Diagn Res,* 10**,** QE01-4.

RENOM ESPINEIRA, A., FERNANDES-ROSA, F. L., BUENO, A. C., DE SOUZA, R. M., MOREIRA, A. C., DE CASTRO, M., BARBIERI, M. A., BETTIOL, H. & ANTONINI, S. R. 2011. Postnatal growth and cardiometabolic profile in young adults born large for gestational age. *Clin Endocrinol (Oxf),* 75**,** 335-41.

RILEY, R. D., ENSOR, J., SNELL, K. I. E., HARRELL, F. E., JR., MARTIN, G. P., REITSMA, J. B., MOONS, K. G. M., COLLINS, G. & VAN SMEDEN, M. 2020. Calculating the sample size required for developing a clinical prediction model. *BMJ,* 368**,** m441.

RUBIO-CABEZAS, O. & ELLARD, S. 2013. Diabetes mellitus in neonates and infants: genetic heterogeneity, clinical approach to diagnosis, and therapeutic options. *Horm Res Paediatr,* 80**,** 137-46.
SAENGER, P., CZERNICHOW, P., HUGHES, I. & REITER, E. O. 2007. Small for gestational age: short stature and beyond. *Endocr Rev,* 28**,** 219-51.
SCHOLL, T. O., SOWERS, M., CHEN, X. & LENDERS, C. 2001. Maternal glucose concentration influences fetal growth, gestation, and pregnancy complications. *Am J Epidemiol,* 154**,** 514-20.
SCIFRES, C. M. 2021. Short- and Long-Term Outcomes Associated with Large for Gestational Age Birth Weight. *Obstet Gynecol Clin North Am,* 48**,** 325-337.
SHIPE, M. E., DEPPEN, S. A., FARJAH, F. & GROGAN, E. L. 2019. Developing prediction models for clinical use using logistic regression: an overview. *J Thorac Dis,* 11**,** S574-S584.
SINGH, V., DWIVEDI, S. N. & DEO, S. V. S. 2020. Ordinal logistic regression model describing factors associated with extent of nodal involvement in oral cancer patients and its prospective validation. *BMC Med Res Methodol,* 20**,** 95.
SMEENK, J. M., BRAAT, D. D., STOLWIJK, A. M. & KREMER, J. A. 2007. Pregnancy is predictable: a large-scale prospective external validation of the prediction of spontaneous pregnancy in subfertile couples. *Hum Reprod,* 22**,** 2344-5; author reply 2345-6.
SPENCER, N. & LOGAN, S. 2002. Social influences on birth weight. *J Epidemiol Community Health,* 56**,** 326-7.
SRIDHAR, S. B., FERRARA, A., EHRLICH, S. F., BROWN, S. D. & HEDDERSON, M. M. 2013. Risk of large-for-gestational-age newborns in women with gestational diabetes by race and ethnicity and body mass index categories. *Obstet Gynecol,* 121**,** 1255-1262.
STEYERBERG, E. W. 2010. *Clinical prediction models : a practical approach to development, validation and updating,* New York, Springer.
STEYERBERG, E. W., HARRELL, F. E., JR., BORSBOOM, G. J., EIJKEMANS, M. J., VERGOUWE, Y. & HABBEMA, J. D. 2001. Internal validation of predictive models: efficiency of some procedures for logistic regression analysis. *J Clin Epidemiol,* 54**,** 774-81.
SUN, H., DEPRAETERE, K., MEESSEMAN, L., CABANILLAS SILVA, P., SZYMANOWSKY, R., FLIEGENSCHMIDT, J., HULDE, N., VON DOSSOW, V., VANBIERVLIET, M., DE BAERDEMAEKER, J., ROCCARO-WALDMEYER, D. M., STIEG, J., DOMINGUEZ HIDALGO, M. & DAHLWEID, F. M. 2022. Machine Learning-Based Prediction Models for Different Clinical Risks in Different Hospitals: Evaluation of Live Performance. *J Med Internet Res,* 24**,** e34295.
SURKAN, P. J., HSIEH, C. C., JOHANSSON, A. L., DICKMAN, P. W. & CNATTINGIUS, S. 2004. Reasons for increasing trends in large for gestational age births. *Obstet Gynecol,* 104**,** 720-6.
TAKAGI, K., IWAMA, N., METOKI, H., UCHIKURA, Y., MATSUBARA, Y., MATSUBARA, K., NISHIGORI, H., SAITO, M., FUJIWARA, I., SAKURAI, K., KURIYAMA, S., ARIMA, T., NAKAI, K., YAEGASHI, N., SUGIYAMA, T., JAPAN, E. & CHILDREN'S STUDY, G. 2019. Paternal height has an impact

on birth weight of their offspring in a Japanese population: the Japan Environment and Children's Study. *J Dev Orig Health Dis,* 10**,** 542-554.
TEKOLA-AYELE, F., WORKALEMAHU, T. & AMARE, A. T. 2018. High burden of birthweight-lowering genetic variants in Africans and Asians. *BMC Med,* 16**,** 70.
TERAUCHI, Y., KUBOTA, N., TAMEMOTO, H., SAKURA, H., NAGAI, R., AKANUMA, Y., KIMURA, S. & KADOWAKI, T. 2000. Insulin effect during embryogenesis determines fetal growth: a possible molecular link between birth weight and susceptibility to type 2 diabetes. *Diabetes,* 49**,** 82-6.
THOMPSON, W. D., BEAUMONT, R. N., KUANG, A., WARRINGTON, N. M., JI, Y., TYRRELL, J., WOOD, A. R., SCHOLTENS, D. M., KNIGHT, B. A., EVANS, D. M., LOWE, W. L., JR., SANTORELLI, G., AZAD, R., MASON, D., HATTERSLEY, A. T., FRAYLING, T. M., YAGHOOTKAR, H., BORGES, M. C., LAWLOR, D. A. & FREATHY, R. M. 2021. Higher maternal adiposity reduces offspring birthweight if associated with a metabolically favourable profile. *Diabetologia,* 64**,** 2790-2802.
THOMPSON, W. D., BEAUMONT, R. N., KUANG, A., WARRINGTON, N. M., JI, Y., TYRRELL, J., WOOD, A. R., SCHOLTENS, D. M., KNIGHT, B. A., EVANS, D. M., LOWE, W. L., JR., SANTORELLI, G., AZAD, R., MASON, D., HATTERSLEY, A. T., FRAYLING, T. M., YAGHOOTKAR, H., BORGES, M. C., LAWLOR, D. A. & FREATHY, R. M. 2022. Fetal alleles predisposing to metabolically favorable adiposity are associated with higher birth weight. *Hum Mol Genet,* 31**,** 1762-1775.
TISE, C. G. & BYERS, H. M. 2021. Genetics of recurrent pregnancy loss: a review. *Curr Opin Obstet Gynecol,* 33**,** 106-111.
TRICHE, E. W. & HOSSAIN, N. 2007. Environmental factors implicated in the causation of adverse pregnancy outcome. *Semin Perinatol,* 31**,** 240-2.
TYRRELL, J., RICHMOND, R. C., PALMER, T. M., FEENSTRA, B., RANGARAJAN, J., METRUSTRY, S., CAVADINO, A., PATERNOSTER, L., ARMSTRONG, L. L., DE SILVA, N. M., WOOD, A. R., HORIKOSHI, M., GELLER, F., MYHRE, R., BRADFIELD, J. P., KREINER-MOLLER, E., HUIKARI, V., PAINTER, J. N., HOTTENGA, J. J., ALLARD, C., BERRY, D. J., BOUCHARD, L., DAS, S., EVANS, D. M., HAKONARSON, H., HAYES, M. G., HEIKKINEN, J., HOFMAN, A., KNIGHT, B., LIND, P. A., MCCARTHY, M. I., MCMAHON, G., MEDLAND, S. E., MELBYE, M., MORRIS, A. P., NODZENSKI, M., REICHETZEDER, C., RING, S. M., SEBERT, S., SENGPIEL, V., SORENSEN, T. I., WILLEMSEN, G., DE GEUS, E. J., MARTIN, N. G., SPECTOR, T. D., POWER, C., JARVELIN, M. R., BISGAARD, H., GRANT, S. F., NOHR, E. A., JADDOE, V. W., JACOBSSON, B., MURRAY, J. C., HOCHER, B., HATTERSLEY, A. T., SCHOLTENS, D. M., DAVEY SMITH, G., HIVERT, M. F., FELIX, J. F., HYPPONEN, E., LOWE, W. L., JR., FRAYLING, T. M., LAWLOR, D. A., FREATHY, R. M. & EARLY GROWTH GENETICS, C. 2016. Genetic Evidence for Causal Relationships Between Maternal Obesity-Related Traits and Birth Weight. *JAMA,* 315**,** 1129-40.
UFFELMANN, E., POSTHUMA, D. & PEYROT, W. J. 2023. Genome-wide association studies of polygenic risk score-derived phenotypes may lead to inflated false positive rates. *Sci Rep,* 13**,** 4219.

VAN DIJK, M. & OUDEJANS, C. 2013. (Epi)genetics of pregnancy-associated diseases. *Front Genet,* 4**,** 180.
VICKERS, A. J. & ELKIN, E. B. 2006. Decision curve analysis: a novel method for evaluating prediction models. *Med Decis Making,* 26**,** 565-74.
VIEIRA, M. C., MCCOWAN, L. M. E., GILLETT, A., POSTON, L., FYFE, E., DEKKER, G. A., BAKER, P. N., WALKER, J. J., KENNY, L. C., PASUPATHY, D. & CONSORTIUM, S. 2017. Clinical, ultrasound and molecular biomarkers for early prediction of large for gestational age infants in nulliparous women: An international prospective cohort study. *PLoS One,* 12**,** e0178484.
VOSKAMP, B. J., PEELEN, M., RAVELLI, A. C. J., VAN DER LEE, R., MOL, B. W. J., PAJKRT, E., GANZEVOORT, W. & KAZEMIER, B. M. 2020. Association between fetal sex, birthweight percentile and adverse pregnancy outcome. *Acta Obstet Gynecol Scand,* 99**,** 48-58.
WAHAB, R. J., JADDOE, V. W. V., VAN KLAVEREN, D., VERMEULEN, M. J., REISS, I. K. M., STEEGERS, E. A. P. & GAILLARD, R. 2022. Preconception and early-pregnancy risk prediction for birth complications: development of prediction models within a population-based prospective cohort. *BMC Pregnancy Childbirth,* 22**,** 165.
WANG, J. W., LYU, Y. N., QIAO, B., LI, Y., ZHANG, Y., DHANYAMRAJU, P. K., BAMME, Y., YU, M. D., YANG, D. & TONG, Y. Q. 2021. Cell-free fetal DNA testing and its correlation with prenatal indications. *BMC Pregnancy Childbirth,* 21**,** 585.
WANG, N., GUO, H., JING, Y., ZHANG, Y., SUN, B., PAN, X., CHEN, H., XU, J., WANG, M., CHEN, X., SONG, L. & CUI, W. 2023. Development and validation of risk prediction models for large for gestational age infants using logistic regression and two machine learning algorithms. *J Diabetes,* 15**,** 338-348.
WARRINGTON, N. M., BEAUMONT, R. N., HORIKOSHI, M., DAY, F. R., HELGELAND, O., LAURIN, C., BACELIS, J., PENG, S., HAO, K., FEENSTRA, B., WOOD, A. R., MAHAJAN, A., TYRRELL, J., ROBERTSON, N. R., RAYNER, N. W., QIAO, Z., MOEN, G. H., VAUDEL, M., MARSIT, C. J., CHEN, J., NODZENSKI, M., SCHNURR, T. M., ZAFARMAND, M. H., BRADFIELD, J. P., GRARUP, N., KOOIJMAN, M. N., LI-GAO, R., GELLER, F., AHLUWALIA, T. S., PATERNOSTER, L., RUEEDI, R., HUIKARI, V., HOTTENGA, J. J., LYYTIKAINEN, L. P., CAVADINO, A., METRUSTRY, S., COUSMINER, D. L., WU, Y., THIERING, E., WANG, C. A., HAVE, C. T., VILOR-TEJEDOR, N., JOSHI, P. K., PAINTER, J. N., NTALLA, I., MYHRE, R., PITKANEN, N., VAN LEEUWEN, E. M., JORO, R., LAGOU, V., RICHMOND, R. C., ESPINOSA, A., BARTON, S. J., INSKIP, H. M., HOLLOWAY, J. W., SANTA-MARINA, L., ESTIVILL, X., ANG, W., MARSH, J. A., REICHETZEDER, C., MARULLO, L., HOCHER, B., LUNETTA, K. L., MURABITO, J. M., RELTON, C. L., KOGEVINAS, M., CHATZI, L., ALLARD, C., BOUCHARD, L., HIVERT, M. F., ZHANG, G., MUGLIA, L. J., HEIKKINEN, J., CONSORTIUM, E. G. G., MORGEN, C. S., VAN KAMPEN, A. H. C., VAN SCHAIK, B. D. C., MENTCH, F. D., LANGENBERG, C., LUAN, J., SCOTT, R. A., ZHAO, J. H., HEMANI, G., RING, S. M., BENNETT, A. J., GAULTON, K. J., FERNANDEZ-TAJES, J., VAN

ZUYDAM, N. R., MEDINA-GOMEZ, C., DE HAAN, H. G., ROSENDAAL, F. R., KUTALIK, Z., MARQUES-VIDAL, P., DAS, S., WILLEMSEN, G., MBAREK, H., MULLER-NURASYID, M., STANDL, M., APPEL, E. V. R., FONVIG, C. E., et al. 2019. Maternal and fetal genetic effects on birth weight and their relevance to cardio-metabolic risk factors. *Nat Genet,* 51**,** 804-814.

WEEDON, M. N., CLARK, V. J., QIAN, Y., BEN-SHLOMO, Y., TIMPSON, N., EBRAHIM, S., LAWLOR, D. A., PEMBREY, M. E., RING, S., WILKIN, T. J., VOSS, L. D., JEFFERY, A. N., METCALF, B., FERRUCCI, L., CORSI, A. M., MURRAY, A., MELZER, D., KNIGHT, B., SHIELDS, B., SMITH, G. D., HATTERSLEY, A. T., DI RIENZO, A. & FRAYLING, T. M. 2006. A common haplotype of the glucokinase gene alters fasting glucose and birth weight: association in six studies and population-genetics analyses. *Am J Hum Genet,* 79**,** 991-1001.

WEI, J. N., SUNG, F. C., LI, C. Y., CHANG, C. H., LIN, R. S., LIN, C. C., CHIANG, C. C. & CHUANG, L. M. 2003. Low birth weight and high birth weight infants are both at an increased risk to have type 2 diabetes among schoolchildren in taiwan. *Diabetes Care,* 26**,** 343-8.

WEST, J., LAWLOR, D. A., FAIRLEY, L., BHOPAL, R., CAMERON, N., MCKINNEY, P. A., SATTAR, N. & WRIGHT, J. 2013. UK-born Pakistani-origin infants are relatively more adipose than white British infants: findings from 8704 mother-offspring pairs in the Born-in-Bradford prospective birth cohort. *J Epidemiol Community Health,* 67**,** 544-51.

WILCOX, A. J. 2001. On the importance--and the unimportance--of birthweight. *Int J Epidemiol,* 30**,** 1233-41.

WOJCIK, M. H., REIMERS, R., POORVU, T. & AGRAWAL, P. B. 2020. Genetic diagnosis in the fetus. *J Perinatol,* 40**,** 997-1006.

WRIGHT, C. M., WILLIAMS, A. F., ELLIMAN, D., BEDFORD, H., BIRKS, E., BUTLER, G., SACHS, M., MOY, R. J. & COLE, T. J. 2010. Using the new UK-WHO growth charts. *BMJ,* 340**,** c1140.

WRIGHT, J., SMALL, N., RAYNOR, P., TUFFNELL, D., BHOPAL, R., CAMERON, N., FAIRLEY, L., LAWLOR, D. A., PARSLOW, R., PETHERICK, E. S., PICKETT, K. E., WAIBLINGER, D., WEST, J. & BORN IN BRADFORD SCIENTIFIC COLLABORATORS, G. 2013. Cohort Profile: the Born in Bradford multi-ethnic family cohort study. *Int J Epidemiol,* 42**,** 978-91.

ZHANG, G., BACELIS, J., LENGYEL, C., TERAMO, K., HALLMAN, M., HELGELAND, O., JOHANSSON, S., MYHRE, R., SENGPIEL, V., NJOLSTAD, P. R., JACOBSSON, B. & MUGLIA, L. 2015. Assessing the Causal Relationship of Maternal Height on Birth Size and Gestational Age at Birth: A Mendelian Randomization Analysis. *PLoS Med,* 12**,** e1001865.

Chapter 5

Developing and validating a multivariable clinical prediction model for babies born LGA in European and South Asian cohorts

Developing and validating a multivariable clinical prediction model for babies born LGA in European and South Asian cohorts

Abstract

Background

In current clinical practice, an Oral fasting glucose tolerance test (OGTT) detects mothers at risk of gestational diabetes, and hence, at risk of delivering a baby large-for-gestational age (LGA), but not all LGA babies are delivered to mothers with gestational diabetes. In this case, a clinical prediction model is very useful in using routinely available data from clinic visits to predict the risk of LGA.

Methods

Multivariable logistic regression models were built with the outcome variable LGA or not LGA and the predictors as mother's weight, height, fasting glucose, parity and smoking status during the pregnancy. The model was built in a UK cohort (n=906, EFSOCH), internally validated in the same cohort by bootstrapping and then externally validated in a separate UK cohort with both European and Pakistani mothers (n=1583, n=1115, BiB). Calibration curves and the ROC Curve were used to assess the model fit. External validation assessed the generalizability of the model.

Results

Higher weight and glucose and primiparity and not smoking were all associated with increased risk of LGA. When combined in a prediction model, these features showed reasonable discriminatory power (ROC AUC=0.761). In external validation, the model calibrated well in Europeans, but overestimated in the Pakistani cohort,

where the proportion with LGA was lower (7.1% v 3.5% in BiB). This improved with model recalibration.

Conclusions

We developed and validated a model for detection of LGA using multiple clinical features that can be used in everyone rather than just those at risk of gestational diabetes.

