Relative maternal protection against type 1 diabetes: a combined analysis of five observational studies

Lowri A Allen[1], Peter N Taylor[1], Annelie Carlsson[2], Diane P Fraser[3,4], William A Hagopian[5], Emma Hedlund[6], Anita V Hill [4,7], Angus G Jones[8,9], Johnny Ludvigsson[10], Georgina L Mortimer[11], Suna Onengut-Gumuscu [12], Maria J Redondo [13], Stephen S Rich [12], Claire L Williams [11], Kathleen M Gillespie [11]*, Colin M Dayan [1]*, Richard A Oram [8,9]*

*Equal contribution.

[1] Diabetes Research Group, Cardiff University, University Hospital of Wales, Cardiff, UK.

[2] Department of Clinical Sciences, Lund, Lund University, Skånes University Hospital, Sweden.

[3] Department of Clinical and Biomedical Sciences, University of Exeter Medical School, Exeter, UK.

[4]NIHR Exeter Biomedical Research Centre, Exeter Centre of Excellence in Diabetes, University of Exeter, Exeter, UK.

[5] Department of Pediatrics, Indiana University School of Medicine, USA.

[6] Department of Clinical Sciences, Lund University, Lund, and Kristianstad Central Hospital, Kristianstad, Sweden.

[7] NIHR Exeter Clinical Research Facility, Royal Devon University Healthcare NHS Foundation Trust, Exeter, UK.

[8] Endocrinology and Diabetes, Royal Devon University Healthcare NHS Foundation Trust, Devon, UK.

[9] Department of Clinical and Biomedical Sciences, University of Exeter Medical School, Exeter, UK.

[10] Crown Princess Victoria Children´s Hospital and Division of Pediatrics, Department of Biomedical and Clinical Sciences, Linköping University, Linköping, Sweden.

[11] Translational Health Sciences, Bristol Medical School, University of Bristol, UK.

[12] Department of Genome Sciences, University of Virginia, Charlottesville, Virginia, USA.

[13] Pediatric Diabetes and Endocrinology, Texas Children's Hospital. Department of Pediatrics, Baylor College of Medicine

ORCID identifiers:

Lowri Allen: 0000-0003-3814-8564

Peter Taylor: 0000-0002-3436-422X

Annelie Carlsson: 0000-0002-5608-3437

Diane Fraser: 0000-0003-0988-8926

William Hagopian: 0000-0003-2979-0475

Emma Hedlund: 0000-0001-7707-4540

Anita Hill: 0000-0002-0901-4834

Angus Jones: 0000-0002-0883-7599

Johnny Ludvigsson: 0000-0003-1695-5234

Georgina Mortimer: 0000-0003-3759-399X

Suna Onengut-Gumuscu: 0000-0002-6563-8334

Maria Redondo: 0000-0001-5871-4645

Stephen Rich: 0000-0003-3872-7793

Claire Williams: 0000-0002-6641-8018

Kathleen Gillespie: 0000-0002-3009-8032

Colin Dayan: 0000-0002-6557-3462

Richard Oram: 0000-0003-3581-8980

Corresponding author:

Dr Lowri Allen

Preferred degree: MBChB

Affiliation: Diabetes Research Group, Cardiff University

Full address: Diabetes Research Group, C2 Link Corridor, University Hospital of Wales, Cardiff, UK. CF14 4XN.

E-mail: AllenLA7@cardiff.ac.uk Telephone: +442920742182

Disclosures/Conflict of interest:

The authors meet criteria for authorship as recommended by the International Committee of Medical Journal Editors (ICMJE) and did not receive payment related to the development of this manuscript. LAA, PNT, AC, DPF, EH, AVH, AGJ, JL, GLM, SOG, CLW, KMG declare no competing interests. MJR is an associate editor at Diabetologia. WAH declares grants, contracts, or payments from NIDDK/NIH, Sanofi, JDRF, Janssen Pharmaceuticals R+D, American Diabetes Association. SSR declares grants from the following entities: NIH NIDDK, Juvenile Diabetes Research Foundation, Leona M & Harry B Helmsley Charitable Trust, the receipt of consulting fees from Westat and his position as Ad Hoc Associate Editor for the American Diabetes Association. CMD declares consulting fees or payments from Sanofi and Provention Bio, holds a patent with Provention Bio, participates on the advisory boards for DOMPE and Diamyd, and is a trustee and the clinical secretary of the British Society for Immunology. RAO declares grants, contracts or payments from Randox Ltd, Janssen, Provention Bio, Sanofi.

Keywords: Familial, Family, Maternal, Offspring, Paternal, Protection, Transmission, Type 1 diabetes.

Abstract (250 words)

Context: Maternal (versus paternal) type 1 diabetes is associated with a relative reduction in type 1 diabetes risk in offspring during early life.

Objective: To determine whether this effect extends into later life. To clarify the importance of intrauterine exposure to maternal type 1 diabetes, and baseline genetic susceptibility in this context.

Methods: We compared the proportion of individuals with type 1 diabetes diagnosed age 0-88 years with affected mothers and fathers across five observational studies (n=11,475), and used random-effects meta-analyses to generate overall effect estimates. We examined this by age at diagnosis, and timing of parental diagnosis relative to offspring birth. We compared the type 1 diabetes genetic risk score (T1D-GRS2) of individuals with affected mothers and fathers.

Results: Almost half as many individuals with type 1 diabetes had an affected mother versus father (OR 0.55 (95% CI 0.48, 0.64), p<0.0001). A lower proportion of individuals with affected mothers than fathers was apparent even amongst individuals diagnosed as adults (>18 years) (OR 0.63 (95% CI 0.43, 0.91), p=0.01). The lower proportion of individuals with maternal versus paternal type 1 diabetes was only observed if maternal diagnosis preceded offspring birth (OR

0.51 (95% CI 0.37, 0.70), p<0.001 versus OR 0.97 (95% CI 0.69, 1.38), p=0.87 after birth). T1D-GRS2 was similar between individuals with affected mothers and fathers (p=0.25).

Conclusion: Our analyses suggest intrauterine exposure to maternal type 1 diabetes is associated with long-lasting relative protection against offspring type 1 diabetes, which is independent of genetic susceptibility as measured by T1D-GRS2.

Introduction

Type 1 diabetes is a chronic autoimmune disease, resulting from a combination of genetic predisposition and environmental exposures. Individuals with a family history are 8-15 times more susceptible than the background population[1-3]. The risk is higher if the affected relative is the father versus mother[4].

Almost 40 years ago, Warram et al. reported, in a cohort of 289 individuals with type 1 diabetes, the risk of type 1 diabetes was approximately four times higher in offspring of men versus women (6.1±1.8% versus 1.3±0.9%)[5]. Larger prospective studies following offspring of individuals with type 1 diabetes have since replicated the finding of fewer offspring of women versus men developing the disease during early life[6-8]. This is consistent with studies reporting fewer individuals with type 1 diabetes have affected mothers than fathers[9-13]. Detailed descriptions of these studies are published elsewhere[4]. Together they demonstrate type 1 diabetes is around half as common during early life amongst offspring of women versus men with type 1 diabetes [4,6,7,9-11,13-15]. Comparison of the risk against that in individuals with affected siblings confirms this is due to a lower risk than expected amongst offspring of affected mothers [9,10,15]. Maternal protection is only relative, as the risk remains higher than in the background population (2-3% lifetime risk versus 0.3%) [4,13].

To date, studies have typically followed offspring to age 20-30 years [4,6,7,9-11,13-15]. A recent study by Wei et al. reported relative maternal protection in offspring diagnosed with type 1 diabetes in childhood (<18 years) and adulthood (>18 years), but did not include offspring diagnosed beyond age 30[14]. Tillil et al. published the only study including offspring diagnosed beyond age 30 years, but included few individuals diagnosed as older adults (n=138 >25 years, n=23 >40 years.)[10] *It remains unclear whether maternal protection simply delays disease onset, which could still be beneficial given later onset is associated with improved outcomes [16], or whether it represents a long-term effect persisting into later life.*

A range of potential mechanisms have been considered, and can be broadly grouped as genetic and environmental[4]. *One approach to disentangling genetic from environmental contributions is to examine whether protection occurs only when mothers are diagnosed before pregnancy, which would implicate intrauterine exposure as an important factor.* A few small studies have explored this, with conflicting results [6,13,15-17].

This study utilised data from five observational studies of individuals with type 1 diabetes (n=11,475). Our aims were first to determine whether any relative protection associated with maternal type 1 diabetes is confined to early life. Second, to clarify whether differences in parental rates of type 1 diabetes amongst individuals with type 1 diabetes depend on parental disease being diagnosed prior to offspring birth as suggested by some reports. Finally, to determine if relative maternal protection depends on differences in type 1 diabetes genetic susceptibility (measured by type 1 diabetes genetic risk score (T1D-GRS2)) between participants with affected mothers and fathers.

Materials and methods

This study utilised data from five observational studies of individuals with type 1 diabetes that collected family history and genetics data in the form of T1D-GRS2.

The Barts Oxford Family Study (BOX) has recruited individuals with newly diagnosed type 1 diabetes prior to age 21 years since 1985 in the Oxfordshire Health Authority Region, UK [18,19]. Participants and relatives (with and without type 1 diabetes) have been prospectively followed. Similarly, Better Diabetes Diagnosis (BDD) and StartRight are also observational studies of individuals with newly diagnosed diabetes (BDD; individuals diagnosed with type 1 diabetes before age 18 years in Sweden since 2005[20], StartRight; individuals with newly diagnosed diabetes ≥17 years in the UK between 2015-2020.) Type 1 Diabetes Genetic Consortium (T1DGC) 'families dataset'[21] and TrialNet Pathway to Prevention (TrialNet PTP)[22] represent two additional sources of family and T1D-GRS2 data collected from individuals with type 1 diabetes, but were designed to over-represent individuals with affected relatives. The former is an international study of predominantly affected sibling pairs with type 1 diabetes conducted between 2004-2009. The latter has recruited and followed first-degree relatives of individuals with type 1 diabetes positive for at least one type 1 diabetes related autoantibody since 2000. Detailed study descriptions are provided (table 1.)