Introduction

Babies born large-for-gestational age (LGA) are at higher risk of adverse pregnancy outcomes, for example, shoulder dystocia which could result in prolonged labor and obstetric emergency, mother requiring a caesarian section or the baby developing type 2 diabetes in the long run(Mi et al., 2017, Wright et al., 2010). To cater for antenatal and postnatal care, it is important to determine which babies are at risk of developing this condition.

Predicting which babies will be LGA is challenging. In the UK, it is common to use an oral glucose tolerance test (OGTT) in current clinical practice to detect mothers at risk of gestational diabetes. These women are also at risk of delivering a baby born LGA. However, not all LGA babies are delivered to mothers with gestational diabetes(Hughes et al., 2018). When only mothers at risk of developing gestational diabetes are followed up for LGA, the remaining proportion of those also at risk of delivering a baby born LGA for other reasons are being dismissed.

LGA is usually a result of maternal factors, race and ethnicity, genetic factors, maternal diabetes, obesity, and excessive weight gain during pregnancy (Hong and Lee, 2021). Risk of a baby being born LGA has been found to be associated with several routinely available maternal characteristics such as mother's age, weight, smoking status, maternal BMI, among others. In addition, other markers such as parity (Meertens et al., 2019), maternal and paternal height(Frick et al., 2016), biomarkers (PAPP-A, fetal NT, free β-hCG and UtA-PI) (Plasencia et al.,

2012) and fetal biometry (the abdominal circumference (AC), head circumference (HC), biparietal Diameter (BPD), fermur length (FL) and estimated fetal weight (EFW) are all predictors of LGA (Papastefanou et al., 2012). In the UK, at least two ultrasound scans are given to expectant mothers at 10 to 14 weeks and between 18 and 21 weeks. Further scans can be allocated if required.

Clinical prediction models make use of a combination of predictors that can predict LGA better than using features in isolation. These can be easily implemented in clinical practice to estimate risk of LGA based on a criterion of available predictors. A prediction model also enables everyone with the available required factors to be assessed rather than just those at risk of gestational diabetes. Clinical prediction models built previously have modeled prediction of LGA with maternal characteristics, and usually included maternal height, weight, parity and smoking status (Poon et al., 2011). Additional maternal characteristics used were mother's age, ethnicity and interpregnancy interval(Frick et al., 2016). Models have used other types of predictors: biomarkers, medical history and fetal biometry to further improve prediction of LGA(Frick et al., 2016). A prediction model for assessing LGA based on maternal characteristics and medical history (Frick et al., 2016) determined that parous women with previous gestational diabetes mellitus (GDM), a history of type 1 diabetes mellitus, increased birthweight in previous pregnancy, increased birth weight z-score and decreased interpregnancy interval were at higher risk of delivering a baby born LGA. This risk also increased with height and weight. It was also observed that the combined screening by maternal factors and fetal biometry could predict a high proportion of pregnancies that would deliver LGA neonates.

Prediction models that have used biomarkers before have all used Pregnancy-associated plasma protein A (PAPP-A) as a common predictor. PAPP-A is defined as a promising biomarker for identification and risk stratification for patients with ACS (Gururajan et al., 2012). A first trimester model (Plasencia et al., 2012) investigated biomarkers; fetal NT, PAPP-A, free β-hCG and UtA-PI's contributions on top of the common maternal characteristics and maternal age and found that

these factors combined allowed the detection of about 34% of women who delivered LGA infants (AUCROC=0.72). In terms of fetal biometry, a first trimester, and a second trimester (Plasencia et al., 2012)showed that maternal characteristics alongside biomarkers were useful in predicting LGA and that fetal ultrasound measurements improved the prediction of this condition when added on top of maternal characteristics and biomarkers. A study carried out to investigate the prediction of LGA based on ultrasound measurements such as EFW or fetal AC showed that the prediction of a LGA neonate by EFW > 90th percentile was modest(Plasencia et al., 2012).

The aim of this study was to develop a model for LGA using data from the Exeter Family Study of Childhood Health (EFSOCH) ((Knight et al., 2006) and to validate the model in an external European as well as a South Asian cohort (Born in Bradford Study) ((Wright et al., 2013) for generalizability.

Methods

This study followed the Transparent Reporting of a Multivariable Prediction Model for Individual Prognosis or Diagnosis (TRIPOD) reporting guideline for prognostic studies (see **Appendix** for checklist).

Study population: development cohort(Knight et al., 2006, Wright et al., 2013)
We used data from the Exeter Family Study of Childhood Health (EFSOCH) ((Knight et al., 2006); a prospective White European cohort based on children born between 2000 and 2004 in postcodes EX1–4 in central Exeter, UK, for developing the prediction model.

Figure 1 shows the inclusion criteria for this study. We included observations where the offspring was born at term (≥37 and < 42 weeks' gestation) and had complete clinical data for mother’s height, weight, fasting glucose level, smoking status, parity and offspring’s birth weight. We excluded observations with mother’s

fasting glucose higher than 5.6mmol/L. We excluded participants with preterm and multiple births.

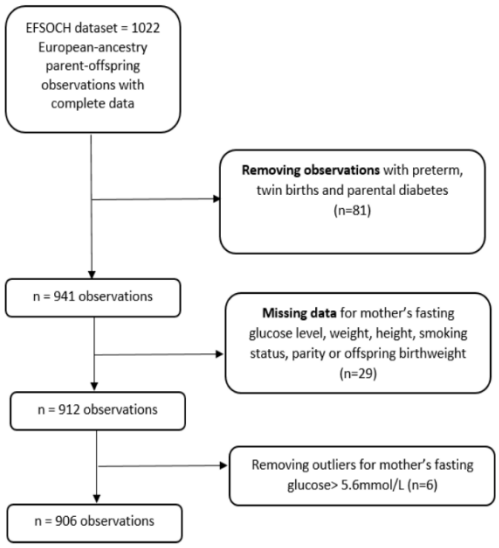


Figure 1: Flowchart of data preparation for EFSOCH

Study population: external validation cohort

For validating the prediction model, we used the Born in Bradford (BiB) study. Most clinical prediction models have been built using data from European cohorts. The risk of LGA is different between European and South Asian ancestries and the model was developed so that it could be used in both a European and South Asian population.

BiB consists of White British and Pakistani cohorts comprising of the obstetric population in Bradford; a city in North of England. The BiB study contains data collected from 12453 women (with 13776 pregnancies) between 2007 and 2010. The participants from the BiB study were mainly from a White European background and about 20% of the population was of South Asian origin (90% were from Mirpur in Pakistan).

Model Outcome

A newborn is considered large-for-gestational age (LGA) if he/she weighs more than 90% of newborns of the same gestational age at birth (above the 90th centile). We calculated sex- and gestational-age specific birth weight centiles based on z-scores generated using the UKWHOterm growth charts (Wright et al., 2010) with the outcome measure, LGA defined as >90th centile.

Model Predictors

Predictors used to build the model were those routinely available in clinics; mother's height, weight, fasting glucose level, parity and smoking status.

For the EFSOCH study, fasting blood samples were taken (fasting for at least 10 h prior to sampling) on both parents at 28 weeks' pregnancy in the early morning, at the parents' home. All measurements were taken three times on the same visits as the blood samples, and an average value calculated and used in analysis. Height was measured to the nearest 0.1 cm, using a Harpenden (Chasmors Ltd, London, UK) pocket stadiometer. Weight was measured to the nearest 100 g, using Tanita digital electric scales (model number THD-305). Parity and smoking status were self-reported variables.

For the BiB study, blood samples were taken for a fasting oral glucose tolerance test (OGTT) at 26 to 28 weeks' gestation. Anthropometric measures were recorded at the same time with height measured to the nearest 0.1 cm and weight measured to the nearest 10g by a trained project worker. Similarly, to EFSOCH, parity and smoking status were self-reported by the mothers.

Sample size

A minimum sample size of 381 is required for a prediction model with a c-statistic of 0.80, 5 parameters and an outcome prevalence of 0.10 (Riley et al., 2020). A sample size of 906 participants was obtained after adjusting for missing data and removing outliers (see **Figure 1**).

Missing data

Analysis was conducted on a complete-case analysis basis. This means that any missing data were removed for mother's height, weight, fasting glucose level, parity, and smoking status. No major differences were found between the included participants and the excluded ones (see **Supplementary table 5**).

Statistical Analysis methods

Selection of predictors

Mother's height, weight and fasting glucose were added to the model as continuous predictors and mother's smoking status and parity were added as factor variables.

We identified mother's systolic blood pressure (SBP) and diastolic blood pressure (DBP) measures closest to 28 weeks' gestation with a margin error of 2 weeks because this coincided with routine scan visits when most blood pressure measures are usually taken.

In the same way as blood pressure measurements were analyzed, the Abdominal circumference (AC), Femur Length (FL) and Head circumference (HC) closest to 20 weeks' gestation were estimated with a margin error of 2 weeks.

Checking of assumptions

Prior to model building, model assumptions for a multivariable logistic regression model were checked. It was ensured that the minimum sample size was met. Data was checked for multicollinearity by ensuring the independent variables were not too highly correlated to each other. Duplicate observations were dropped to remove those observations that come from repeated measurements or matched data. Linearity of observations were checked by plots of log odds of LGA against the independent variables (see **supplementary figures 1-4**). The outcome variable was defined as LGA-whether the newborn has a weight considered as

large for gestational age (1) or not (0) to ensure the condition is met for binary logistic regression.

Model Development

Once the model assumptions were checked and the predictors (mother's height, weight, fasting glucose, smoking status and parity) were chosen, logistic regression analysis was used to develop the prediction model with LGA as the outcome measure. Two models were built, one which used features available before the third trimester: mother's height, pre-pregnancy weight, smoking status and parity and another one which used easily available features in the third trimester: height, weight at 28 weeks, fasting glucose level, smoking status and parity. We used the receiver operating characteristic (ROC) area and the calibration curve as the main indicators of model performance.

Overfitting occurs when a model is too complex, and the statistical model then explains the random error in the model rather than the relationship between the variables. Hence, once the model had been developed, it was important to determine the degree of overfitting (optimism). To do this, bootstrapping was used. Bootstrapping with 250 samples was used as a method of internal validation, with measures of optimism and calibration performance assessed including Emax and Dxy. Emax is the maximum absolute difference in predicted and calibrated probabilities over the entire interval, that is Emax (0,1). A value close to zero would indicate good model performance. Somer's Dxy is a measure of unreliability where this statistic provides an estimate of rank correlation of observed binary response variable and the predicted probabilities. A value close to 1 would indicate good model performance. (Harrell et al., 1984)

When the model was built in EFSOCH (Model 1), the coefficients of the model were firstly applied to the BiB European cohort and then to the BiB Pakistani cohort for external validation. Re-calibration was required for external validation in the BiB Pakistani cohort due to differences in the proportions in LGA between Europeans and South Asian ethnicities. This was done by setting the initial model (Model 1) as a new linear predictor (LP1) and then fitting it into a new model (New Model) with

LGA as outcome variable. The coefficients from this new model were then used to re-calibrate Model 1.

Results

Participants

Table 1 summarizes the characteristics of the participants from EFSOCH (n=906). Babies in the EFSOCH study had a mean birthweight of 3508 g and 52% were males.

Phenotype	Mean or %
Maternal Height (cm)	165.0 (6.3)
Maternal weight(kg)	76.1 (12.8)
Maternal pre-pregnancy weight (kg)	65(11.5)
Gestational Duration (weeks)	40.1 (1.2)
Birth weight (g)	3508 (475)
Smoking Status (% Yes)	13
Primiparity (%1st pregnancy)	45
Sex of the baby (% Male)	52
Maternal fasting glucose(mmol/L)	4.4(0.4)

Table 1: Characteristics of participants in EFSOCH

Model development

The model was built using data from the EFSOCH after data cleaning as shown in Figure 1. Out of 906 participants, 97 (10.7%) were LGA.

Model specification

The final model met the assumptions for a multivariable logistic regression model.

After data preparation and the model assumptions check, the variables; mother's height, weight at 28 weeks' gestation, fasting glucose at 28 weeks' gestation were found to be associated with an increased risk of LGA whereas parity and smoking status were found to be associated with a decreased risk of LGA (**Table 2**). Mother's average SBP, average DBP (see supplementary **table 6,7,8**) and the scan measures of fetal head circumference (HC). Fetal abdominal circumference (AC) and fetal femur length (FL) were not found to be associated with LGA. (See supplementary **table 9 and 10**).

The final prediction model for LGA was summarized by the following equation:

Log (Odds of LGA) = - 19.55+(0.0398*mother's weight) + (0.0630*mother's height) + (0.9495*mother's fasting glucose-(0.7013*primiparity) - (1.0712* mother's smoking status)

To use the model and find the log odds for LGA for a particular baby, the variables of mother's height, weight, fasting glucose, parity and mother's smoking status are inserted in the above equation, for that observation. Table 2 provides a summary of the full model. Log odds can be converted to a probability using the equation below:
Let the linear predictor, LP1

LP1 = - 19.55+(0.0398*mother's weight) + (0.0630*mother's height) + (0.9495*mother's fasting glucose-(0.7013*primiparity) - (1.0712* mother's smoking status)

Odds of LGA = exp (LP1)

Probability of LGA = exp (LP1)/ (1+exp (LP1))

Model description

Clinical features	Coefficient	Standard Error	Odds ratio (95%CI)	Wald Z	Pr(>|z|)
Intercept	-15.72	3.08	1.48e-07 (3.54e-10,6.25e-05)	-5.10	<0.0001
Mother's height (cm)	0.068	0.019	1.07 (1.03, 1.11)	3.64	0.0003
Mother's pre-pregnancy weight (kg)	0.040	0.0082	1.04 (1.02, 1.06)	4.94	<0.0001
Primiparity (1st Pregnancy vs. subsequent)	-0.824	0.25	0.439(0.271,0.711)	-3.34	0.0008
Mother's smoking status (Yes vs. no)	-1.00	0.44	0.367 (0.154, 0.874)	-2.26	0.0236

Table 2: Summary of the multivariable logistic regression model, First Trimester Model

The R^2 statistic for this First trimester model (**Table 2**) was 0.137. The odds ratio for mother's height and weight show that a unit increase in these features (by 1 cm and 1 kg, respectively), would result in an increase in odds of LGA by 1.07 and 1.04, respectively. In terms of parity, the odds of LGA decrease by 56% for each subsequent offspring and if the mother smoked during the pregnancy, the odds of her delivering a baby born LGA decreased by a factor of 63%.

Clinical features	Coefficient	Standard Error	Odds ratio (95%CI)	Wald Z	Pr(>|z|)
Intercept	-19.55	3.47	3.24e-09 (3.63e-12, 2.89e-06)	-5.64	<0.0001
Mother's height (cm)	0.063	0.018	1.07 (1.03, 1.10)	4.61	0.0006
Mother's weight (kg)	0.040	0.0086	1.04 (1.02,1.06)	3.43	<0.0001
Mother's fasting glucose (mmol/L)	0.950	0.33	2.58 (1.35, 4.95)	2.87	0.0042
Primiparity (1st Pregnancy vs. subsequent)	-0.701	0.24	0.496 (0.309, 0.795)	-2.91	0.0036
Mother's smoking status (Yes vs. no)	-1.07	0.43	0.34 (0.146, 0.803)	-2.47	0.0137

Table 3: Summary of the multivariable logistic regression model, Model 1

Table 3 shows the model with the routinely available clinical features in addition to maternal fasting glucose where glucose added significantly to the other routine clinical feature with each mmol/L of glucose leading to 2.6 times increase in the odds of LGA. The addition of glucose did not change the associations between the other variables and risk of LGA. The R^2 statistic for this model was 0.160.

Assessment of model performance

The area under the ROC curve for the model with glucose and clinical features was found to be 0.761. This is illustrated by Figure 2; where the ROC curve shows a plot of true positive rate (sensitivity) against true negative rate (specificity) and the calibration curve assessed how well the predicted probabilities agreed with the observed probabilities. The calibration curve illustrated that the predicted probabilities were closely fitted to the observed ones in Figure 3.

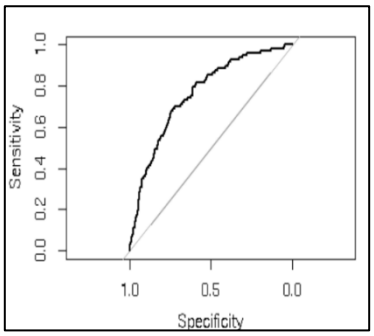


Figure 2: ROC Curve for model in EFSOCH

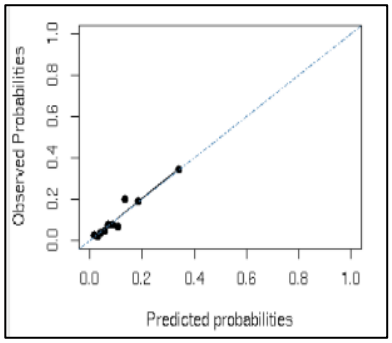


Figure 3: Calibration Curve for model in EFSOCH

Internal validation for model performance

The results of the bootstrap samples (sample size=250) indicated low levels of optimism due to little error because of overfitting; the apparent Somer's Dxy was 0.5219 and the bias-corrected Dxy was 0.5035. The maximum absolute error in predicted probability, Emax, was estimated to be 0.0242. (Harrell et al., 1984, Steyerberg et al., 2001)

Discrimination and validation in a European cohort

External validation was assessed by applying the final regression equation from EFSOCH into BiB cohort. Table 4 summarizes the characteristics of the participants from BiB (Europeans, n=1583 and Pakistanis, n=1115). On average,

babies of European ancestry weighed about 200g heavier than those of South Asian ancestry.

Phenotype	Europeans (n=1583) Mean or % (SD)	Pakistanis (n=1115) Mean or % (SD)
Maternal Height (cm)	164.5(6.3)	159.5(5.7)
Maternal weight(kg)	78.4(16.1)	70.6(13.1)
Gestational Duration (weeks)	39.7(1.2)	39.4(1.2)
Birth weight (g)	3470(492)	3195(445)
Smoking Status (% Yes)	31	4
Primiparity (%1st pregnancy)	49	34
Sex of the baby (% Male)	51	52

Table 4: Characteristics of participants in BiB (Europeans and Pakistanis)

In Europeans, the ROC area under the curve was 0.6745 (95% CI: 0.6351-0.7138). The model calibrated well overall with the mean predicted probability for LGA was found to be 0.110 which was similar to the actual percentage of LGA in the BiB European cohort (n=1583) which was 11.4%. The calibration was generally good, but the model overestimated slightly in the top decile. Mothers in the highest decile in BiB were heavier but had lower rates of primiparity than mothers in EFSOCH (Table 11). This showed that the model was performing well in a different cohort of similar ethnicity, but caution may be needed at the highest probabilities.

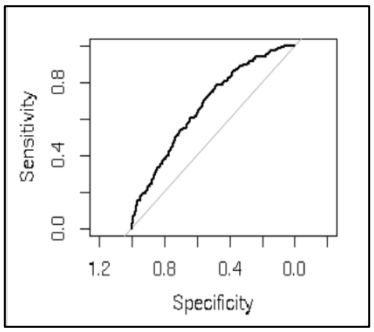


Figure 4: ROC Curve for external validation in a European cohort

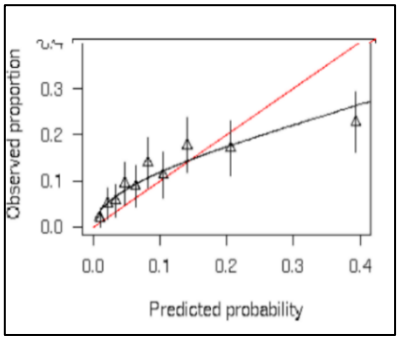


Figure 5: Calibration curve for external validation in a European cohort

The model externally validated well in a South Asian cohort but required a re-calibration to ensure the probabilities reflected the overall lower population risk of LGA in this cohort

External validation was assessed by the ROC curve and the area under the curve was 0. 707 (95% CI: 0.637-0.7769). The mean predicted probability for LGA was found to be 0.071 which was higher than the actual percentage of LGA in the BiB Pakistani cohort (n=1115) which was 3.5%, suggesting the model was over-estimating in this cohort with different ancestry. This is also further illustrated by the calibration curve (see **Figure 7**).

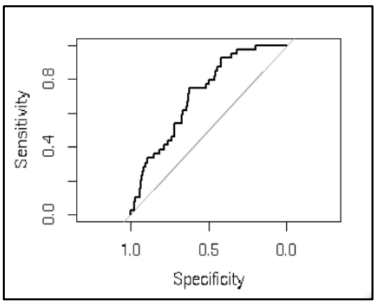


Figure 6: ROC Curve for external validation in the SA cohort

Re-calibration in the South Asian cohort

LP1 = - 19.55+(0.0398*mother's weight) + (0.0630*mother's height) + (0.9495*mother's fasting glucose-(0.7013*primiparity) - (1.0712* mother's smoking status) **[equation 1]**

$LGA \sim 0 + LP_1$ **[equation 2]**

$\hat{Y} = -1.3014 + 0.8746\ (LP1)$ **[equation 3]**

We applied recalibration, adjusting the intercept by –1.3014 and the slope by 0.8746(see Methods; **equations 1-3)** and the model fitted better in the South Asian cohort (see Figure 8). The area under the curve was 0. 707 (95% CI: 0.637-0.7769) and mean predicted probability for LGA was found to be 0.03498.

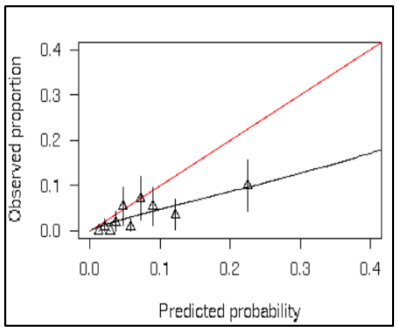


Figure 7: Calibration curve without re-calibration

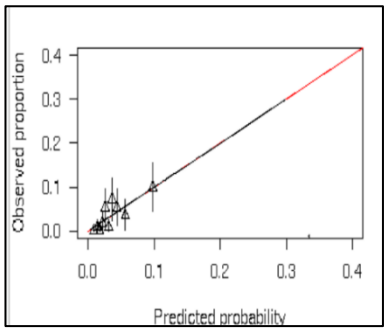


Figure 8: Calibration curve with re-calibration

Discussion

We have developed a clinical prediction model for LGA that discriminates well and independently validates in an external European population, and, with recalibration, in a South Asian population. Detection of LGA in current clinical practice includes following up with additional ultrasound scans, expectant mothers that classified for

an oral glucose tolerance test (OGTT) for risk of developing gestational diabetes (Rani and Begum, 2016). However, not all women with gestational diabetes will give birth to babies born LGA and not all LGA babies are born to women with gestational diabetes. Hence, the idea of developing a clinical prediction model for LGA for all women was considered. The model was built using data from the Exeter Family Study of Childhood Health (EFSOCH), internally and externally validated in the BiB European and South Asian cohorts.

This model was built to improve on prediction of LGA in current clinical practice, where although clinical prediction models have previously been built, no actual model is currently used to estimate LGA. Easily and routinely available clinical features; mother's height, mother's weight, mother's fasting glucose, parity and mother's smoking status were used to build this model. The outcome variable was large-for-gestational age (LGA). Like most models for LGA, this model also used the common predictors of mother's height, weight, parity and smoking status and in contrast to previous models, the model used maternal fasting glucose which had not been previously used in prediction models. It achieved an area under the curve of 0.761, which indicated a better performance than previously built clinical prediction models(Meertens et al., 2019).

The key idea was to have a model that is easy to use with the available information at hand in clinics. When a mother goes for a regular checkup at a clinic, it is easy to obtain information about her height, weight at 28 weeks', whether she has given birth before or not (parity) and her smoking status during the pregnancy. Mother's fasting glucose is also easily obtained at 28 weeks' on a clinic visit. Hence, the availability of all these variables makes this model very easy to implement and predict LGA. Another main strength of this model is the robust modeling that it underwent before being built (data cleaning and checking of model assumptions). The sample size for both the first trimester model (n=821) and the final model (n=906) were very good with the requirement being only a minimum sample size of 351 observations. A good prediction model is one that makes valid predictions for

new subjects in novel settings. and it was found that the model externally validated well in a different cohort of the same ancestry (European); if Model 1 was applied in a different population of similar ancestry, it would still perform well in estimating the odds of LGA (probability of estimated LGA was 0.114). The predicted probabilities were slightly overestimated for the top decile in BiB Europeans (about 0.4) as compared to the observed probability (about 0.2) and it is unclear why as birthweights were lower on average in this decile in BiB, as maternal weights were higher and there were lower rates of primiparity compared to EFSOCH (see **Table 11**).

Furthermore, external validation in a cohort of different ancestry (South Asian), further improved on the generalizability of our prediction model. An initial try gave a good discrimination, but the model overestimated on average (probability of LGA=0.071) but this was easily resolved if a re-calibration was applied which then produced a mean predicted probability of LGA very close to the actual probability of LGA in a South Asian population (probability of LGA=0.035). In general, the predictive performance of a prediction model is often decreased in new patients because the outcome incidence is different in the validating cohort. Overall, this model can be summarized as easy to implement in both a European and South Asian cohort, with good predictive ability.

While this was a good model, there are still limitations that need to be addressed. One of the limitations of this model is its use being limited to the third trimester, when the variables for fasting glucose and weight were available. A first trimester model that we developed (see **Table 2**) showed that while using mother's pre-pregnancy weight and excluding maternal fasting glucose could still give a good prediction for LGA (R^2 =0.137, AUCROC=0.732), the addition of maternal fasting glucose to the model did significantly improve prediction of LGA (R^2 =0.160; AUCROC=0.761). Also, the model was built in a European population and only tested in a South Asian population. LGA is a common problem affecting a lot of other ancestries which this model did not address. In terms of other risk groups, mothers with diagnosed gestational diabetes (Sridhar et al., 2013) and those who

could classify as being obese (Hong and Lee, 2021) who are high risk groups for LGA, were not considered. Finally, while the model was considered to have a good predictive ability with an area under the curve of 0.761, this could still be improved.

While our model assessed the gap in literature in using maternal fasting glucose as a predictor for LGA in a clinical prediction model, there remain other areas to be explored. Other factors that interplay with fetal growth such as genetics, intrauterine environment, nutrition, and placental function need to be further assessed. No other clinical prediction models have assessed the contribution of non-genetic and genetic factors in relation to LGA while being born LGA has been found to be associated with increased global placental function of DNA methylation(Dwi Putra et al., 2020, Haulder et al., 2022). Additionally, in order to implement this model in clinical practice, an online calculator could be developed. Finally, the model has considered only healthy mothers with no diabetes but in a realistic population, some mothers would have diabetes and it would be worth assessing how the model would work in a population with diabetic mothers.

In summary, we have found that improving on current clinical practice for predicting LGA can be done using a clinical prediction model developed from routinely available clinical features. This model can be easily implemented in a European population. It can also be used to effectively predict LGA in a South Asian population, with the application of a simple re-calibration. By building this model, we were also able to explore the significant contribution of maternal fasting glucose in a prediction model for LGA, which had not been assessed before. It was found that the use of simple, routinely available clinical features could be used in combination to predict LGA. The simple model also demonstrated that it was better to use a combination of a few predictors to accurately predict LGA than a complex model. Although the model had good predictive ability, it still contained several limitations and there was still room for further improvement by the addition of other factors such as genetics.

Supplementary information

Linearity check in multivariable logistic regression model

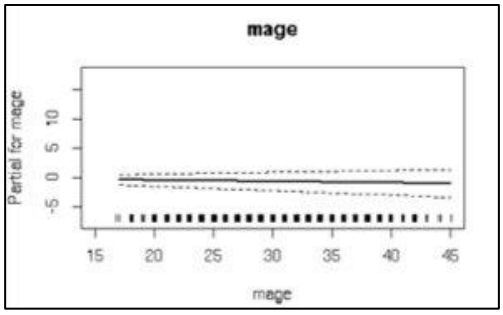


Figure 1: Plot of log odds of LGA against mother's age

The plot shows that the variable of mother's age in EFSOCH is linearly associated with log odds of LGA.

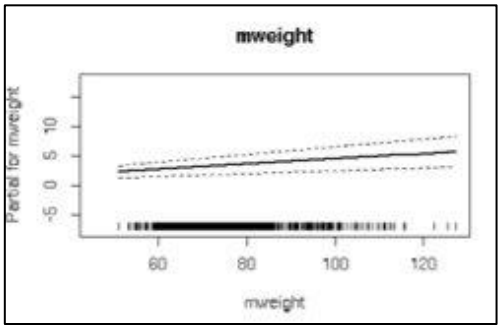


Figure 2: Plot of log odds of LGA against mother's weight

The plot shows that the variable of mother's weight in EFSOCH is linearly associated with log odds of LGA.

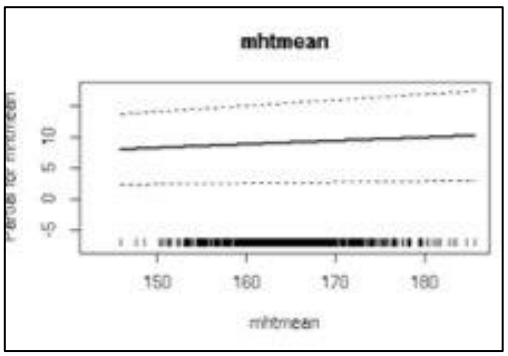


Figure 3: Plot of log odds of LGA against mother's age

The plot shows that the variable of mother's height in EFSOCH is linearly associated with log odds of LGA.

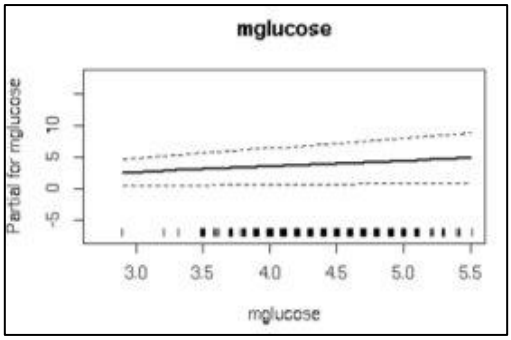


Figure 4: Plot of log odds of LGA against mother's fasting glucose

The plot shows that the variable of mother's fasting glucose in EFSOCH is linearly associated with log odds of LGA.

Phenotype	Included (n=906) Mean or %	Excluded (n=116) Mean or %	P-value
Maternal Height (cm)	165.0 (6.3)	164.7 (6.1)	0.576
Maternal weight(kg)	76.1 (12.8)	75.0(12.8)	0.443
Gestational Duration (weeks)	40.1 (1.2)	37.5 (2.7)	<0.001
Maternal fasting glucose	4.3(0.4)	4.5(0.6)	0.003

Birth weight (g)	3508 (475)	2972(694)	<0.001
Smoking Status (% Yes)	13	17	0.354
Primiparity (%1st pregnancy)	45	53	0.602
Sex of the baby (% Male)	52	46	0.256

Table 5: Comparison between the included participants and the excluded participants

Blood pressure exploration

Mother's average SBP and DBP were computed and analyzed to determine their relationship with LGA. The following tables show the analyses with several models having been tested to determine the contribution of Mother's SBP and DBP in predicting LGA.

Phenotype	Coefficient	Wald	Pr(>|z|)
Intercept	-18.6	-5.13	<0.0001
Mother's height (cm)	0.043	4.55	<0.0001
Mother's weight (kg)	0.056	2.90	0.037
Mother's fasting glucose (mmol/L)	0.804	2.27	0.024
Parity (1st Pregnancy)	0.779	3.07	0.022
Mother's smoking status (Yes)	-1.00	-2.13	0.033

Table 6: Multivariable logistic regression model with clinical features only (n=775)

Phenotype	Coefficient	Wald	Pr(>|z|)
Intercept	-15.8	-5.13	<0.0001
Mother's height (cm)	0.052	4.55	<0.0001
Mother's weight (kg)	0.049	2.90	0.037
Mother's fasting glucose (mmol/L)	0.834	2.27	0.024
Parity (1st Pregnancy)	0.783	3.07	0.022
Mother's smoking status (Yes)	-1.11	-2.13	0.033

Mother's systolic blood pressure (Hg/mm)	-0.0103	-0.96	0.336

Table 7: Multivariable logistic regression model with clinical features and systolic blood pressure (n=775)

Phenotype	Coefficient	Odds ratio	Wald Z	Pr(>|z|)
Intercept	-15.76(-23.2,-8.31)	$1.43x10^{-7}$	-4.15	<0.0001
Mother's height (cm)	0.049(-0.047,0.144)	1.05	5.08	0.0128
Mother's weight (kg)	0.052(0.032,0.072)	1.05	2.49	<0.0001
Mother's fasting glucose (mmol/L)	0.834(0.14,1.53)	2.30	2.34	0.0192
Parity (subsequent pregnancy)	0.783(0.28,1.28)	2.19	3.07	0.0021
Mother's smoking status (Yes)	-1.11(-2.04,-0.18)	0.33	-2.33	0.0199
Mother's Diastolic BP	-0.0381 (-0.068,-0.0077)	0.96	-2.45	0.0141

Table 8: Multivariable logistic regression model with clinical features and diastolic blood pressure (n=775)

Mother's systolic blood pressure was not found to be a good predictor of LGA and was therefore, dropped from the final model.

Scan data exploration

Abdominal circumference (AC) and head circumference (HC) were analyzed to investigate their relationship with LGA. The following tables show the analyses with several models having been tested to determine the contribution of AC and HC in predicting LGA.

Phenotype	Coefficient	Wald	Pr(>|z|)
Intercept	-18.6	-4.82	<0.0001
Mother's height (cm)	0.043	4.41	<0.0001
Mother's weight (kg)	0.060	3.03	0.0024
Mother's fasting glucose (mmol/L)	0.944	2.57	0.0101
Parity (1st Pregnancy)	0.811	3.09	0.002
Mother's smoking status (Yes)	-1.11	-2.18	0.029
Abdominal Circumference (mm)	-0.0096	-1.38	0.167

Table 9: Multivariable logistic regression model with clinical features and abdominal circumference (n=750)

Phenotype	Coefficient	Wald	Pr(>|z|)
Intercept	-18.1	-4.67	<0.0001
Mother's height (cm)	0.058	4.51	0.0038
Mother's weight (kg)	0.044	2.89	<0.0001
Mother's fasting glucose (mmol/L)	0.905	2.47	0.0135
Parity (1st Pregnancy)	0.792	3.01	0.0026

Mother's smoking status (Yes)	-0.927	-1.95	0.0507
Head circumference (mm)	-0.0077	-1.16	0.248

Table 10: Multivariable logistic regression model with clinical features and head circumference (n=747)

Analysis of the scan data from the EFSOCH study showed that the abdominal circumference (AC) and the head circumference (HC) were not good predictors of LGA, and they were therefore removed from the final model.