Inclusion criteria for this study were type 1 diabetes and the availability of family history. Individuals were excluded if their relation to an affected relative was uncertain. All individuals in BOX (n=3,040), BDD (n=3,896), T1DGC 'families dataset' (n=2,662) were eligible. In TrialNet PTP, 1,316 individuals progressed to type 1 diabetes and were eligible. Of 1,800 individuals recruited to StartRight, 561 had type 1 diabetes and were eligible. 11,475 participants were ultimately included (supplementary figure 1[23]).

Data collection:

Family history was obtained through questioning of participants and/or relatives at recruitment and during follow up. Parents with type 1 diabetes were recorded in all studies except StartRight, in which relatives with diabetes were recorded, but diabetes type was not. Consistent with our prespecified analysis plan, treatment with insulin only was used as a surrogate for type 1 diabetes in relatives in StartRight[24]. Results using more restrictive definitions were similar (supplementary table 1[23].)

Data regarding the timing of parents' diagnoses relative to offspring birth were available in BOX and TrialNet PTP. Data regarding affected siblings were available for all studies except BDD.

T1D-GRS2 is a validated measure of polygenic susceptibility to type 1 diabetes. T1D-GRS2 was generated as previously described in T1DGC and StartRight[25,26]. T1D-GRS2 was generated in BDD

and BOX with a custom SNP genotyping panel developed by Dr. Hagopian with LGC Genomics using KASP genotyping. T1D-GRS2 in TrialNet PTP was generated as previously described[25], using genotyping from an Illumina Infinium T1DExomeChip SNP array, imputed to the TOPMed reference panel (supplementary text 1[23]).

T1D-GRS2 was available for all BDD, TrialNet PTP, T1DGC participants. In StartRight, T1D-GRS2 was available for 511 (95%) participants. In BOX, initiated in 1985, T1D-GRS2 was available for 1,388 (46%) individuals. Prospective and retrospective DNA sampling commenced in 1998 but fewer existing families provided samples. Age at diagnosis and parental type 1 diabetes were comparable between individuals who did and did not provide samples (median diagnosis age 10.5 vs 10.1 years, 2.7% affected mothers vs 2.9%, 5.0% affected fathers vs 5.0%).

Statistical analysis:

Analyses were undertaken separately within each study initially. Due to the heterogeneity of studies, unadjusted random effects meta-analyses were used to derive effect estimates across studies.

The proportion of missing data are described. Since missing data were minimal (<1%) for most analyses, complete case analyses were undertaken. 11% of cases had missing T1D-GRS2,

predominantly due to missing data in BOX. BOX was attributed lower weighting accordingly. BOX results were consistent with other studies.

The proportion of individuals with type 1 diabetes with affected mothers and fathers are presented as frequencies (percentages) for individual studies. Participants with both parents affected (0.26% overall, supplementary table 2[23]) were included in both counts. Due to sibling data being unavailable for BDD, and differences in study design resulting in over-representation of affected siblings in TrialNet PTP and T1DGC (supplementary table 3[23]), individuals with and without affected siblings were not treated differently.

The ratio of the odds of an affected mother to the odds of an affected father, with 95% confidence intervals (OR, (95%CIs)) is presented for each study. Random effects meta-analysis was used to derive an overall OR across studies. The same methods were used to generate results for individuals diagnosed $\leq$18 years and >18 years.

The mean difference in age and T1D-GRS2 were compared between those with affected mothers versus fathers. Standardised mean differences across studies were derived from random effects meta-analyses.

Using data from BOX and TrialNet PTP, the proportion of individuals with an affected mother versus father was compared between those whose parents were diagnosed before and after birth.

Additional analyses

A limitation of our study is that complete data regarding offspring who did not develop type 1 diabetes were not available for all cohorts. Reporting the proportion of individuals with type 1 diabetes with affected mothers versus fathers is consistent with the approach of several publications[9-13]. These publications generated results similar to those obtained from prospective studies including all offspring of men and women with type 1 diabetes[6-8]. However, an observed difference in parental rates of type 1 diabetes could theoretically be explainable by: (i) an excess of males relative to females with type 1 diabetes, (ii) more offspring being born to men than women with type 1 diabetes, (iii) an excess risk of type 1 diabetes amongst offspring of affected men, as opposed to relative maternal protection. We undertook a series of analyses to address each possibility.

(i) Type 1 diabetes is relatively more common in males than females[27-29]. This study was not designed to adjust for this. However, we examined the sex distribution of participants. Published literature indicates that the recognised male preponderance

of type 1 diabetes is driven by an excess of male diagnoses at older ages[27]. Depending on the population studied, the ratio of male to females amongst individuals diagnosed after age 15 years is 1.3-2.2. However, prior to age 15 years the sex distribution is relatively equal [27-29]. In a Swedish nationwide study, a significant male bias was observed amongst individuals diagnosed age 15-19 and 20-24 years (ratios 1.59, 2.08 respectively) but not 10-14 years (0.94) [30]. We therefore undertook a sensitivity analysis restricted to individuals with parents diagnosed prior to age 15 years.