Comparison between last deciles of EFOSCH and BiB

Mean predicted probability range for last decile in EFSOCH (0.226,0.729]

Mean predicted probability range for last decile in BiB Europeans (0.222,0.596]

Phenotype	EFSOCH(n=91) Mean or %	BiB (n=159) Mean or %
Maternal Height (cm)	171.0(6.1)	168.8(5.3)
Maternal weight(kg)	96.3 (12.8)	105.8(14.8)
Gestational Duration (weeks)	40.1 (1.2)	40.0(1.1)
Maternal fasting glucose	4.7(0.3)	4.7(0.4)
Birth weight (g)	3905 (479)	3793(502)
Smoking Status (% Yes)	4	6
Primiparity (%1st pregnancy)	85	77
Sex of the baby (% Male)	57	57

Table 11: Comparison of characteristics for the last decile between EFSOCH and BiB

Appendix

TRIPOD Checklist: Prediction Model Development and Validation

Section/Topic	Item		Checklist Item	Page
Title and abstract				
Title	1	D;V	Identify the study as developing and/or validating a multivariable prediction model, the target population, and the outcome to be predicted.	
Abstract	2	D;V	Provide a summary of objectives, study design, setting, participants, sample size, predictors, outcome, statistical analysis, results, and conclusions.	
Introduction				
Background and objectives	3a	D;V	Explain the medical context (including whether diagnostic or prognostic) and rationale for developing or validating the multivariable prediction model, including references to existing models.	
	3b	D;V	Specify the objectives, including whether the study describes the development or validation of the model or both.	
Methods				
Source of data	4a	D;V	Describe the study design or source of data (e.g., randomized trial, cohort, or registry data), separately for the development and validation data sets, if applicable.	
	4b	D;V	Specify the key study dates, including start of accrual; end of accrual; and, if applicable, end of follow-up.	
Participants	5a	D;V	Specify key elements of the study setting (e.g., primary care, secondary care, general population) including number and location of centres.	
	5b	D;V	Describe eligibility criteria for participants.	
	5c	D;V	Give details of treatments received, if relevant.	
Outcome	6a	D;V	Clearly define the outcome that is predicted by the prediction model, including how and when assessed.	
	6b	D;V	Report any actions to blind assessment of the outcome to be predicted.	
Predictors	7a	D;V	Clearly define all predictors used in developing or validating the multivariable prediction model, including how and when they were measured.	
	7b	D;V	Report any actions to blind assessment of predictors for the outcome and other predictors.	
Sample size	8	D;V	Explain how the study size was arrived at.	
Missing data	9	D;V	Describe how missing data were handled (e.g., complete-case analysis, single imputation, multiple imputation) with details of any imputation method.	
Statistical analysis methods	10a	D	Describe how predictors were handled in the analyses.	
	10b	D	Specify type of model, all model-building procedures (including any predictor selection), and method for internal validation.	
	10c	V	For validation, describe how the predictions were calculated.	
	10d	D;V	Specify all measures used to assess model performance and, if relevant, to compare multiple models.	
	10e	V	Describe any model updating (e.g., recalibration) arising from the validation, if done.	
Risk groups	11	D;V	Provide details on how risk groups were created, if done.	
Development vs. validation	12	V	For validation, identify any differences from the development data in setting, eligibility criteria, outcome, and predictors.	
Results				
Participants	13a	D;V	Describe the flow of participants through the study, including the number of participants with and without the outcome and, if applicable, a summary of the follow-up time. A diagram may be helpful.	
	13b	D;V	Describe the characteristics of the participants (basic demographics, clinical features, available predictors), including the number of participants with missing data for predictors and outcome.	
	13c	V	For validation, show a comparison with the development data of the distribution of important variables (demographics, predictors and outcome).	
Model development	14a	D	Specify the number of participants and outcome events in each analysis.	
	14b	D	If done, report the unadjusted association between each candidate predictor and outcome.	
Model specification	15a	D	Present the full prediction model to allow predictions for individuals (i.e., all regression coefficients, and model intercept or baseline survival at a given time point).	
	15b	D	Explain how to the use the prediction model.	
Model performance	16	D;V	Report performance measures (with CIs) for the prediction model.	
Model-updating	17	V	If done, report the results from any model updating (i.e., model specification, model performance).	
Discussion				
Limitations	18	D;V	Discuss any limitations of the study (such as nonrepresentative sample, few events per predictor, missing data).	
Interpretation	19a	V	For validation, discuss the results with reference to performance in the development data, and any other validation data.	
	19b	D;V	Give an overall interpretation of the results, considering objectives, limitations, results from similar studies, and other relevant evidence.	
Implications	20	D;V	Discuss the potential clinical use of the model and implications for future research.	
Other information				
Supplementary information	21	D;V	Provide information about the availability of supplementary resources, such as study protocol, Web calculator, and data sets.	
Funding	22	D;V	Give the source of funding and the role of the funders for the present study.	

*Items relevant only to the development of a prediction model are denoted by D, items relating solely to a validation of a prediction model are denoted by V, and items relating to both are denoted D;V. We recommend using the TRIPOD Checklist in conjunction with the TRIPOD Explanation and Elaboration document.

References

Dwi Putra, S. E., C. Reichetzeder, A. A. Hasan, T. Slowinski, C. Chu, B. K. Kramer, B. Kleuser and B. Hocher (2020). "Being Born Large for Gestational Age is Associated with Increased Global Placental DNA Methylation." Sci Rep **10**(1): 927.

Frick, A. P., A. Syngelaki, M. Zheng, L. C. Poon and K. H. Nicolaides (2016). "Prediction of large-for-gestational-age neonates: screening by maternal factors and biomarkers in the three trimesters of pregnancy." Ultrasound Obstet Gynecol **47**(3): 332-339.

Gururajan, P., P. Gurumurthy, P. Nayar, G. S. Rao, R. S. Babu, A. Sarasabharati and K. M. Cherian (2012). "Pregnancy associated plasma protein-A (PAPP-A) as an early marker for the diagnosis of acute coronary syndrome." Indian Heart J **64**(2): 141-145.

Harrell, F. E., Jr., K. L. Lee, R. M. Califf, D. B. Pryor and R. A. Rosati (1984). "Regression modelling strategies for improved prognostic prediction." Stat Med **3**(2): 143-152.

Haulder, M., A. E. Hughes, R. N. Beaumont, B. A. Knight, A. T. Hattersley, B. M. Shields and R. M. Freathy (2022). "Assessing whether genetic scores explain extra variation in birthweight, when added to clinical and anthropometric measures." BMC Pediatr **22**(1): 504.

Hong, Y. H. and J. E. Lee (2021). "Large for Gestational Age and Obesity-Related Comorbidities." J Obes Metab Syndr **30**(2): 124-131.

Hughes, A. E., M. Nodzenski, R. N. Beaumont, O. Talbot, B. M. Shields, D. M. Scholtens, B. A. Knight, W. L. Lowe, Jr., A. T. Hattersley and R. M. Freathy (2018). "Fetal Genotype and Maternal Glucose Have Independent and Additive Effects on Birth Weight." Diabetes **67**(5): 1024-1029.

Knight, B., B. M. Shields and A. T. Hattersley (2006). "The Exeter Family Study of Childhood Health (EFSOCH): study protocol and methodology." Paediatr Perinat Epidemiol **20**(2): 172-179.

Meertens, L., L. Smits, S. van Kuijk, R. Aardenburg, I. van Dooren, J. Langenveld, I. M. Zwaan, M. Spaanderman and H. Scheepers (2019). "External validation and clinical usefulness of first-trimester prediction models for small- and large-for-gestational-age infants: a prospective cohort study." BJOG **126**(4): 472-484.

Mi, D., H. Fang, Y. Zhao and L. Zhong (2017). "Birth weight and type 2 diabetes: A meta-analysis." Exp Ther Med **14**(6): 5313-5320.
Papastefanou, I., A. P. Souka, A. Pilalis, M. Eleftheriades, V. Michalitsi and D. Kassanos (2012). "First trimester prediction of small- and large-for-gestation neonates by an integrated model incorporating ultrasound parameters, biochemical indices and maternal characteristics." Acta Obstet Gynecol Scand **91**(1): 104-111.
Plasencia, W., E. Gonzalez Davila, V. Tetilla, E. Padron Perez, J. A. Garcia Hernandez and N. L. Gonzalez Gonzalez (2012). "First-trimester screening for large-for-gestational-age infants." Ultrasound Obstet Gynecol **39**(4): 389-395.
Poon, L. C., G. Karagiannis, V. Stratieva, A. Syngelaki and K. H. Nicolaides (2011). "First-trimester prediction of macrosomia." Fetal Diagn Ther **29**(2): 139-147.
Rani, P. R. and J. Begum (2016). "Screening and Diagnosis of Gestational Diabetes Mellitus, Where Do We Stand." J Clin Diagn Res **10**(4): QE01-04.
Riley, R. D., J. Ensor, K. I. E. Snell, F. E. Harrell, Jr., G. P. Martin, J. B. Reitsma, K. G. M. Moons, G. Collins and M. van Smeden (2020). "Calculating the sample size required for developing a clinical prediction model." BMJ **368**: m441.
Sridhar, S. B., A. Ferrara, S. F. Ehrlich, S. D. Brown and M. M. Hedderson (2013). "Risk of large-for-gestational-age newborns in women with gestational diabetes by race and ethnicity and body mass index categories." Obstet Gynecol **121**(6): 1255-1262.
Steyerberg, E. W., F. E. Harrell, Jr., G. J. Borsboom, M. J. Eijkemans, Y. Vergouwe and J. D. Habbema (2001). "Internal validation of predictive models: efficiency of some procedures for logistic regression analysis." J Clin Epidemiol **54**(8): 774-781.
Wright, C. M., A. F. Williams, D. Elliman, H. Bedford, E. Birks, G. Butler, M. Sachs, R. J. Moy and T. J. Cole (2010). "Using the new UK-WHO growth charts." BMJ **340**: c1140.
Wright, J., N. Small, P. Raynor, D. Tuffnell, R. Bhopal, N. Cameron, L. Fairley, D. A. Lawlor, R. Parslow, E. S. Petherick, K. E. Pickett, D. Waiblinger, J. West and G. Born in Bradford Scientific Collaborators (2013). "Cohort Profile: the Born in Bradford multi-ethnic family cohort study." Int J Epidemiol **42**(4): 978-991.

Chapter 6

Discussion, future results, and conclusions

Discussion

There are several factors that influence a baby's birthweight: the fetal genotype and the maternal genotype directly influence birthweight via transmission to the fetus and indirectly influence maternal characteristics such as glucose levels which may then affect birth weight. Several maternal characteristics and environmental factors influence birthweight via the maternal intrauterine environment. Conducted GWAS of own birthweight and offspring birthweight have identified 243 loci directly associated with birthweight, indicating the importance of genetics in relation to birthweight. Previous studies carried out have demonstrated that there are several other key factors that explain the variation in offspring birthweight: maternal characteristics, environmental factors, and social demographics. The third chapter of this thesis covers the gap between assessing the contribution of genetics to birthweight alongside other clinical features. Ethnicity is also an important predictor of birthweight and what constitutes a small baby in one ancestral link might not be a small baby in a different ancestry. While previous research has demonstrated similar contributions of genetics to offspring birthweight in White Europeans and South Asians, the fourth chapter of this thesis compares the contribution of genetics in these two ethnic groups alongside other clinical features. It is desirable to be able to predict a baby's birthweight before its birth to cater for complications with delivery. Thus, it is possible to accommodate for any adverse pregnancy outcomes or postnatal care. Methods used in current clinical practice are not entirely effective in assessing small and large-for-gestational age babies. The fifth chapter expands on building a clinical prediction model for large-for-gestational age babies in European babies by using routinely available maternal characteristics in clinics and validating it in both European and SA mothers and babies. In summary, this thesis addresses some of the main questions in relation to contribution of genetics to birthweight alongside other predictors of birthweight, and it also investigates the implementation of a clinical prediction model which uses maternal fasting glucose as one of its main predictors in determining risk of LGA in Europeans and South Asian populations.

Chapter 3: Assessing whether genetic scores explain extra variation in birthweight, when added to clinical and anthropometric measures

Summary

This chapter addresses the contribution of genetics alongside other key maternal characteristics to offspring birthweight. Several maternal characteristics such as mother's weight, BMI, height, fasting glucose, smoking status during pregnancy and parity have previously been found to influence birthweight through the maternal intrauterine environment (Cogswell and Yip, 1995). We also know that a baby's birthweight is partly explained by its inherited set of genes. It has been 18 years since the publication of the first Genome Wide Association Study (GWAS) in 2005 (Klein et al., 2005) and this method of making genetic information easily accessible has enabled us to quantify genetic information and create genetic scores. Individually, maternal characteristics and genetics have been analyzed in terms of their effects on birthweight (Cogswell and Yip, 1995) but the genetic effects that contribute to birthweight alongside clinical features have not been assessed. To do this, we aimed to investigate variance in birthweight explained by genetic scores in addition to easily measurable clinical and anthropometric variables in a European group by using multivariable linear regression models. It was found that genetics explain additional variance on top of that explained by routinely available clinical features.

Conclusions

This study, which is now published (Haulder et al., 2022) found that a multivariable linear regression model containing maternal age, weight, smoking status, parity, and 28-week fasting glucose explained 21.7% of variation in birthweight in a European population. Maternal and fetal genetic scores together, just on their own, explained a 6.0% variance in birthweight. Also, the maternal genetic score explained additional variance in birthweight when added to maternal characteristics (Adj-R^2 = 0.233 vs Adj-R^2 = 0.210, $p < 0.001$) similarly to the fetal genetic score

which also improved explanation of variance in birthweight (Adj-R^2 = 0.264 vs 0.248, $p < 0.001$) when added to maternal characteristics and parental heights. Mother's height and father's height partially explain the fetus' genetic contribution to birthweight but in general, the genetic scores explain additional variance.

Implications of the findings

The results clearly demonstrate how genetics play a key role in explaining the variation in birthweight independent of what is captured from routinely available clinical features. On their own, a small proportion of variation in birthweight is explained (6.0%) whereas in addition to other clinical features, the explanation in variation is much more (21.7%). This result highlights how clinical features play a significant role in explaining offspring birthweight and the contribution of genetics on their own is not as high. This is in line with other findings which state that offspring birthweight is a product of several factors such as environmental and genetic rather than just one set of predictors (Spencer and Logan, 2002). Also, while the contribution of genetics is not a lot (3% of variance explained for maternal genetics and 2% for fetal genetics), it is still comparable to other predictors such as parity (3% of variation explained) which makes it an important variable in explaining birthweight. The association between maternal height and fetal growth has been found to be mainly defined by fetal genetics (Zhang et al., 2015) and it is also important to note that some of the maternal height effect is independent of fetal genetics(Warrington et al., 2019). Paternal height has been found to independently (but not independently of fetal genetics) influence birth size (Knight et al., 2005). Our results show that parental heights with clinical features explained 24.8% of variance in birthweight as compared to clinical features on their own which explained 21.7% of variance. However, when fetal genetics is added to a model with clinical features and parental heights, there was additional variance explained (27.7%) which suggests that parental heights capture features of fetal genetics only partially. Maternal genetics with paternal genetics on top of clinical features are only slightly better at explaining birthweight variation and the best

model uses clinical features, parental heights, maternal genetics and fetal genetics for the maximum explanation of variance in birthweight.

We also found that in contrast with a previous study(Griffiths et al., 2007) where the parental heights made similar contributions to birthweight, father's height makes a higher and significant contribution to birthweight when compared to mother's height, that is a unit change in father's height resulted in 60g (23, 98) increase in birthweight as compared to only 23g (13, 62) for a unit increase in mother's height. Mother's birthweight has a stronger association with birthweight compared to father's birthweight where a unit increase in mother's birthweight resulted in 103g (66, 140) increase in birthweight and a unit increase in father's birthweight resulted in 47g (10, 83) increase in birthweight. The results for parental birthweights are not statistically significant as the confidence intervals for both maternal and paternal birthweights do not include the values of paternal birthweight and maternal birthweight, respectively. This could be due to the small sample size available for parental birthweights. In contrast, a previous study found that parental birthweights significantly influenced offspring birthweight (Little, 1987).

Strengths and limitations of the work

This study comprehensively assessed the contributions of maternal characteristics to birthweight alongside maternal and fetal genetics whereas previous studies have either assessed the contributions of clinical features on their own (Cogswell and Yip, 1995) or genetics on their own(Beaumont et al., 2018). It gave insight on the contributions of maternal, paternal and fetal genetics to birthweight on top of routinely available clinical features.

In addition to this, this study further investigated the contributions of paternal genetics as well as the father's and mother's height, father's and mother's birthweight to birthweight using multivariable linear regression models. Previously carried out studies related to birthweight focused more on the child and mother rather than the father as there is limited amount of paternal information available (Magnus et al., 2001).

The genetic scores used in this study were created using GWAS of European ancestry(Warrington et al., 2019) and if applied to other ancestries, the GS created for these other ethnic groups would not explain variation in birthweight as well as when GS are created for Europeans using GWAS of European ancestry because the underlying genetic associations might be different in every ethnic group (Uffelmann et al., 2021).

The smoking status of the mother is the only environmental factor considered in this study while other factors such as social demographics, such as education level, have not been assessed. Previous studies (Cogswell and Yip, 1995) have shown that a parent's education level is an important factor in determining variance in birthweight. This was not available in EFSOCH and therefore, was not included in modeling of birthweight.

Also, the parental birthweights were self-reported which resulted in smaller sample sizes and less accurate models. It is important to have an accurate sample size because this minimizes the risk of a false-negative finding, that minimizes the risk of Type 2 error, or it helps estimate how precise the study is (Biau et al., 2008).

Future work

Other predictors of environmental factors and social demographics such as education level attainment could also be included in a multivariable linear regression model in future work to assess their contributions in explaining birthweight on top of clinical variables and genetics. Education level is an indication of the quality of life of the mother or the possible lifestyle she would be eligible for; for example, a higher education level would signal better access to healthcare facilities and better diet. This could then give us an indication of whether a specific lifestyle significantly influences variation in birthweight.

Fetal genetics were included in the analyses in the form of fetal genetic scores, but the genetic information of the baby is usually not available before birth. Currently used in the screening of down syndrome, cell-free fetal DNA (cffDNA), that is, fetal

DNA which freely circulates in the blood, can be the source of fetal genetic material for prenatal diagnosis (Everett and Chitty, 2015). In the future, this could be used for extracting fetal genetic information prior to birth and moving forward this would be a significant step in making genetic information available for use in pregnancy. At the moment, this is being done to test for Down Syndrome and other rare conditions. Also, currently, the availability of genetics in pregnancy is limited to studies that have investigated genetic disorders in the prenatal period (Wojcik et al., 2020) (Hoffner and Surti, 2012),the genetic conflicts of gene expression in pregnancy, genetic abnormalities that could cause pregnancy loss (Tise and Byers, 2021)and the genetics and epigenetics of pregnancy-associated diseases such as pre-eclampsia(van Dijk and Oudejans, 2013).

Limited studies have investigated common SNPs that influence both birthweight and another trait in the mother. The GCK gene has a common haplotype that influences both birthweight and maternal fasting glucose (Weedon et al., 2006) but there is still a lot to uncover in terms of the genetics and common SNPs that influence birthweight and other traits.

Chapter 4: The contributions of genetic scores, maternal glycemia, and other maternal characteristics to variation in birth weight in South Asian compared with European babies

Summary

Babies of South Asian ancestry weigh less than (about 200g difference) those of European ancestry even though South Asian (SA) mothers have higher maternal fasting glucose levels, higher parity, and lower smoking levels. The genetic associations alongside other maternal characteristics with birthweight were compared between the two groups. We have previously assessed the contributions of routinely available clinical features to birthweight in babies of European ancestry and the contributions of genetics on top of these features(Haulder et al., 2022). The contributions of maternal and fetal genetics to birthweight have been explored in the SA group (Nongmaithem et al., 2022) but the contributions of genetics on top of clinical features have not been assessed before nor compared to the European group. The main aim of this project was to compare the associations with birthweight of mother's age, height, parity, smoking status during the pregnancy, maternal genetics and fetal genetics between SA and European babies. Maternal and fetal genetics were represented by maternal and fetal genetic scores (GS), and these were derived from the same GWAS of birthweight of European ancestry for both groups. Multivariable linear regression models were then used to build separate models in the two ethnic groups and make comparisons. Overall, the associations were similar in both groups except for parity and smoking status.

Conclusions

In both ethnic groups, fetal and maternal genetic scores showed similar associations with birthweight. Birthweight, on average, was lower in SAs compared with Europeans (3193(445) g vs. 3475 (483) g; $p<0.001$), and Fasting Plasma Glucose (FPG) was higher (4.6(0.5) vs. 4.4(0.4) mmol/L, $p<0.001$). Except for smoking and parity, the other characteristics had similar associations with

birthweight in both groups. Maternal FPG and adjusted birthweight demonstrated similar contributions in both populations (SA v European regression coefficient 81g [95%CI: 39, 102] per 1 SD higher FPG vs. 62g [39, 91] per 1 SD higher FPG; variance explained 2.2% v 3.3%). GS explained additional information to variation in birthweight but the full model in SA explained less overall variation in birthweight compared to Europeans (Adj-R^2 = 0.175 vs 0.223). Overall, we also found that the underlying difference between birthweights cannot be explained by genetics.

Implications of the findings

The characteristics all had similar associations to birthweight in SA and Europeans except for smoking and parity. This implies that birthweight is mostly explained to the same extent in both ethnic groups by the assessed characteristics. Smoking contributes less to birthweight variation in Europeans as compared to SA, likely due to the low prevalence of smoking in the SA group (add the amount). This is in line with previous findings that have found that only about 1·2% (0·7–1·7) SA mothers smoke during pregnancy (Lange et al., 2018).

Despite a higher mean fasting glucose level in SAs, higher parity and lower rate of smoking among the pregnant mothers, mean birthweight of SA babies is smaller than that of European babies. It was suggested to explore the genetic associations on top of the routinely available clinical features, and since they were the same, this was consistent with a previous study (Nongmaithem et al., 2022) where it has been found that the underlying difference in birthweight between SAs and Europeans was not explained by genetics.

In the SA group, mothers tend to deliver more babies than in the European group and it has been found that birthweight increases with increasing parity (Hinkle et al., 2014). Birthweight is larger with increasing parity. The results show that the association between birthweight and parity is less in SAs than in Europeans since

a higher number of babies means overall, less influence on reducing birthweight.

The fetal genetic score showed a slightly stronger association with birthweight in Europeans than in the SAs and the maternal genetic score had a similar association with birthweight in both SAs and Europeans. Similarly, to what has been found before, fetal genetics contributes more to birthweight than maternal genetics (Warrington et al., 2019) and the genetic scores give more accurate results when they have been created from GWAS of matching ancestry(Warrington et al., 2019). In this case, since both the scores were created with GWAS of European ancestry, although the associations are not far apart in the two ethnic groups, Europeans show a slightly stronger association, most likely as the genetic score was more accurate for that ethnic group.

The maternal genetic score was associated with birthweight to the same extent in South Asians and Europeans. This was different from what was found in a previous study where the maternal GS was less strongly associated with birthweight in SA than in Europeans when the GS were assessed on their own. The one possible explanation for different findings could be because in this study, the samples being analysed were from the same city in the UK, hence making them similar to one another while in the other study, the samples were from various locations (mostly outside the UK). (Nongmaithem et al., 2022)

Strengths and limitations

This study assessed SA and European babies who share the same environment (City of Bradford). Comparisons of the contributions of maternal characteristics in both ethnic groups are more accurate than studies which have used cohorts from different geographical locations (Babies of South Asian and European ancestry show similar associations with genetic risk score for birth weight despite the smaller size of South Asian newborns(Nongmaithem et al., 2022). When these groups share the same environment, the differences due to environmental exposures are removed and hence, makes the groups quite comparable. Both cohorts had a good sample size for accurate comparisons to be made and these

were consistent with previous studies where the sample sizes were about the same and generated similar power(Nongmaithem et al., 2022).

The study addressed an ethnic group which is at high risk of adverse pregnancy outcomes. A study carried out has found that women of SA ethnicity are at higher risk of adverse pregnancy outcomes such as prenatal death (De Graaff et al., 2023). It gave useful insight on how genetics further explain variance in birthweight on top of other routinely available clinical features. Previous studies have explored interventions in terms of diabetes in this group (Greenhalgh et al., 2015), prevalence of GDM in women of SA ancestry and other related pregnancy complications (Ahkter et al., 1996) low birthweight in the SA group (Metgud et al., 2012)and genetic studies have assessed the relationship between DNA methylation and cardiometabolic traits (Fragoso-Bargas et al., 2021).

One of the limitations to this project was the fact that the individual contributions of the predictors to birthweight could not be directly comparable from the linear regression models. This is why the R-squared statistic from univariate analysis were used to make direct comparisons.

The genetic score in SA was created from GWAS of European ancestry. The accuracy of these genetic scores were, therefore, not as good as the genetic score for the European group as the two ethnicities have different underlying genetics (Tekola-Ayele et al., 2018).

Future work

Despite their lower birthweights, SAs have higher skinfold thickness(Anand et al., 2016). Further exploration of this could lead to better understanding of what causes the difference in birthweight between these two groups. One way of measuring skinfold thickness is by the measurement of favorable adiposity. Adiposity is also defined as body fat, and it is more desirable for it to be stored under the skin than around organs such as the liver and the heart. Genes dictate where this fat is stored, and this can then be termed as having “favourable adiposity” or “unfavourable adiposity” (Martin et al., 2021).

Favorable adiposity could be the next step in investigating the underlying mechanisms that causes the difference in birthweight as it has different patterns in SAs and Europeans (Anand et al., 2016). SAs have greater skinfold thickness than Europeans due to a higher glucose level and higher levels of maternal body fat. The thin-fat phenotype that has been put forward previously (Kurpad et al., 2011) suggests that it is possible to have lower birthweight despite a greater amount of fat and it amounts to the fat being added to an already thin frame. To investigate whether this hypothesis holds true in SAs as compared to Europeans, the contribution of adiposity to birthweight could be assessed by adding it to multivariable linear regression models alongside other characteristics. It would be interesting to compare the contribution of the genetic score of favorable adiposity to birthweight, in SAs and Europeans. To do this, we would need access to data that has recorded favorable and unfavorable adiposity and the GWAS of favorable and unfavorable adiposity to create the genetic scores.

Chapter 5: Developing and validating a multivariable clinical prediction model for babies born LGA in European and South Asian cohorts

Summary

It is important to be able to predict if a baby is at risk of LGA to reduce and prevent adverse pregnancy outcomes. Prediction models for LGA have been developed before but these are not used in clinical practice due to their limited clinical utility. The aim of this project was to develop a model for LGA using data from the Exeter Family Study of Childhood Health (EFSOCH) and to validate the model in an external European as well as a South Asian cohort (Born in Bradford Study) for generalizability. Compared to other prediction models that used a varied selection of predictors such as ultrasound, biomarkers and medical history in addition to maternal characteristics, the idea of this prediction model was that it would improve on current detection of LGA in everyone, and not just in mothers at risk of gestational diabetes and because it uses routinely available clinical features, including maternal fasting glucose, it would be fairly easy to implement in clinical practice. The final model achieved a good level of predictive performance (AUC=0.761), but it was not good enough to be implemented in clinical practice. It validated well internally, as well as externally in a European cohort but required a re-calibration to externally validate well in a South Asian cohort.

Conclusions

This clinical prediction model for LGA had an ROC curve of 0.761, suggesting good discrimination, but its predictive power as it stands, is not strong enough for it to be adopted in clinical practice as it did not reach 0.80. It also has a good predictive ability indicated by its internal validation of bootstrapping where Emax was 0.0242 and Somer's Dxy was 0.5035, respectively. Emax represents the maximum error in predicted probabilities and since the model's Emax was close to 0, this shows that there were not a lot of errors in predicted probabilities. Somer's Dxy is the rank correlation assessing the difference between predicted probabilities and observed responses; a value close to 1 indicates that the model can make

predictions close to actual values. The model externally validated well in a South Asian cohort with the area under the curve as 0. 707 (95% CI: 0.637-0.7769), but it required re-calibration as LGA was less common in the SA. The mean predicted probability for LGA was found to be 0.071 and 3.5% of LGA was determined after re-calibration.

Implications of the findings

The area under the curve of the model is 0.761 which is better than current published models for LGA built in European cohorts. Examples include AUC between 0.60 and 0.69 for six models in Netherlands(Meertens et al., 2019), AUC between 0.66 and 0.75 for twelve models in a Canadian population(Kuhle et al., 2018) and AUC was 0.67 for a prediction model based on the SCOPE study. (SCOPE is an international prospective cohort study involving centers in Auckland, New Zealand; Adelaide, Australia; London, Manchester and Leeds, UK; and Cork, Ireland (Vieira et al., 2017)).

The model's c-statistic shows that while it makes good predictions, it is not good enough to be used in clinical practice on its own (Shipe et al., 2019). However, in line with additional information and monitoring, it could then be implemented. In current clinical practice, pregnant women have their own customized growth chart which has been prepared based on information about their height, weight, ethnicity and parity. This chart is prepared to show the predicted pattern of growth of the baby during the pregnancy. The symphysis fundal height (that is, the distance between top of the womb and the bone at the front of the pelvis) is also measured during antenatal visits for women pregnant over 26 weeks and compared to the growth chart. If on more than two occasions, the measurement is greater than expected, the woman is then set up for an ultrasound scan. If the baby is possibly larger than the 90th centile, the mother is given an OGTT if she is less than 36 weeks pregnant (Okonofua and Akaba, 2021). This implies that only women with specific criteria based on their symphysis fundal height and ultrasound scan are further monitored or have preventive measures taken for risk of LGA. A study assessed the accuracy of ultrasound measurements in calculating the Estimated

Fetal Weight (EFW) and has found that while a Mean Percentage Error (MPE) within 5% of accuracy is desirable in practice, currently, the level of errors associated with ultrasound scan estimates of fetal birthweight is below 10% (Milner and Arezina, 2018). In contrast, this prediction model assesses any pregnant woman for risk of delivering a baby born LGA and not only those that meet certain criteria, and it avoids the uncertainty generated by ultrasound scans.

Other models for LGA have used the same predictors that have been used in this one (mother's smoking status, parity, mother's weight, mother's height(Meertens et al., 2019), but none of these that assess healthy pregnant women have used maternal fasting glucose. Models assessing mothers with GDM have used maternal fasting glucose as a predictor (Wang et al., 2023, Cooray et al., 2022).

The model uses maternal fasting glucose as one of its predictors and it added significantly to the other routine clinical feature with each mmol/L of glucose leading to 2.6 times increase in the odds of LGA. In line with what has been found before, maternal fasting glucose is a key driver of fetal growth (Tyrrell et al., 2016) and therefore, provides valuable additional predictive power in predicting LGA as compared to other prediction models that did not use maternal fasting glucose as a predictor.

The prediction model was developed in a European cohort and externally validated in both European and South Asian groups. While it validated well in the European group, providing a probability of having a baby born LGA as 0.110 which is very close to the true mean probability in the actual population (10%), a re-calibration was required in the SA group. This model can be used in both a European population and a SA one (but with the application of a re-calibration).

Strengths and limitations

This prognostic model was built based on a cohort of healthy pregnant women and can easily be used in two distinct ethnic groups, which makes it very good for generalizability. The model is, therefore, able to correctly reflect the new target population. As compared to other published models, only the models in one study

performed external validation (Meertens et al., 2019) and this was still within the same ethnic group. The other models only go as far as internal validation(Wang et al., 2023, Wahab et al., 2022, Kuhle et al., 2018).

In terms of different ethnic groups, the model was successfully validated in a group with high risk of GDM(Fragoso-Bargas et al., 2021). This model uses maternal fasting glucose as a predictor in healthy pregnant women whereas this predictor has only been used before, in models with women having GDM (Cooray et al., 2022).

Many of the previously published models do not mention checking of assumptions such as sample size. The sample size for this model is very good when compared to many prediction models where model assumptions like these were not checked before the model was created. About 73% (95% CI: 63–82%) of 94 assessed prediction models with binary outcomes used smaller sample sizes than required to estimate risk of event(Dhiman et al., 2023). The clinical usefulness of this prognostic model was not checked. While a value of 0.761 for the c-statistic can indicate that a model would not be clinically ideal, this statistic on its own is not enough to assess the clinical usefulness of the model (Shipe et al., 2019).

Future work

The next step in this project would be to use decision curve analysis to formally evaluate the clinical utility of this model. This is to assess whether this model is useful in making medical decision of classifying a baby of being at risk of LGA or not. There is a specific threshold at which the probability of delivering a baby born LGA could influence the mother to choose a certain clinical option-treatment or no treatment. This potential relationship is used to derive the net benefit of the model over a range of different threshold probabilities. Plotting net benefit against threshold probability creates the “decision curve.” Currently, if detection of possible LGA happens after 26 weeks, no further treatment is provided to the mother. By choosing a certain threshold as the key factor, a decision curve could be designed by plotting the net benefit over different various threshold probabilities (Vickers and

Elkin, 2006). This, then, helps to see over which range the model was of value and the associated benefit.

To improve the model, other predictors could be added to this that are already available. Clinical notes show that biomarkers are usually recorded in urinalysis. Other types of models could also be used; for example, Machine Learning (ML) or ordinal regression. In terms of ordinal regression (Singh et al., 2020), the model could be further developed into predicting LGA (birthweight above the 90th centile and below the 95th centile) or severe LGA (birthweight above the 95th centile). ML techniques such as Artificial Neural Networks (ANNs) and Decision Trees (DTs) have been used in cancer detection(Kourou et al., 2015). Similarly, these could be used for detection of risk of LGA.

Future work

This thesis extensively investigated the contributions of several factors to birthweight, including parental birthweights and heights, blood pressure, maternal genetics, fetal genetics, and maternal fasting glucose. Although it has not been explored in this project, maternal BMI as well as pre-pregnancy maternal BMI (Cogswell and Yip, 1995) mostly influence birthweight. However, BMI represents the ratio of weight to the squared of height (weight/ (height)^2) and this formula could result in significant loss of information because it is using the inverse of height rather than height itself (which has been shown to be a direct, significant predictor of birthweight. In this case, we suggest that future investigations could explore the contribution to birthweight by maternal height and weight alongside other characteristics and compare this to the contribution of birthweight by maternal BMI alongside other characteristics.

The second project explored the associations of several characteristics with birthweight in two different ethnic groups, including maternal and fetal genetic scores. As mentioned previously, data about fetal genetics would not be available

prior to a baby's birth and the method known as cell-free fetal DNA (cffDNA) genotyping could be used to extract genetic data about the fetus before birth. cffDNA (Wang et al., 2021) is fetal DNA that circulates freely in the maternal blood. This test is usually offered to mothers at 16 weeks' gestation and the availability of fetal genetic information before birth would make it possible to use this information and increase the accuracy of predictions. This also improves the idea of genetic scores being used in the future and expands on the potential this could bring about in making genetics a readily accessible tool.

Finally, the risk of LGA has been explored in healthy, pregnant women and LGA has been known to be a possible adverse outcome in women with gestational diabetes (GDM). The Hyperglycemia and Adverse Pregnancy Outcomes (HAPO) Study (Coustan et al., 2010) has shown that maternal glucose levels during pregnancy vary significantly among women, and this could lead to adverse pregnancy outcomes. For example, women with GDM have higher fasting glucose levels and higher maternal glucose leads to increased fetal growth (Scholl et al., 2001) as maternal fasting glucose explains about twice the amount of variance in birthweight compared to the fetal genetic score(Hughes et al., 2018). This makes this group of women susceptible to adverse pregnancy outcomes such as LGA.

Research has also found that not all women with GDM deliver big babies (Surkan et al., 2004). Hence, being able to predict LGA in this group is quite important. The model did not reach the threshold to be used in clinical practice but since glucose is a more significant predictor in a group of women with GDM, the model might make more accurate predictions. Glycemic control for women with GDM has been found to influence birthweight (Langer et al., 1989) Therefore, being able to predict the risk of LGA in this group and using this information for better glycemic control could reduce the risk of LGA in women with GDM significantly.

Conclusions

Modeling of birthweight and risk of LGA has been extensively done in this research work. Different modeling approaches such as linear regression and logistic regression have been used, and there are new findings as well as results that are in line with previous research studies. The exciting novel findings include maternal and fetal genetics contributing to about 6% to birthweight on their own and explaining additional variance when added on top of routinely available clinical features (15.7%).

We found possible evidence that supports the suggestion that the contribution of maternal genetics to birthweight is influenced by the exposure of the maternal intrauterine environment(Nongmaithem et al., 2022). When the maternal intrauterine environment is the same, maternal genetics has similar contributions to birthweight in SAs and Europeans (Born in Bradford European and Pakistani groups had the same location and the maternal genetic score had similar associations with birthweight) and when the environment is different, maternal genetics contributes less to birthweight of SAs. Overall, most characteristics had similar associations to birthweight in the two different ethnic groups.

Finally, the risk of LGA can be assessed using routinely available clinical features in clinics and the model we developed can be easily implemented in a European cohort and a South Asian cohort after re-calibration. Previously developed and published clinical prediction models have not externally validated their models in a different ethnic group to the one it was developed in. The model we developed was found to perform better than currently published models for assessing the risk of delivering a baby born LGA and can also easily be used in two separate ethnic groups.

References

ABRAHAM, G. & INOUYE, M. 2014. Fast principal component analysis of large-scale genome-wide data. *PLoS One,* 9**,** e93766.

AHKTER, J., QURESHI, R., RAHIM, F., MOOSVI, S., REHMAN, A., JABBAR, A., ISLAM, N. & KHAN, M. A. 1996. Diabetes in pregnancy in Pakistani women: prevalence and complications in an indigenous south Asian community. *Diabet Med,* 13**,** 189-91.