(ii) Data regarding the total number of children born to men and women with type 1 diabetes were only available in BOX. An unpaired student t-test was used to compare the mean number of children born to men and women with type 1 diabetes.

(iii) BOX was the only study to record complete sibling data *and* not bias recruitment towards individuals with affected siblings. We described the proportion of participants with affected siblings compared with affected mothers and fathers.

All statistical analyses were performed using STATAv17.0. $p<0.05$ was deemed significant.

Ethical approval was obtained prior to establishing each individual study[18-20,22,26,31,32]. This study fell within the remit of original approvals. All participants gave informed consent, with specific assent procedures for children, and oversight from at least one ethical review board (minimum one per country for international studies), in accordance with the Declaration of Helsinki).

Results

11,475 individuals with type 1 diabetes were eligible for inclusion. Participants were diagnosed age 0-88 years (median 10, IQR 6,14). 1,052 (9.2% of participants) individuals were diagnosed after age 18 years, 648 (5.6%) >25 years, 515 (4.5%) >30 years, 292 (2.5%) >40 years (supplementary table 4[23].)

Fewer individuals with type 1 diabetes had an affected mother versus father (OR 0.55 (95%CI 0.48, 0.64), p<0.0001) (figure 1, supplementary tables 2 and 3[23]). Results were comparable across studies. Specifically, results were similar in StartRight (individuals diagnosed with type 1 diabetes as adults (≥17 years) (4.5% versus 7.8%, OR 0.55 (0.33, 0.91) p=0.02)) and studies of individuals diagnosed in early life (BOX (<21 years), 2.8% versus 5.0%, OR 0.55 (0.42, 0.72), p<0.0001, and BDD (<18 years) 2.9% versus 5.5%, OR 0.52 (0.41, 0.65), p<0.0001) (figure 1).

Significantly fewer individuals had an affected mother compared with father in meta-analyses of participants diagnosed >18 years (OR 0.63 (0.43, 0.91) p=0.01) and ≤18 years (OR 0.55 (0.47, 0.64), p<0.0001) (figure 2). Analyses using cut-offs between 13-19 years (supplementary table 5[23]) yielded similar results.

Age at diagnosis was similar amongst participants with affected mothers and fathers (overall Hedges's g standardised mean difference 0.05 years (95%CI -0.07, 0.16), p=0.45) (figure 3).

Relative maternal protection

To ascertain whether our findings indicate relative maternal protection against offspring type 1 diabetes we sought to exclude alternative explanations.

i) Is the effect due to type 1 diabetes being more common in males than females?

The ratio of male to female participants was 1.15 (supplementary table 6[23]). This is similar to published results from comparable populations[28]. Assuming the sex distribution of type 1 diabetes is stable across generations, as described by Gale et al. [27], this slight male excess is too small to account for the magnitude of the difference in maternal versus paternal disease in our study. Furthermore, restricting analysis to individuals with parents diagnosed before age 15

years, amongst whom no significant sex bias is expected, we still found a lower proportion of individuals with affected mothers (OR 0.69 (0.50, 0.95) p=0.02) (supplementary figure 2[23]).

ii) Is it due to more offspring being born to men than women with type 1 diabetes?

In BOX, the mean number of children born to men and women with type 1 diabetes was similar (mean 2.4 children for both, p=0.83) (supplementary table 7[23]).

iii) Is it due to an excess risk of type 1 diabetes amongst offspring of men with type 1 diabetes?

In BOX (n=3,040), the proportion of participants with affected siblings and fathers was comparable (n=168 (5.5%), n=152 (5.0%)). The proportion with an affected mother was lower (n=85 (2.8%.)) This is in-keeping with offspring of affected mothers being at relatively lower risk, as opposed to excessive risk amongst offspring of affected fathers.

Intrauterine exposure to maternal type 1 diabetes

Amongst participants with parents diagnosed prior to their birth, significantly fewer had an affected mother than father (OR 0.51 (0.37, 0.70), p<0.001) (figure 4A.) A lower proportion of individuals with an affected mother was not apparent amongst individuals with parents diagnosed after their birth (OR 0.97 (0.69, 1.38), p=0.87, figure 4B).

Parents diagnosed prior to offspring birth are inevitably younger at diagnosis than parents diagnosed after offspring birth. Earlier disease onset is associated with increased genetic susceptibility and increased transmission to offspring. We therefore expected more participants to have parents diagnosed before versus after birth. This is what we observed for fathers (table2) (BOX: n=38 (1.3%) after birth, n=114 (3.8%) pre-birth, TrialNet PTP n=27 (2.1%) after birth, n=193 (14.7%) pre-birth). This was less apparent for mothers, due to fewer than expected participants having mothers diagnosed prior to their birth (BOX: n=33 (1.1%) after birth, n=49 (1.6%) pre-birth, TrialNet PTP n=31 (2.4%) after birth, n=120 (9.1%) pre-birth.) These findings suggest intrauterine exposure to maternal type 1 diabetes is important.