AMARK, H., WESTGREN, M. & PERSSON, M. 2019. Prediction of large-for-gestational-age infants in pregnancies complicated by obesity: A population-based cohort study. *Acta Obstet Gynecol Scand,* 98**,** 769-776.

AMBLER, G., OMAR, R. Z., ROYSTON, P., KINSMAN, R., KEOGH, B. E. & TAYLOR, K. M. 2005. Generic, simple risk stratification model for heart valve surgery. *Circulation,* 112**,** 224-31.

ANAND, S. S., GUPTA, M. K., SCHULZE, K. M., DESAI, D., ABDALLA, N., WAHI, G., WADE, C., SCHEUFLER, P., MCDONALD, S. D., MORRISON, K. M., VASUDEVAN, A., DWARAKANATH, P., SRINIVASAN, K., KURPAD, A., GERSTEIN, H. C. & TEO, K. K. 2016. What accounts for ethnic differences in newborn skinfold thickness comparing South Asians and White Caucasians? Findings from the START and FAMILY Birth Cohorts. *Int J Obes (Lond),* 40**,** 239-44.

BAHRAMI TAGHANAKI, H., HASHEMIAN, M., LOTFALIZADEH, M. & NORAS, M. 2016. The relationship between Body Mass Index (BMI) and birth weight and some pregnancy outcomes. *The Iranian Journal of Obstetrics, Gynecology and Infertility,* 19**,** 1-8.

BARKER, D. J. 1995. Fetal origins of coronary heart disease. *BMJ,* 311**,** 171-4.

BARKER, D. J., HALES, C. N., FALL, C. H., OSMOND, C., PHIPPS, K. & CLARK, P. M. 1993. Type 2 (non-insulin-dependent) diabetes mellitus, hypertension and hyperlipidaemia (syndrome X): relation to reduced fetal growth. *Diabetologia,* 36**,** 62-7.

BARKER, D. J. & OSMOND, C. 1986. Infant mortality, childhood nutrition, and ischaemic heart disease in England and Wales. *Lancet,* 1**,** 1077-81.

BARRON, S. L. 1983. Birthweight and ethnicity. *Br J Obstet Gynaecol,* 90**,** 289-90.

BATTERSBY, C., STATNIKOV, Y., SANTHAKUMARAN, S., GRAY, D., MODI, N., COSTELOE, K., COLLABORATIVE, U. K. N. & MEDICINES FOR NEONATES INVESTIGATOR, G. 2018. The United Kingdom National Neonatal Research Database: A validation study. *PLoS One,* 13**,** e0201815.

BEAUMONT, R. N., WARRINGTON, N. M., CAVADINO, A., TYRRELL, J., NODZENSKI, M., HORIKOSHI, M., GELLER, F., MYHRE, R., RICHMOND, R. C., PATERNOSTER, L., BRADFIELD, J. P., KREINER-MOLLER, E., HUIKARI, V., METRUSTRY, S., LUNETTA, K. L., PAINTER, J. N., HOTTENGA, J. J., ALLARD, C., BARTON, S. J., ESPINOSA, A., MARSH, J. A., POTTER, C., ZHANG, G., ANG, W., BERRY, D. J., BOUCHARD, L., DAS, S., EARLY GROWTH GENETICS, C., HAKONARSON, H., HEIKKINEN, J., HELGELAND, O., HOCHER, B., HOFMAN, A., INSKIP, H. M., JONES, S. E., KOGEVINAS, M., LIND, P. A., MARULLO, L., MEDLAND, S. E., MURRAY, A., MURRAY, J. C., NJOLSTAD, P. R., NOHR, E. A., REICHETZEDER, C., RING, S. M., RUTH, K. S., SANTA-MARINA, L., SCHOLTENS, D. M., SEBERT, S., SENGPIEL, V., TUKE, M. A., VAUDEL, M., WEEDON, M. N., WILLEMSEN, G., WOOD, A. R., YAGHOOTKAR, H., MUGLIA, L. J., BARTELS, M., RELTON, C. L., PENNELL, C. E., CHATZI, L., ESTIVILL, X., HOLLOWAY, J. W., BOOMSMA, D. I., MONTGOMERY, G. W., MURABITO, J. M., SPECTOR, T. D., POWER, C., JARVELIN, M. R., BISGAARD, H., GRANT, S. F. A., SORENSEN, T. I. A., JADDOE, V. W., JACOBSSON, B., MELBYE, M., MCCARTHY, M. I., HATTERSLEY, A. T., HAYES, M. G., FRAYLING, T. M., HIVERT, M. F., FELIX, J. F., HYPPONEN, E., LOWE, W. L., JR., EVANS, D. M., LAWLOR, D. A., FEENSTRA, B. & FREATHY, R. M. 2018. Genome-wide association study of offspring birth weight in 86 577 women identifies five novel loci and highlights maternal genetic effects that are independent of fetal genetics. *Hum Mol Genet,* 27**,** 742-756.

BELBASIS, L., SAVVIDOU, M. D., KANU, C., EVANGELOU, E. & TZOULAKI, I. 2016. Birth weight in relation to health and disease in later life: an umbrella review of systematic reviews and meta-analyses. *BMC Med,* 14**,** 147.

BIAU, D. J., KERNEIS, S. & PORCHER, R. 2008. Statistics in brief: the importance of sample size in the planning and interpretation of medical research. *Clin Orthop Relat Res,* 466**,** 2282-8.

BIKS, G. A., BLENCOWE, H., HARDY, V. P., GEREMEW, B. M., ANGAW, D. A., WAGNEW, A., ABEBE, S. M., GUADU, T., MARTINS, J. S. D., FISKER, A. B., IMAM, M. A., NETTEY, O. E. A., KASASA, S., DI STEFANO, L., AKUZE, J., KWESIGA, D., LAWN, J. E. & EVERY NEWBORN, I. S. C. G. 2021. Birthweight data completeness and quality in population-based surveys: EN-INDEPTH study. *Popul Health Metr,* 19**,** 17.

BOYD, M. E., USHER, R. H. & MCLEAN, F. H. 1983. Fetal macrosomia: prediction, risks, proposed management. *Obstet Gynecol,* 61**,** 715-22.

BRESCHI, M. C., SEGHIERI, G., BARTOLOMEI, G., GIRONI, A., BALDI, S. & FERRANNINI, E. 1993. Relation of birthweight to maternal plasma glucose and insulin concentrations during normal pregnancy. *Diabetologia,* 36**,** 1315-21.

BROWN, A. E. & WALKER, M. 2016. Genetics of Insulin Resistance and the Metabolic Syndrome. *Curr Cardiol Rep,* 18**,** 75.

CAMERON, N. & HAWLEY, N. L. 2010. Should the UK use WHO growth charts? *Paediatrics and Child Health,* 20**,** 151-156.

CATALANO, P. M., DRAGO, N. M. & AMINI, S. B. 1995. Factors affecting fetal growth and body composition. *Am J Obstet Gynecol,* 172**,** 1459-63.

CAUZZO, C., CHIAVAROLI, V., DI VALERIO, S. & CHIARELLI, F. 2023. Birth size, growth trajectory and later cardio-metabolic risk. *Front Endocrinol (Lausanne),* 14**,** 1187261.

CETIN, I., MANDO, C. & CALABRESE, S. 2013. Maternal predictors of intrauterine growth restriction. *Curr Opin Clin Nutr Metab Care,* 16**,** 310-9.

CHOWDHURY, M. Z. I. & TURIN, T. C. 2020. Variable selection strategies and its importance in clinical prediction modelling. *Fam Med Community Health,* 8**,** e000262.

COGSWELL, M. E. & YIP, R. 1995. The influence of fetal and maternal factors on the distribution of birthweight. *Semin Perinatol,* 19**,** 222-40.

COLE, T. J., FREEMAN, J. V. & PREECE, M. A. 1998. British 1990 growth reference centiles for weight, height, body mass index and head circumference fitted by maximum penalized likelihood. *Stat Med,* 17**,** 407-29.

COLLINS, G. S., REITSMA, J. B., ALTMAN, D. G. & MOONS, K. G. 2015. Transparent reporting of a multivariable prediction model for individual prognosis or diagnosis (TRIPOD): the TRIPOD statement. *BMJ,* 350**,** g7594.

COLLISTER, J. A., LIU, X. & CLIFTON, L. 2022. Calculating Polygenic Risk Scores (PRS) in UK Biobank: A Practical Guide for Epidemiologists. *Front Genet,* 13**,** 818574.

COORAY, S. D., BOYLE, J. A., SOLDATOS, G., ALLOTEY, J., WANG, H., FERNANDEZ-FELIX, B. M., ZAMORA, J., THANGARATINAM, S. & TEEDE, H. J. 2022. Development, validation and clinical utility of a risk prediction model for adverse pregnancy outcomes in women with gestational diabetes: The PeRSonal GDM model. *EClinicalMedicine,* 52**,** 101637.

COUSTAN, D. R., LOWE, L. P., METZGER, B. E., DYER, A. R., INTERNATIONAL ASSOCIATION OF, D. & PREGNANCY STUDY, G. 2010. The Hyperglycemia and Adverse Pregnancy Outcome (HAPO) study: paving the way for new diagnostic criteria for gestational diabetes mellitus. *Am J Obstet Gynecol,* 202**,** 654 e1-6.

CRAMER, J. C. 1995. Racial and ethnic differences in birthweight: the role of income and financial assistance. *Demography,* 32**,** 231-47.

DE GRAAFF, E., SADLER, L., LAKHDHIR, H., SIMON-KUMAR, R., PEIRIS-JOHN, R., BURGESS, W., OKESENE-GAFA, K., CRONIN, R., MCCOWAN, L. & ANDERSON, N. 2023. Grouping women

of South Asian ethnicity for pregnancy research in New Zealand. *Aust N Z J Obstet Gynaecol,* 63**,** 499-508.

DHIMAN, P., MA, J., QI, C., BULLOCK, G., SERGEANT, J. C., RILEY, R. D. & COLLINS, G. S. 2023. Sample size requirements are not being considered in studies developing prediction models for binary outcomes: a systematic review. *BMC Med Res Methodol,* 23**,** 188.

DONG, Y., LUO, Z. C., NUYT, A. M., AUDIBERT, F., WEI, S. Q., ABENHAIM, H. A., BUJOLD, E., JULIEN, P., HUANG, H., LEVY, E., FRASER, W. D. & GROUP, D. C. S. 2018. Large-for-Gestational-Age May Be Associated With Lower Fetal Insulin Sensitivity and beta-Cell Function Linked to Leptin. *J Clin Endocrinol Metab,* 103**,** 3837-3844.

DUTTON, P. J., WARRANDER, L. K., ROBERTS, S. A., BERNATAVICIUS, G., BYRD, L. M., GAZE, D., KROLL, J., JONES, R. L., SIBLEY, C. P., FROEN, J. F. & HEAZELL, A. E. 2012. Predictors of poor perinatal outcome following maternal perception of reduced fetal movements--a prospective cohort study. *PLoS One,* 7**,** e39784.

DWI PUTRA, S. E., REICHETZEDER, C., HASAN, A. A., SLOWINSKI, T., CHU, C., KRAMER, B. K., KLEUSER, B. & HOCHER, B. 2020. Being Born Large for Gestational Age is Associated with Increased Global Placental DNA Methylation. *Sci Rep,* 10**,** 927.

EVERETT, T. R. & CHITTY, L. S. 2015. Cell-free fetal DNA: the new tool in fetal medicine. *Ultrasound Obstet Gynecol,* 45**,** 499-507.

FADISTA, J., MANNING, A. K., FLOREZ, J. C. & GROOP, L. 2016. The (in)famous GWAS P-value threshold revisited and updated for low-frequency variants. *Eur J Hum Genet,* 24**,** 1202-5.

FLANAGAN, S. E., PATCH, A. M., MACKAY, D. J., EDGHILL, E. L., GLOYN, A. L., ROBINSON, D., SHIELD, J. P., TEMPLE, K., ELLARD, S. & HATTERSLEY, A. T. 2007. Mutations in ATP-sensitive K+ channel genes cause transient neonatal diabetes and permanent diabetes in childhood or adulthood. *Diabetes,* 56**,** 1930-7.

FRAGOSO-BARGAS, N., OPSAHL, J. O., KIRYUSHCHENKO, N., BOTTCHER, Y., LEE-ODEGARD, S., QVIGSTAD, E., RICHARDSEN, K. R., WAAGE, C. W., SLETNER, L., JENUM, A. K., PRASAD, R. B., GROOP, L. C., MOEN, G. H., BIRKELAND, K. I. & SOMMER, C. 2021. Cohort profile: Epigenetics in Pregnancy (EPIPREG) - population-based sample of European and South Asian pregnant women with epigenome-wide DNA methylation (850k) in peripheral blood leukocytes. *PLoS One,* 16**,** e0256158.

FRICK, A. P., SYNGELAKI, A., ZHENG, M., POON, L. C. & NICOLAIDES, K. H. 2016. Prediction of large-for-gestational-age neonates: screening by maternal factors and biomarkers in the three trimesters of pregnancy. *Ultrasound Obstet Gynecol,* 47**,** 332-9.

GARIN, I., EDGHILL, E. L., AKERMAN, I., RUBIO-CABEZAS, O., RICA, I., LOCKE, J. M., MAESTRO, M. A., ALSHAIKH, A., BUNDAK, R., DEL CASTILLO, G., DEEB, A., DEISS, D., FERNANDEZ, J. M., GODBOLE, K., HUSSAIN, K., O'CONNELL, M., KLUPA, T., KOLOUSKOVA, S., MOHSIN, F., PERLMAN, K., SUMNIK, Z., RIAL, J. M., UGARTE, E., VASANTHI, T., NEONATAL DIABETES INTERNATIONAL, G., JOHNSTONE, K., FLANAGAN, S. E., MARTINEZ, R., CASTANO, C., PATCH, A. M., FERNANDEZ-REBOLLO, E., RAILE, K., MORGAN, N., HARRIES, L. W., CASTANO, L., ELLARD, S., FERRER, J., PEREZ DE NANCLARES, G. & HATTERSLEY, A. T. 2010. Recessive mutations in the INS gene result in neonatal diabetes through reduced insulin biosynthesis. *Proc Natl Acad Sci U S A,* 107**,** 3105-10.

GREENHALGH, T., CLINCH, M., AFSAR, N., CHOUDHURY, Y., SUDRA, R., CAMPBELL-RICHARDS, D., CLAYDON, A., HITMAN, G. A., HANSON, P. & FINER, S. 2015. Socio-cultural influences on the behaviour of South Asian women with diabetes in pregnancy: qualitative study using a multi-level theoretical approach. *BMC Med,* 13**,** 120.

GRIFFITHS, L. J., DEZATEUX, C. & COLE, T. J. 2007. Differential parental weight and height contributions to offspring birthweight and weight gain in infancy. *Int J Epidemiol,* 36**,** 104-7.

GUL, R., IQBAL, S., ANWAR, Z., AHDI, S. G., ALI, S. H. & PIRZADA, S. 2020. Pre-pregnancy maternal BMI as predictor of neonatal birth weight. *PLoS One,* 15**,** e0240748.

GURURAJAN, P., GURUMURTHY, P., NAYAR, P., RAO, G. S., BABU, R. S., SARASABHARATI, A. & CHERIAN, K. M. 2012. Pregnancy associated plasma protein-A (PAPP-A) as an early marker for the diagnosis of acute coronary syndrome. *Indian Heart J,* 64**,** 141-5.

HAJIAN-TILAKI, K. 2013. Receiver Operating Characteristic (ROC) Curve Analysis for Medical Diagnostic Test Evaluation. *Caspian J Intern Med,* 4**,** 627-35.

HARDER, T., RODEKAMP, E., SCHELLONG, K., DUDENHAUSEN, J. W. & PLAGEMANN, A. 2007. Birth weight and subsequent risk of type 2 diabetes: a meta-analysis. *Am J Epidemiol,* 165**,** 849-57.

HARRELL, F. E., JR., LEE, K. L., CALIFF, R. M., PRYOR, D. B. & ROSATI, R. A. 1984. Regression modelling strategies for improved prognostic prediction. *Stat Med,* 3**,** 143-52.

HATTERSLEY, A. T., BEARDS, F., BALLANTYNE, E., APPLETON, M., HARVEY, R. & ELLARD, S. 1998. Mutations in the glucokinase gene of the fetus result in reduced birth weight. *Nat Genet,* 19**,** 268-70.

HATTERSLEY, A. T. & TOOKE, J. E. 1999. The fetal insulin hypothesis: an alternative explanation of the association of low birthweight with diabetes and vascular disease. *Lancet,* 353**,** 1789-92.

HAULDER, M., HUGHES, A. E., BEAUMONT, R. N., KNIGHT, B. A., HATTERSLEY, A. T., SHIELDS, B. M. & FREATHY, R. M. 2022. Assessing whether genetic scores explain extra variation in birthweight, when added to clinical and anthropometric measures. *BMC Pediatr,* 22**,** 504.

HINKLE, S. N., ALBERT, P. S., MENDOLA, P., SJAARDA, L. A., YEUNG, E., BOGHOSSIAN, N. S. & LAUGHON, S. K. 2014. The association between parity and birthweight in a longitudinal consecutive pregnancy cohort. *Paediatr Perinat Epidemiol,* 28**,** 106-15.

HOFFNER, L. & SURTI, U. 2012. The genetics of gestational trophoblastic disease: a rare complication of pregnancy. *Cancer Genet,* 205**,** 63-77.

HONG, Y. H. & LEE, J. E. 2021. Large for Gestational Age and Obesity-Related Comorbidities. *J Obes Metab Syndr,* 30**,** 124-131.

HUGHES, A. E., DE FRANCO, E., FREATHY, R. M., FETAL, I., GROWTH, C., FLANAGAN, S. E. & HATTERSLEY, A. T. 2023. Monogenic disease analysis establishes that fetal insulin accounts for half of human fetal growth. *J Clin Invest,* 133.

HUGHES, A. E., HATTERSLEY, A. T., FLANAGAN, S. E. & FREATHY, R. M. 2021. Two decades since the fetal insulin hypothesis: what have we learned from genetics? *Diabetologia,* 64**,** 717-726.

HUGHES, A. E., NODZENSKI, M., BEAUMONT, R. N., TALBOT, O., SHIELDS, B. M., SCHOLTENS, D. M., KNIGHT, B. A., LOWE, W. L., JR., HATTERSLEY, A. T. & FREATHY, R. M. 2018. Fetal Genotype and Maternal Glucose Have Independent and Additive Effects on Birth Weight. *Diabetes,* 67**,** 1024-1029.

JOHNSTON, L. B., CLARK, A. J. & SAVAGE, M. O. 2002. Genetic factors contributing to birth weight. *Arch Dis Child Fetal Neonatal Ed,* 86**,** F2-3.

JULIUSDOTTIR, T., STEINTHORSDOTTIR, V., STEFANSDOTTIR, L., SVEINBJORNSSON, G., IVARSDOTTIR, E. V., THOROLFSDOTTIR, R. B., SIGURDSSON, J. K., TRAGANTE, V., HJORLEIFSSON, K. E., HELGADOTTIR, A., FRIGGE, M. L., THORGEIRSSON, G., BENEDIKTSSON, R., SIGURDSSON, E. L., ARNAR, D. O., STEINGRIMSDOTTIR, T., JONSDOTTIR, I., HOLM, H., GUDBJARTSSON, D. F., THORLEIFSSON, G., THORSTEINSDOTTIR, U. & STEFANSSON, K. 2021. Distinction between the effects of parental and fetal genomes on fetal growth. *Nat Genet,* 53**,** 1135-1142.

KALAFAT, E., MORALES-ROSELLO, J., THILAGANATHAN, B., DHOTHER, J. & KHALIL, A. 2019. Risk of neonatal care unit admission in small for gestational age fetuses at term: a prediction model and internal validation. *J Matern Fetal Neonatal Med,* 32**,** 2361-2368.

KATAOKA, M. C., CARVALHEIRA, A. P. P., FERRARI, A. P., MALTA, M. B., DE BARROS LEITE CARVALHAES, M. A. & DE LIMA PARADA, C. M. G. 2018. Smoking during pregnancy and harm reduction in birth weight: a cross-sectional study. *BMC Pregnancy Childbirth,* 18**,** 67.

KC, K., SHAKYA, S. & ZHANG, H. 2015. Gestational diabetes mellitus and macrosomia: a literature review. *Ann Nutr Metab,* 66 Suppl 2**,** 14-20.

KELLY, Y., PANICO, L., BARTLEY, M., MARMOT, M., NAZROO, J. & SACKER, A. 2009. Why does birthweight vary among ethnic groups in the UK? Findings from the Millennium Cohort Study. *J Public Health (Oxf),* 31**,** 131-7.

KLEIN, R. J., ZEISS, C., CHEW, E. Y., TSAI, J. Y., SACKLER, R. S., HAYNES, C., HENNING, A. K., SANGIOVANNI, J. P., MANE, S. M., MAYNE, S. T., BRACKEN, M. B., FERRIS, F. L., OTT, J., BARNSTABLE, C. & HOH, J. 2005. Complement factor H polymorphism in age-related macular degeneration. *Science,* 308**,** 385-9.

KNIGHT, B., SHIELDS, B. M. & HATTERSLEY, A. T. 2006. The Exeter Family Study of Childhood Health (EFSOCH): study protocol and methodology. *Paediatr Perinat Epidemiol,* 20**,** 172-9.

KNIGHT, B., SHIELDS, B. M., TURNER, M., POWELL, R. J., YAJNIK, C. S. & HATTERSLEY, A. T. 2005. Evidence of genetic regulation of fetal longitudinal growth. *Early Hum Dev,* 81**,** 823-31.

KNOP, M. R., GENG, T. T., GORNY, A. W., DING, R., LI, C., LEY, S. H. & HUANG, T. 2018. Birth weight and risk of type 2 diabetes mellitus, cardiovascular disease, and hypertension in adults: a meta-analysis of 7 646 267 participants from 135 studies. *Journal of the American Heart Association,* 7**,** e008870.

KOUROU, K., EXARCHOS, T. P., EXARCHOS, K. P., KARAMOUZIS, M. V. & FOTIADIS, D. I. 2015. Machine learning applications in cancer prognosis and prediction. *Comput Struct Biotechnol J,* 13**,** 8-17.

KROOK, A., BRUETON, L. & O'RAHILLY, S. 1993. Homozygous nonsense mutation in the insulin receptor gene in infant with leprechaunism. *Lancet,* 342**,** 277-8.

KUHLE, S., MAGUIRE, B., ZHANG, H., HAMILTON, D., ALLEN, A. C., JOSEPH, K. S. & ALLEN, V. M. 2018. Comparison of logistic regression with machine learning methods for the prediction of fetal growth abnormalities: a retrospective cohort study. *BMC Pregnancy Childbirth,* 18**,** 333.

KURPAD, A. V., VARADHARAJAN, K. S. & AEBERLI, I. 2011. The thin-fat phenotype and global metabolic disease risk. *Curr Opin Clin Nutr Metab Care,* 14**,** 542-7.

LABARERE, J., RENAUD, B. & FINE, M. J. 2014. How to derive and validate clinical prediction models for use in intensive care medicine. *Intensive Care Med,* 40**,** 513-27.

LANGE, S., PROBST, C., REHM, J. & POPOVA, S. 2018. National, regional, and global prevalence of smoking during pregnancy in the general population: a systematic review and meta-analysis. *Lancet Glob Health,* 6**,** e769-e776.

LANGER, O., LEVY, J., BRUSTMAN, L., ANYAEGBUNAM, A., MERKATZ, R. & DIVON, M. 1989. Glycemic control in gestational diabetes mellitus--how tight is tight enough: small for gestational age versus large for gestational age? *Am J Obstet Gynecol,* 161**,** 646-53.

LEE, Y., SHIN, J. H., PARK, H. C., KIM, S. G. & CHOI, S. I. 2014. A prediction model for renal artery stenosis using carotid ultrasonography measurements in patients undergoing coronary angiography. *BMC Nephrol,* 15**,** 60.

LITTLE, R. E. 1987. Mother's and father's birthweight as predictors of infant birthweight. *Paediatr Perinat Epidemiol,* 1**,** 19-31.

MAGNUS, P., GJESSING, H. K., SKRONDAL, A. & SKJAERVEN, R. 2001. Paternal contribution to birth weight. *J Epidemiol Community Health,* 55**,** 873-7.

MAHAJAN, A., WESSEL, J., WILLEMS, S. M., ZHAO, W., ROBERTSON, N. R., CHU, A. Y., GAN, W., KITAJIMA, H., TALIUN, D., RAYNER, N. W., GUO, X., LU, Y., LI, M., JENSEN, R. A., HU, Y., HUO, S., LOHMAN, K. K., ZHANG, W., COOK, J. P., PRINS, B. P., FLANNICK, J., GRARUP, N., TRUBETSKOY, V. V., KRAVIC, J., KIM, Y. J., RYBIN, D. V., YAGHOOTKAR, H., MULLER-

NURASYID, M., MEIDTNER, K., LI-GAO, R., VARGA, T. V., MARTEN, J., LI, J., SMITH, A. V., AN, P., LIGTHART, S., GUSTAFSSON, S., MALERBA, G., DEMIRKAN, A., TAJES, J. F., STEINTHORSDOTTIR, V., WUTTKE, M., LECOEUR, C., PREUSS, M., BIELAK, L. F., GRAFF, M., HIGHLAND, H. M., JUSTICE, A. E., LIU, D. J., MAROULI, E., PELOSO, G. M., WARREN, H. R., EXOME, B. P. C., CONSORTIUM, M., CONSORTIUM, G., AFAQ, S., AFZAL, S., AHLQVIST, E., ALMGREN, P., AMIN, N., BANG, L. B., BERTONI, A. G., BOMBIERI, C., BORK-JENSEN, J., BRANDSLUND, I., BRODY, J. A., BURTT, N. P., CANOUIL, M., CHEN, Y. I., CHO, Y. S., CHRISTENSEN, C., EASTWOOD, S. V., ECKARDT, K. U., FISCHER, K., GAMBARO, G., GIEDRAITIS, V., GROVE, M. L., DE HAAN, H. G., HACKINGER, S., HAI, Y., HAN, S., TYBJAERG-HANSEN, A., HIVERT, M. F., ISOMAA, B., JAGER, S., JORGENSEN, M. E., JORGENSEN, T., KARAJAMAKI, A., KIM, B. J., KIM, S. S., KOISTINEN, H. A., KOVACS, P., KRIEBEL, J., KRONENBERG, F., LALL, K., LANGE, L. A., LEE, J. J., LEHNE, B., LI, H., LIN, K. H., et al. 2018. Refining the accuracy of validated target identification through coding variant fine-mapping in type 2 diabetes. *Nat Genet,* 50**,** 559-571.

MAKGOBA, M., SAVVIDOU, M. D. & STEER, P. J. 2012. The effect of maternal characteristics and gestational diabetes on birthweight. *BJOG,* 119**,** 1091-7.

MANICHAIKUL, A., MYCHALECKYJ, J. C., RICH, S. S., DALY, K., SALE, M. & CHEN, W. M. 2010. Robust relationship inference in genome-wide association studies. *Bioinformatics,* 26**,** 2867-73.

MARTIN-GRONERT, M. S. & OZANNE, S. E. 2010. Mechanisms linking suboptimal early nutrition and increased risk of type 2 diabetes and obesity. *J Nutr,* 140**,** 662-6.

MARTIN, S., CULE, M., BASTY, N., TYRRELL, J., BEAUMONT, R. N., WOOD, A. R., FRAYLING, T. M., SOROKIN, E., WHITCHER, B., LIU, Y., BELL, J. D., THOMAS, E. L. & YAGHOOTKAR, H. 2021. Genetic Evidence for Different Adiposity Phenotypes and Their Opposing Influences on Ectopic Fat and Risk of Cardiometabolic Disease. *Diabetes,* 70**,** 1843-1856.

MASIAKWALA, E., NYATI, L. H. & NORRIS, S. A. 2023. The association of intrauterine and postnatal growth patterns and nutritional status with toddler body composition. *BMC Pediatr,* 23**,** 342.

MEERTENS, L., SMITS, L., VAN KUIJK, S., AARDENBURG, R., VAN DOOREN, I., LANGENVELD, J., ZWAAN, I. M., SPAANDERMAN, M. & SCHEEPERS, H. 2019. External validation and clinical usefulness of first-trimester prediction models for small- and large-for-gestational-age infants: a prospective cohort study. *BJOG,* 126**,** 472-484.

MENOTTI, A., PUDDU, P. E. & LANTI, M. 2000. Comparison of the Framingham risk function-based coronary chart with risk function from an Italian population study. *Eur Heart J,* 21**,** 365-70.

METGUD, C. S., NAIK, V. A. & MALLAPUR, M. D. 2012. Factors affecting birth weight of a newborn--a community based study in rural Karnataka, India. *PLoS One,* 7**,** e40040.

MI, D., FANG, H., ZHAO, Y. & ZHONG, L. 2017. Birth weight and type 2 diabetes: A meta-analysis. *Exp Ther Med,* 14**,** 5313-5320.

MILNER, J. & AREZINA, J. 2018. The accuracy of ultrasound estimation of fetal weight in comparison to birth weight: A systematic review. *Ultrasound,* 26**,** 32-41.

MIRANDA, M. L., EDWARDS, S. E. & MYERS, E. R. 2011. Adverse birth outcomes among nulliparous vs. multiparous women. *Public Health Rep,* 126**,** 797-805.

MONARI, F., MENICHINI, D., SPANO' BASCIO, L., GRANDI, G., BANCHELLI, F., NERI, I., D'AMICO, R. & FACCHINETTI, F. 2021. A first trimester prediction model for large for gestational age infants: a preliminary study. *BMC Pregnancy Childbirth,* 21**,** 654.

MORLEY, R. 2006. Fetal origins of adult disease. *Semin Fetal Neonatal Med,* 11**,** 73-8.

NAHUM, G. G., STANISLAW, H. & HUFFAKER, B. J. 1999. Accurate prediction of term birth weight from prospectively measurable maternal characteristics. *J Reprod Med,* 44**,** 705-12.

NIGHTINGALE, C. M., RUDNICKA, A. R., OWEN, C. G., COOK, D. G. & WHINCUP, P. H. 2011. Patterns of body size and adiposity among UK children of South Asian, black African-Caribbean and

white European origin: Child Heart And health Study in England (CHASE Study). *Int J Epidemiol,* 40**,** 33-44.

NONGMAITHEM, S. S., BEAUMONT, R. N., DEDANIYA, A., WOOD, A. R., OGUNKOLADE, B. W., HASSAN, Z., KRISHNAVENI, G. V., KUMARAN, K., POTDAR, R. D., SAHARIAH, S. A., KRISHNA, M., DI GRAVIO, C., MALI, I. D., SANKARESWARAN, A., HUSSAIN, A., BHOWMIK, B. W., KHAN, A. K. A., KNIGHT, B. A., FRAYLING, T. M., FINER, S., FALL, C. H. D., YAJNIK, C. S., FREATHY, R. M., HITMAN, G. A. & CHANDAK, G. R. 2022. Babies of South Asian and European Ancestry Show Similar Associations With Genetic Risk Score for Birth Weight Despite the Smaller Size of South Asian Newborns. *Diabetes,* 71**,** 821-836.

NORDMAN, H., JAASKELAINEN, J. & VOUTILAINEN, R. 2020. Birth Size as a Determinant of Cardiometabolic Risk Factors in Children. *Horm Res Paediatr,* 93**,** 144-153.

OKONOFUA, F. & AKABA, G. 2021. Royal College of Obstetricians and Gynecologists (RCOG) champions' network: A new agenda for women's health reform in sub-Saharan Africa. *Afr J Reprod Health,* 25**,** 10-13.

ONG, K. K., AHMED, M. L., EMMETT, P. M., PREECE, M. A. & DUNGER, D. B. 2000. Association between postnatal catch-up growth and obesity in childhood: prospective cohort study. *BMJ,* 320**,** 967-71.

OPARA, E. I. & ZAIDI, J. 2007. The interpretation and clinical application of the word 'parity': a survey. *BJOG,* 114**,** 1295-7.

OSUCHUKWU, O. O. & REED, D. J. 2023. Small for Gestational Age. *StatPearls.* Treasure Island (FL).

PAPASTEFANOU, I., SOUKA, A. P., PILALIS, A., ELEFTHERIADES, M., MICHALITSI, V. & KASSANOS, D. 2012. First trimester prediction of small- and large-for-gestation neonates by an integrated model incorporating ultrasound parameters, biochemical indices and maternal characteristics. *Acta Obstet Gynecol Scand,* 91**,** 104-111.

PETTITT, D. J. & JOVANOVIC, L. 2001. Birth weight as a predictor of type 2 diabetes mellitus: the U-shaped curve. *Curr Diab Rep,* 1**,** 78-81.

PLASENCIA, W., AKOLEKAR, R., DAGKLIS, T., VEDUTA, A. & NICOLAIDES, K. H. 2011. Placental volume at 11-13 weeks' gestation in the prediction of birth weight percentile. *Fetal Diagn Ther,* 30**,** 23-8.

PLASENCIA, W., GONZALEZ DAVILA, E., TETILLA, V., PADRON PEREZ, E., GARCIA HERNANDEZ, J. A. & GONZALEZ GONZALEZ, N. L. 2012. First-trimester screening for large-for-gestational-age infants. *Ultrasound Obstet Gynecol,* 39**,** 389-95.

POON, L. C., KARAGIANNIS, G., STRATIEVA, V., SYNGELAKI, A. & NICOLAIDES, K. H. 2011. First-trimester prediction of macrosomia. *Fetal Diagn Ther,* 29**,** 139-47.

POYRAZOGLU, S., DARENDELILER, F., AHMED, S. F., HUGHES, I., BRYCE, J., JIANG, J., RODIE, M., HIORT, O., HANNEMA, S. E., BERTELLONI, S., LISA, L., GURAN, T., COOLS, M., DESLOOVERE, A., CLAAHSEN-VAN DER GRINTEN, H. L., NORDENSTROM, A., HOLTERHUS, P. M., KOHLER, B., NIEDZIELA, M. & KRONE, N. 2017. Birth Weight in Different Etiologies of Disorders of Sex Development. *J Clin Endocrinol Metab,* 102**,** 1044-1050.

RAMSPEK, C. L., JAGER, K. J., DEKKER, F. W., ZOCCALI, C. & VAN DIEPEN, M. 2021. External validation of prognostic models: what, why, how, when and where? *Clin Kidney J,* 14**,** 49-58.

RANI, P. R. & BEGUM, J. 2016. Screening and Diagnosis of Gestational Diabetes Mellitus, Where Do We Stand. *J Clin Diagn Res,* 10**,** QE01-4.

RENOM ESPINEIRA, A., FERNANDES-ROSA, F. L., BUENO, A. C., DE SOUZA, R. M., MOREIRA, A. C., DE CASTRO, M., BARBIERI, M. A., BETTIOL, H. & ANTONINI, S. R. 2011. Postnatal growth and cardiometabolic profile in young adults born large for gestational age. *Clin Endocrinol (Oxf),* 75**,** 335-41.

RILEY, R. D., ENSOR, J., SNELL, K. I. E., HARRELL, F. E., JR., MARTIN, G. P., REITSMA, J. B., MOONS, K. G. M., COLLINS, G. & VAN SMEDEN, M. 2020. Calculating the sample size required for developing a clinical prediction model. *BMJ,* 368**,** m441.

ROTHMAN, K. J. 2008. BMI-related errors in the measurement of obesity. *Int J Obes (Lond),* 32 Suppl 3**,** S56-9.

RUBIO-CABEZAS, O. & ELLARD, S. 2013. Diabetes mellitus in neonates and infants: genetic heterogeneity, clinical approach to diagnosis, and therapeutic options. *Horm Res Paediatr,* 80**,** 137-46.

SACKS, D. A., LIU, A. I., WOLDE-TSADIK, G., AMINI, S. B., HUSTON-PRESLEY, L. & CATALANO, P. M. 2006. What proportion of birth weight is attributable to maternal glucose among infants of diabetic women? *Am J Obstet Gynecol,* 194**,** 501-7.