Genetic susceptibility

T1D-GRS2 was similar between participants with affected mothers versus fathers (overall Hedges's g standardised mean difference -0.07 (95%CI -0.20, 0.05), p=0.25) (figure 5). T1D-GRS2 was lower in StartRight than other studies, in-keeping with the older age at diagnosis of participants (figure 5). Results of a sensitivity analysis excluding StartRight were comparable (overall Hedges's g standardised mean difference -0.09 (95% CI -0.22, 0.05), p=0.20 (supplementary figure3[23]).

Discussion

In this large study (n=11,475) incorporating meta-analyses across five studies, we confirm around half as many individuals with type 1 diabetes have affected mothers than fathers (figure 1). We report for the first time, that this is evident even amongst individuals diagnosed in later adulthood (figure 2), and demonstrate no significant difference in age at diagnosis between individuals with affected mothers and fathers (figure 3). We only observed a lower proportion of participants with affected mothers when maternal diagnosis was prior to offspring birth, resolving earlier uncertainty regarding this (figure 4). *Through* exploratory analyses, we demonstrate our findings to be in-keeping with relative maternal protection against offspring type 1 diabetes. *Finally, we show for the first time, that genetic susceptibility (T1D-GRS2) was similar in individuals with affected mothers and fathers (figure 5), suggesting that the observed difference is not explained by inherited genetic risk*. Together, our data provide novel insights into relative maternal protection, demonstrating this is an enduring phenomenon that follows intrauterine exposure to maternal type 1 diabetes, which is independent of inherited type 1 diabetes genetic susceptibility (measured by T1D-GRS2), and continues to protect offspring into adulthood.

The study population was confined to individuals with type 1 diabetes. Offspring without type 1 diabetes were not included. Therefore, to be confident our observation of fewer participants having affected mothers than fathers was indicative of relative maternal protection, we sought

to exclude alternative explanations. First, type 1 diabetes is relatively more common in males, and this could account for fewer participants having affected mothers than fathers. Consistent with published results from comparable populations, we observed a slight male bias amongst participants (male:female ratio 1.15, supplementary table 6[23].) This is insufficient to account for the magnitude of the difference in the proportion of affected mothers and fathers. Furthermore, even after restricting analysis to individuals with parents diagnosed prior to age 15 years (amongst whom no significant sex bias is expected [27-30]), we still found fewer individuals had affected mothers than fathers (supplementary figure 2[23]). These findings suggest our results are not due to an excess of males with type 1 diabetes.

An alternative possibility is that more children are born to men than women with type 1 diabetes. For example, it is well documented that maternal type 1 diabetes is associated with an increased risk of complications for both mother and baby, and that pregnancy can make the management of type 1 diabetes significantly more challenging e.g. due to pregnancy being a state of relative insulin resistance[33]. It is therefore possible that some women with type 1 diabetes may choose to have fewer pregnancies or even avoid pregnancy altogether, due to concerns about the associated risks. However, we report that in BOX, men and women with type 1 diabetes had a similar number of children (mean 2.4 for both, supplementary table 7[23].) This is consistent with findings from a Danish cohort, which reported fewer individuals with type 1 diabetes had affected mothers than fathers despite men and women with type 1 diabetes having a similar number of children (2.16 vs 1.96)[15]. Taken together our analyses suggest that

1 our results indicate a true difference in type 1 diabetes risk between offspring of affected
2 mothers and fathers.

3

4 BOX was the only study to collect complete sibling data, and where participant recruitment was
5 not biased towards individuals with affected siblings. In BOX, a similar proportion of
6 participants had affected siblings and fathers (5.5%, 5.0% respectively), but fewer had affected
7 mothers (2.8%). This is consistent with findings of published studies[9,10,15], and in the context of
8 our other results, supports the phenomenon of relative maternal protection against offspring
9 type 1 diabetes, rather than an excess of paternal transmission.

10

11 We confirm that relative maternal protection is only observed if the mother already has
12 diabetes prior to offspring birth. Women diagnosed prior to pregnancy will inevitably be
13 younger at diagnosis than mothers diagnosed after pregnancy. Therefore, maternal age at
14 diagnosis could be an important confounder. This study was not designed to directly examine
15 this. Harjutsalo et al. reported that amongst offspring of mothers diagnosed with type 1
16 diabetes age 0-39 years, relative maternal protection was greater if mothers were diagnosed
17 before age 10 years[16]. Since a younger age at diagnosis is associated with high-risk type 1
18 diabetes susceptibility genes[34,35], it is plausible that if maternal protection is driven by reduced
19 maternal transmission of these genes, the impact might be greatest amongst offspring of
20 parents diagnosed earlier in life. However, our study contained approximately three times as
21 many children born to mothers with type 1 diabetes (n=436 vs 150), and we found that type 1

diabetes genetic susceptibility measured by T1D-GRS2 was similar between participants with affected mothers and fathers. We acknowledge that T1D-GRS2 is an imperfect measure of inherited genetic susceptibility, based on 67 specific SNPs, but in the absence of sufficient power to study individual genetic variants, our results suggest that aggregate genetic susceptibility to type 1 diabetes was not significantly different in individuals with type 1 diabetes with affected mothers versus fathers. Our results do not exclude the possibility of a genetic mechanism not detectable using T1D-GRS2, including a difference in rare variant inheritance between offspring of men and women with type 1 diabetes, nor the possibility of genetic imprinting, whereby the expression of inherited genes differs according to the parent from whom they are inherited. However, neither (as sole mechanisms) would account for our observation that maternal protection is only apparent when maternal disease is diagnosed prior to offspring birth. Furthermore, two large studies (including a GWAS) have concluded genetic imprinting is unlikely to significantly contribute to type 1 diabetes susceptibility in offspring of affected men and women [36,37]. Larger studies are required to definitively examine the interaction between timing of parental diagnosis and parental age at diagnosis.