SAENGER, P., CZERNICHOW, P., HUGHES, I. & REITER, E. O. 2007. Small for gestational age: short stature and beyond. *Endocr Rev,* 28**,** 219-51.

SCHOLL, T. O., SOWERS, M., CHEN, X. & LENDERS, C. 2001. Maternal glucose concentration influences fetal growth, gestation, and pregnancy complications. *Am J Epidemiol,* 154**,** 514-20.

SCIFRES, C. M. 2021. Short- and Long-Term Outcomes Associated with Large for Gestational Age Birth Weight. *Obstet Gynecol Clin North Am,* 48**,** 325-337.

SEIDMAN, D. S., EVER-HADANI, P., STEVENSON, D. K., SLATER, P. E., HARLAP, S. & GALE, R. 1988. Birth order and birth weight reexamined. *Obstetrics and gynecology,* 72**,** 158-162.

SHAH, P. S. & BIRTHS, K. S. G. O. D. O. L. P. 2010. Parity and low birth weight and preterm birth: a systematic review and meta-analyses. *Acta obstetricia et gynecologica Scandinavica,* 89**,** 862-875.

SHIPE, M. E., DEPPEN, S. A., FARJAH, F. & GROGAN, E. L. 2019. Developing prediction models for clinical use using logistic regression: an overview. *J Thorac Dis,* 11**,** S574-S584.

SINGH, K. & XIE, M. 2008. Bootstrap: a statistical method. *Unpublished manuscript, Rutgers University, USA. Retrieved from http://www. stat. rutgers. edu/home/mxie/RCPapers/bootstrap. pdf***,** 1-14.

SINGH, V., DWIVEDI, S. N. & DEO, S. V. S. 2020. Ordinal logistic regression model describing factors associated with extent of nodal involvement in oral cancer patients and its prospective validation. *BMC Med Res Methodol,* 20**,** 95.

SMEENK, J. M., BRAAT, D. D., STOLWIJK, A. M. & KREMER, J. A. 2007. Pregnancy is predictable: a large-scale prospective external validation of the prediction of spontaneous pregnancy in subfertile couples. *Hum Reprod,* 22**,** 2344-5; author reply 2345-6.

SPENCER, N. & LOGAN, S. 2002. Social influences on birth weight. *J Epidemiol Community Health,* 56**,** 326-7.

SRIDHAR, S. B., FERRARA, A., EHRLICH, S. F., BROWN, S. D. & HEDDERSON, M. M. 2013. Risk of large-for-gestational-age newborns in women with gestational diabetes by race and ethnicity and body mass index categories. *Obstet Gynecol,* 121**,** 1255-1262.

STEYERBERG, E. W. 2010. *Clinical prediction models : a practical approach to development, validation and updating,* New York, Springer.

STEYERBERG, E. W. 2019. Overfitting and Optimism in Prediction Models. *Clinical Prediction Models: A Practical Approach to Development, Validation, and Updating.* Cham: Springer International Publishing.

STEYERBERG, E. W., HARRELL, F. E., JR., BORSBOOM, G. J., EIJKEMANS, M. J., VERGOUWE, Y. & HABBEMA, J. D. 2001. Internal validation of predictive models: efficiency of some procedures for logistic regression analysis. *J Clin Epidemiol,* 54**,** 774-81.

SUN, H., DEPRAETERE, K., MEESSEMAN, L., CABANILLAS SILVA, P., SZYMANOWSKY, R., FLIEGENSCHMIDT, J., HULDE, N., VON DOSSOW, V., VANBIERVLIET, M., DE BAERDEMAEKER, J., ROCCARO-WALDMEYER, D. M., STIEG, J., DOMINGUEZ HIDALGO, M. &

DAHLWEID, F. M. 2022. Machine Learning-Based Prediction Models for Different Clinical Risks in Different Hospitals: Evaluation of Live Performance. *J Med Internet Res,* 24**,** e34295.

SURKAN, P. J., HSIEH, C. C., JOHANSSON, A. L., DICKMAN, P. W. & CNATTINGIUS, S. 2004. Reasons for increasing trends in large for gestational age births. *Obstet Gynecol,* 104**,** 720-6.

SYKES, A. O. 1993. An introduction to regression analysis.

TAKAGI, K., IWAMA, N., METOKI, H., UCHIKURA, Y., MATSUBARA, Y., MATSUBARA, K., NISHIGORI, H., SAITO, M., FUJIWARA, I., SAKURAI, K., KURIYAMA, S., ARIMA, T., NAKAI, K., YAEGASHI, N., SUGIYAMA, T., JAPAN, E. & CHILDREN'S STUDY, G. 2019. Paternal height has an impact on birth weight of their offspring in a Japanese population: the Japan Environment and Children's Study. *J Dev Orig Health Dis,* 10**,** 542-554.

TEKOLA-AYELE, F., WORKALEMAHU, T. & AMARE, A. T. 2018. High burden of birthweight-lowering genetic variants in Africans and Asians. *BMC Med,* 16**,** 70.

TENNANT, P., DOXFORD-HOOK, E., FLYNN, L., KERSHAW, K., GODDARD, J. & STACEY, T. 2022. Fasting plasma glucose, diagnosis of gestational diabetes and the risk of large for gestational age: a regression discontinuity analysis of routine data. *BJOG,* 129**,** 82-89.

TERAUCHI, Y., KUBOTA, N., TAMEMOTO, H., SAKURA, H., NAGAI, R., AKANUMA, Y., KIMURA, S. & KADOWAKI, T. 2000. Insulin effect during embryogenesis determines fetal growth: a possible molecular link between birth weight and susceptibility to type 2 diabetes. *Diabetes,* 49**,** 82-6.

THOMPSON, W. D., BEAUMONT, R. N., KUANG, A., WARRINGTON, N. M., JI, Y., TYRRELL, J., WOOD, A. R., SCHOLTENS, D. M., KNIGHT, B. A., EVANS, D. M., LOWE, W. L., JR., SANTORELLI, G., AZAD, R., MASON, D., HATTERSLEY, A. T., FRAYLING, T. M., YAGHOOTKAR, H., BORGES, M. C., LAWLOR, D. A. & FREATHY, R. M. 2021. Higher maternal adiposity reduces offspring birthweight if associated with a metabolically favourable profile. *Diabetologia,* 64**,** 2790-2802.

THOMPSON, W. D., BEAUMONT, R. N., KUANG, A., WARRINGTON, N. M., JI, Y., TYRRELL, J., WOOD, A. R., SCHOLTENS, D. M., KNIGHT, B. A., EVANS, D. M., LOWE, W. L., JR., SANTORELLI, G., AZAD, R., MASON, D., HATTERSLEY, A. T., FRAYLING, T. M., YAGHOOTKAR, H., BORGES, M. C., LAWLOR, D. A. & FREATHY, R. M. 2022. Fetal alleles predisposing to metabolically favorable adiposity are associated with higher birth weight. *Hum Mol Genet,* 31**,** 1762-1775.

TISE, C. G. & BYERS, H. M. 2021. Genetics of recurrent pregnancy loss: a review. *Curr Opin Obstet Gynecol,* 33**,** 106-111.

TRICHE, E. W. & HOSSAIN, N. 2007. Environmental factors implicated in the causation of adverse pregnancy outcome. *Semin Perinatol,* 31**,** 240-2.

TYRRELL, J., RICHMOND, R. C., PALMER, T. M., FEENSTRA, B., RANGARAJAN, J., METRUSTRY, S., CAVADINO, A., PATERNOSTER, L., ARMSTRONG, L. L., DE SILVA, N. M., WOOD, A. R., HORIKOSHI, M., GELLER, F., MYHRE, R., BRADFIELD, J. P., KREINER-MOLLER, E., HUIKARI, V., PAINTER, J. N., HOTTENGA, J. J., ALLARD, C., BERRY, D. J., BOUCHARD, L., DAS, S., EVANS, D. M., HAKONARSON, H., HAYES, M. G., HEIKKINEN, J., HOFMAN, A., KNIGHT, B., LIND, P. A., MCCARTHY, M. I., MCMAHON, G., MEDLAND, S. E., MELBYE, M., MORRIS, A. P., NODZENSKI, M., REICHETZEDER, C., RING, S. M., SEBERT, S., SENGPIEL, V., SORENSEN, T. I., WILLEMSEN, G., DE GEUS, E. J., MARTIN, N. G., SPECTOR, T. D., POWER, C., JARVELIN, M. R., BISGAARD, H., GRANT, S. F., NOHR, E. A., JADDOE, V. W., JACOBSSON, B., MURRAY, J. C., HOCHER, B., HATTERSLEY, A. T., SCHOLTENS, D. M., DAVEY SMITH, G., HIVERT, M. F., FELIX, J. F., HYPPONEN, E., LOWE, W. L., JR., FRAYLING, T. M., LAWLOR, D. A., FREATHY, R. M. & EARLY GROWTH GENETICS, C. 2016. Genetic Evidence for Causal Relationships Between Maternal Obesity-Related Traits and Birth Weight. *JAMA,* 315**,** 1129-40.

UFFELMANN, E., HUANG, Q. Q., MUNUNG, N. S., DE VRIES, J., OKADA, Y., MARTIN, A. R., MARTIN, H. C., LAPPALAINEN, T. & POSTHUMA, D. 2021. Genome-wide association studies. *Nature Reviews Methods Primers,* 1**,** 59.

UFFELMANN, E., POSTHUMA, D. & PEYROT, W. J. 2023. Genome-wide association studies of polygenic risk score-derived phenotypes may lead to inflated false positive rates. *Sci Rep,* 13**,** 4219.

VAN DIJK, M. & OUDEJANS, C. 2013. (Epi)genetics of pregnancy-associated diseases. *Front Genet,* 4**,** 180.

VICKERS, A. J. & ELKIN, E. B. 2006. Decision curve analysis: a novel method for evaluating prediction models. *Med Decis Making,* 26**,** 565-74.

VIDMAR, S. I., COLE, T. J. & PAN, H. 2013. Standardizing Anthropometric Measures in Children and Adolescents with Functions for Egen: Update. *The Stata Journal,* 13**,** 366-378.

VIEIRA, M. C., MCCOWAN, L. M. E., GILLETT, A., POSTON, L., FYFE, E., DEKKER, G. A., BAKER, P. N., WALKER, J. J., KENNY, L. C., PASUPATHY, D. & CONSORTIUM, S. 2017. Clinical, ultrasound and molecular biomarkers for early prediction of large for gestational age infants in nulliparous women: An international prospective cohort study. *PLoS One,* 12**,** e0178484.

VOSKAMP, B. J., PEELEN, M., RAVELLI, A. C. J., VAN DER LEE, R., MOL, B. W. J., PAJKRT, E., GANZEVOORT, W. & KAZEMIER, B. M. 2020. Association between fetal sex, birthweight percentile and adverse pregnancy outcome. *Acta Obstet Gynecol Scand,* 99**,** 48-58.

WAHAB, R. J., JADDOE, V. W. V., VAN KLAVEREN, D., VERMEULEN, M. J., REISS, I. K. M., STEEGERS, E. A. P. & GAILLARD, R. 2022. Preconception and early-pregnancy risk prediction for birth complications: development of prediction models within a population-based prospective cohort. *BMC Pregnancy Childbirth,* 22**,** 165.

WANG, J. W., LYU, Y. N., QIAO, B., LI, Y., ZHANG, Y., DHANYAMRAJU, P. K., BAMME, Y., YU, M. D., YANG, D. & TONG, Y. Q. 2021. Cell-free fetal DNA testing and its correlation with prenatal indications. *BMC Pregnancy Childbirth,* 21**,** 585.

WANG, N., GUO, H., JING, Y., ZHANG, Y., SUN, B., PAN, X., CHEN, H., XU, J., WANG, M., CHEN, X., SONG, L. & CUI, W. 2023. Development and validation of risk prediction models for large for gestational age infants using logistic regression and two machine learning algorithms. *J Diabetes,* 15**,** 338-348.

WARRINGTON, N. M., BEAUMONT, R. N., HORIKOSHI, M., DAY, F. R., HELGELAND, O., LAURIN, C., BACELIS, J., PENG, S., HAO, K., FEENSTRA, B., WOOD, A. R., MAHAJAN, A., TYRRELL, J., ROBERTSON, N. R., RAYNER, N. W., QIAO, Z., MOEN, G. H., VAUDEL, M., MARSIT, C. J., CHEN, J., NODZENSKI, M., SCHNURR, T. M., ZAFARMAND, M. H., BRADFIELD, J. P., GRARUP, N., KOOIJMAN, M. N., LI-GAO, R., GELLER, F., AHLUWALIA, T. S., PATERNOSTER, L., RUEEDI, R., HUIKARI, V., HOTTENGA, J. J., LYYTIKAINEN, L. P., CAVADINO, A., METRUSTRY, S., COUSMINER, D. L., WU, Y., THIERING, E., WANG, C. A., HAVE, C. T., VILOR-TEJEDOR, N., JOSHI, P. K., PAINTER, J. N., NTALLA, I., MYHRE, R., PITKANEN, N., VAN LEEUWEN, E. M., JORO, R., LAGOU, V., RICHMOND, R. C., ESPINOSA, A., BARTON, S. J., INSKIP, H. M., HOLLOWAY, J. W., SANTA-MARINA, L., ESTIVILL, X., ANG, W., MARSH, J. A., REICHETZEDER, C., MARULLO, L., HOCHER, B., LUNETTA, K. L., MURABITO, J. M., RELTON, C. L., KOGEVINAS, M., CHATZI, L., ALLARD, C., BOUCHARD, L., HIVERT, M. F., ZHANG, G., MUGLIA, L. J., HEIKKINEN, J., CONSORTIUM, E. G. G., MORGEN, C. S., VAN KAMPEN, A. H. C., VAN SCHAIK, B. D. C., MENTCH, F. D., LANGENBERG, C., LUAN, J., SCOTT, R. A., ZHAO, J. H., HEMANI, G., RING, S. M., BENNETT, A. J., GAULTON, K. J., FERNANDEZ-TAJES, J., VAN ZUYDAM, N. R., MEDINA-GOMEZ, C., DE HAAN, H. G., ROSENDAAL, F. R., KUTALIK, Z., MARQUES-VIDAL, P., DAS, S., WILLEMSEN, G., MBAREK, H., MULLER-NURASYID, M., STANDL, M., APPEL, E. V. R., FONVIG, C. E., et al. 2019. Maternal and fetal genetic effects on birth weight and their relevance to cardio-metabolic risk factors. *Nat Genet,* 51**,** 804-814.

WEEDON, M. N., CLARK, V. J., QIAN, Y., BEN-SHLOMO, Y., TIMPSON, N., EBRAHIM, S., LAWLOR, D. A., PEMBREY, M. E., RING, S., WILKIN, T. J., VOSS, L. D., JEFFERY, A. N., METCALF, B., FERRUCCI, L., CORSI, A. M., MURRAY, A., MELZER, D., KNIGHT, B., SHIELDS, B., SMITH, G. D., HATTERSLEY, A. T., DI RIENZO, A. & FRAYLING, T. M. 2006. A common haplotype of the glucokinase gene alters fasting glucose and birth weight: association in six studies and population-genetics analyses. *Am J Hum Genet,* 79**,** 991-1001.

WEI, J. N., SUNG, F. C., LI, C. Y., CHANG, C. H., LIN, R. S., LIN, C. C., CHIANG, C. C. & CHUANG, L. M. 2003. Low birth weight and high birth weight infants are both at an increased risk to have type 2 diabetes among schoolchildren in taiwan. *Diabetes Care,* 26**,** 343-8.

WEISSMANN-BRENNER, A., SIMCHEN, M. J., ZILBERBERG, E., KALTER, A., WEISZ, B., ACHIRON, R. & DULITZKY, M. 2012. Maternal and neonatal outcomes of large for gestational age pregnancies. *Acta Obstet Gynecol Scand,* 91**,** 844-9.

WEST, J., LAWLOR, D. A., FAIRLEY, L., BHOPAL, R., CAMERON, N., MCKINNEY, P. A., SATTAR, N. & WRIGHT, J. 2013. UK-born Pakistani-origin infants are relatively more adipose than white British infants: findings from 8704 mother-offspring pairs in the Born-in-Bradford prospective birth cohort. *J Epidemiol Community Health,* 67**,** 544-51.

WILCOX, A. J. 2001. On the importance--and the unimportance--of birthweight. *Int J Epidemiol,* 30**,** 1233-41.

WOJCIK, M. H., REIMERS, R., POORVU, T. & AGRAWAL, P. B. 2020. Genetic diagnosis in the fetus. *J Perinatol,* 40**,** 997-1006.

WRIGHT, C. M., WILLIAMS, A. F., ELLIMAN, D., BEDFORD, H., BIRKS, E., BUTLER, G., SACHS, M., MOY, R. J. & COLE, T. J. 2010. Using the new UK-WHO growth charts. *BMJ,* 340**,** c1140.

WRIGHT, J., SMALL, N., RAYNOR, P., TUFFNELL, D., BHOPAL, R., CAMERON, N., FAIRLEY, L., LAWLOR, D. A., PARSLOW, R., PETHERICK, E. S., PICKETT, K. E., WAIBLINGER, D., WEST, J. & BORN IN BRADFORD SCIENTIFIC COLLABORATORS, G. 2013. Cohort Profile: the Born in Bradford multi-ethnic family cohort study. *Int J Epidemiol,* 42**,** 978-91.

YAJNIK, C. S. 2001. The Insulin Resistance Epidemic in India: Fetal Origins, Later Lifestyle, or Both? *Nutrition Reviews,* 59**,** 1-9.

YU, Z., HAN, S., ZHU, J., SUN, X., JI, C. & GUO, X. 2013. Pre-pregnancy body mass index in relation to infant birth weight and offspring overweight/obesity: a systematic review and meta-analysis. *PLoS One,* 8**,** e61627.

ZHANG, G., BACELIS, J., LENGYEL, C., TERAMO, K., HALLMAN, M., HELGELAND, O., JOHANSSON, S., MYHRE, R., SENGPIEL, V., NJOLSTAD, P. R., JACOBSSON, B. & MUGLIA, L. 2015. Assessing the Causal Relationship of Maternal Height on Birth Size and Gestational Age at Birth: A Mendelian Randomization Analysis. *PLoS Med,* 12**,** e1001865.