Potentially important intrauterine exposures may include maternal hyperglycaemia, maternal islet antibodies, exogenous insulin therapy, maternal antibodies to insulin, and maternal antibodies to enterovirus (a type 1 diabetes trigger) [4,38-41]. Toxicity from hyperglycaemic exposure is deemed the principal driver for increased spontaneous abortions in women with type 1 diabetes[4,42-44]. Consequently, it is plausible that selective loss of fetuses exposed to severe hyperglycaemia may contribute to relative maternal protection. This is difficult to study,

as many pregnancy losses occur early in pregnancy and go unrecorded. However, our data suggest the number of children born to men and women with type 1 diabetes is comparable, (supplementary table 7[23]), consistent with a previous report [15].

For surviving offspring, intrauterine exposure to one or more of the exposures listed above may impact offspring type 1 diabetes susceptibility by directly influencing the development and maturation of the pancreatic beta cells and/or the immune system. As an example, intrauterine exposure to hyperglycaemia could plausibly stimulate beta cell growth, development and maturation thereby reducing offspring type 1 diabetes susceptibility. However, there may be a threshold whereby exposure to very marked degrees of hyperglycaemia may lead to beta cell exhaustion and therefore an increased risk of type 1 diabetes[45]. Either separately, or in concert, the transplacental transfer of maternal antibodies could directly induce immunotolerance against autoantigens and/or promote efficient clearance of autoreactive T-cells[46]. A more detailed description of potentially relevant intrauterine exposures and the effects they could plausibly exert on the development of the pancreas and/or immune system are provided elsewhere[4].

Intrauterine exposure to these different components of maternal type 1 diabetes could also have more indirect effects on offspring type 1 diabetes susceptibility e.g. via epigenetics. Epigenetic mechanisms have been documented to influence T cell differentiation and maturation, insulin gene expression and beta cell function and autoantigen expression[47]. Such

epigenetic changes can be detected even prior to the development of islet autoimmunity, suggesting a possible role for epigenetics in the pathogenesis of the disease[48]. It is plausible that these epigenetic changes could be driven, at least in part, by intrauterine exposures, including potentially maternal type 1 diabetes. Knorr et al. reported evidence of epigenetic changes in adolescent offspring of mothers with type 1 diabetes who had been exposed to hyperglycaemia in utero[49]. Larger studies are required to validate this finding. Future studies should consider that there may not be one sole mechanism through which these exposures influence type 1 diabetes susceptibility, and that some of these mechanisms could complement or interact with one another.

Insights into the relative importance of exposure to maternal hyperglycaemia could be gained from comparing the type 1 diabetes risk of offspring of parents with different forms of diabetes. A systematic review published in 2019 reported that the risk of type 1 diabetes in offspring of mothers with gestational diabetes was higher than for mothers without diabetes, but lower than for mothers with type 1 diabetes[50]. The risk of type 1 diabetes was not significantly different between offspring of mothers with type 2 diabetes and mothers without diabetes. However, maternal diabetes diagnoses were not consistently and well characterised, relatively few mothers had type 2 diabetes, and important potential confounders were not addressed. Large, robustly designed studies of offspring of parents with different forms of diabetes are required. Ultimately, studies that include measures of maternal glycaemia at different time points during pregnancy are required to define the relationship between exposure to maternal hyperglycaemia in different forms of maternal diabetes and offspring type 1 diabetes risk. Such

studies should consider that birthweight may be either a confounder or an effect mediator in the relationship between exposure to maternal hyperglycaemia and offspring type 1 diabetes risk[4].

Strengths of our study include the use of data from five large studies (n=11,475), encompassing individuals diagnosed with type 1 diabetes between ages 0-88 years, over more than three decades, recruited from four continents. The heterogeneity of the population suggests our findings likely apply to a broad population of individuals with type 1 diabetes. The availability of T1D-GRS2 as a measure of genetic susceptibility incorporating HLA- and non-HLA- genetic loci in these individuals provides important novel insights. Furthermore, our study includes more individuals diagnosed in later adulthood than published studies. The largest study including participants with adult-onset type 1 diabetes published by Wei et al. (n=3,240) included only offspring diagnosed before age 30 years. Tillil et al. published the only study including individuals diagnosed at older ages. Our study included over three times as many offspring diagnosed beyond age 25 years (n=648 (38 with an affected mother, 47 with an affected father) versus 138 [10]). Larger studies of participants diagnosed in later life are required to confirm our findings.

The main limitation is that offspring without type 1 diabetes were not included, which restricts causal inference. Prospective lifelong follow up of all offspring of men and women with type 1 diabetes (regardless of diabetes status) would be optimal. Publications attempting

this approach to date have only followed offspring into early adult life [4,6,7,9-11,13-15]. As registries of routinely-collected clinical data mature, they will become invaluable resources for examining type 1 diabetes risk over the entire life-course of offspring. However, they are unlikely to routinely collect data such as T1D-GRS2.

Additional limitations include data regarding parental diagnoses timing only being available in selected studies. Another potential limitation was that family history was obtained by questioning participants and families. Parental type 1 diabetes could have been missed if an individual was estranged from a parent, but this is unlikely to disproportionately exclude more affected mothers than fathers. In StartRight, not specifically recording the type of diabetes in relatives posed a misclassification risk. However, use of increasingly restrictive definitions of parental type 1 diabetes did not impact our results, which were consistent with those from the studies which accurately recorded parental diagnoses. Despite its relatively low weighting in meta-analyses (reflecting n=561), StartRight was important to include as the largest, best characterised study of adult-onset type 1 diabetes including family and T1D-GRS2 data.

In conclusion, our findings suggest that, independently of genetic susceptibility, intrauterine exposure to maternal type 1 diabetes is associated with relative protection against offspring type 1 diabetes that persists into adulthood. Further studies are required to identify the key exposures involved and to confirm these observations in larger prospective datasets.

A better understanding of how maternal type 1 diabetes results in a relative reduction in type 1 diabetes in offspring may provide opportunities to develop interventions that could reduce type 1 diabetes risk in predisposed individuals.

Data availability Statement:

Data can be requested directly from the data holders for each study and/or accessed via public repositories including dbGaP, EGA.

References

1. Weires MB, Tausch B, Haug PJ, Edwards CQ, Wetter T, Cannon-Albright LA. Familiality of diabetes mellitus. *Exp Clin Endocrinol Diabetes*. Nov 2007;115(10):634-40. doi:10.1055/s-2007-984443
2. Barrett JC CD, Concannon P, Akolkar B, Cooper JD, Erlich HA, Julier C, Morahan G, Nerup J, Nierras C, Plagnol V, Pociot F, Schuilenburg H, Smyth DJ, Stevens H, Todd JA, Walker NM, Rich SS; Type 1 Diabetes Genetics Consortium. . Genome-wide association study and meta-analysis find that over 40 loci affect risk of type 1 diabetes. . *Nat Genet*. 2009;41(6):703-7. doi:10.1038/ng.381
3. Parkkola A HT, Ryhanen SJ, Ilonen J, Knip M, the Finnish Pediatric Diabetes Register. Extended family history of type 1 diabetes and phenotype and genotype of newly diagnosed children. *Diabetes Care*. 2013;36(2):348-354. doi:10.2337/dc12-0445
4. Allen LA, Taylor PN, Gillespie KM, Oram RA, Dayan CM. Maternal type 1 diabetes and relative protection against offspring transmission. *The Lancet Diabetes & Endocrinology*. 2023/10/01/ 2023;11(10):755-767. doi:https://doi.org/10.1016/S2213-8587(23)00190-0
5. Warram JH, Krolewski AS, Gottlieb MS, Kahn CR. Differences in risk of insulin-dependent diabetes in offspring of diabetic mothers and diabetic fathers. *N Engl J Med*. Jul 19 1984;311(3):149-52. doi:10.1056/nejm198407193110304
6. Bleich D, Polak M, Eisenbarth GS, Jackson RA. Decreased Risk of Type I Diabetes in Offspring of Mothers Who Acquire Diabetes During Adrenarchy. *Diabetes*. 1993;42(10):1433-1439. doi:10.2337/diab.42.10.1433
7. Tuomilehto J PT, Tuomilehto-Wolf E, Virtala E. Evidence for importance of gender and birth cohort for risk of IDDM in offspring of IDDM parents. *Diabetologia*. 1995 Aug 1995;38(8):975-82. doi:10.1007/BF00400588
8. Harjutsalo V RA, Tuomilehto J. . Differential transmission of type 1 diabetes from diabetic fathers and mothers to their offspring. *Diabetes*. 2006 May 2006;55(5):1517-24. doi:10.2337/db05-1296
9. The Eurodiab Ace Study Group and The Eurodiab Ace Substudy 2 Study Group. Familial risk of type I diabetes in European children. *Diabetologia*. 1998;41(10):1151-6. Diabetologia 1999 Feb; 42(2):262. doi:10.1007/s001250051044
10. Tillil H, Köbberling J. Age-corrected empirical genetic risk estimates for first-degree relatives of IDDM patients. *Diabetes*. Jan 1987;36(1):93-9. doi:10.2337/diab.36.1.93
11. Dahlquist G BL, Holmgren G, Hägglöf B, Larsson Y, Sterky G, Wall S. The epidemiology of diabetes in Swedish children 0-14 years--a six-year prospective study. *Diabetologia*. 1985 Nov 1985;28(11):802-8. doi:10.1007/BF00291068.
12. Pociot F NK, Hobolth N, Andersen O, Nerup J. A nationwide population-based study of the familial aggregation of type 1 (insulin-dependent) diabetes mellitus in Denmark. Danish Study Group of Diabetes in Childhood. . *Diabetologia*. 1993 Sept 1993;36(9):870-5. doi:10.1007/BF00400364

13. Turtinen M, Härkönen T, Parkkola A, Ilonen J, Knip M. Characteristics of familial type 1 diabetes: effects of the relationship to the affected family member on phenotype and genotype at diagnosis. *Diabetologia*. Nov 2019;62(11):2025-2039. doi:10.1007/s00125-019-4952-8
14. Wei Y, Liu S, Andersson T, Feychting M, Kuja-Halkola R, Carlsson S. Familial aggregation and heritability of childhood-onset and adult-onset type 1 diabetes: a Swedish register-based cohort study. *Lancet Diabetes Endocrinol*. May 2024;12(5):320-329. doi:10.1016/s2213-8587(24)00068-8
15. Pociot F, Nørgaard K, Hobolth N, Andersen O, Nerup J. A nationwide population-based study of the familial aggregation of type 1 (insulin-dependent) diabetes mellitus in Denmark. Danish Study Group of Diabetes in Childhood. *Diabetologia*. Sep 1993;36(9):870-5. doi:10.1007/bf00400364
16. Harjutsalo V, Lammi N, Karvonen M, Groop PH. Age at onset of type 1 diabetes in parents and recurrence risk in offspring. *Diabetes*. Jan 2010;59(1):210-4. doi:10.2337/db09-0344
17. Lorenzen T, Pociot F, Stilgren L, et al. Predictors of IDDM recurrence risk in offspring of Danish IDDM patients. Danish IDDM Epidemiology and Genetics Group. *Diabetologia*. Jun 1998;41(6):666-73. doi:10.1007/s001250050966
18. Gillespie KM, Fareed R, Mortimer GL. Four decades of the Bart's Oxford study: Improved tests to predict type 1 diabetes. *Diabet Med*. Dec 2021;38(12):e14717. doi:10.1111/dme.14717
19. Gardner SG, Bingley PJ, Sawtell PA, Weeks S, Gale EA. Rising incidence of insulin dependent diabetes in children aged under 5 years in the Oxford region: time trend analysis. The Bart's-Oxford Study Group. *Bmj*. Sep 20 1997;315(7110):713-7. doi:10.1136/bmj.315.7110.713
20. Persson M, Becker C, Elding Larsson H, et al. The Better Diabetes Diagnosis (BDD) study – A review of a nationwide prospective cohort study in Sweden. *Diabetes Research and Clinical Practice*. 2018/06/01/ 2018;140:236-244. doi:https://doi.org/10.1016/j.diabres.2018.03.057
21. Onengut-Gumuscu S, Chen WM, Burren O, et al. Fine mapping of type 1 diabetes susceptibility loci and evidence for colocalization of causal variants with lymphoid gene enhancers. *Nat Genet*. Apr 2015;47(4):381-6. doi:10.1038/ng.3245
22. Mahon JL, Sosenko JM, Rafkin-Mervis L, et al. The TrialNet Natural History Study of the Development of Type 1 Diabetes: objectives, design, and initial results. *Pediatric diabetes*. 2009;10(2):97-104.
23. Allen LA TP, Carlsson A, Fraser DP, Hagopian WA, Hedlund E, Hill AV, Jones AG, Ludvigsson J, Mortimer GL, Onengut-Gumuscu S, Redondo MJ, Rich SS, Williams CL, Gillespie KM, Dayan CM, Oram RA. Relative maternal protection against type 1 diabetes: a combined analysis of five observational studies: Supplementary materials. Accessed 24/1/26, https://orca.cardiff.ac.uk/id/eprint/182597[.
24. Allen LA TP, Carlsson A, Fraser DP, Hagopian WA, Hedlund E, Hill AV, Jones AG, Ludvigsson J, Mortimer GL, Onengut-Gumuscu S, Redondo MJ, Rich SS, Williams CL, Gillespie KM, Dayan CM, Oram RA. A study of relative maternal protection against type 1 diabetes in offspring: Statistical analysis plan. Accessed 25/04/2024, 2024. https://orca.cardiff.ac.uk/id/eprint/168151

25. Sharp SA, Rich SS, Wood AR, et al. Development and standardization of an improved type 1 diabetes genetic risk score for use in newborn screening and incident diagnosis. *Diabetes care*. 2019;42(2):200-207.
26. Thomas NJ, Hill AV, Dayan CM, et al. Age of Diagnosis Does Not Alter the Presentation or Progression of Robustly Defined Adult-Onset Type 1 Diabetes. *Diabetes Care*. 2023;46(6):1156-1163. doi:10.2337/dc22-2159
27. Gale EA, Gillespie KM. Diabetes and gender. *Diabetologia*. Jan 2001;44(1):3-15. doi:10.1007/s001250051573
28. Karvonen M, Viik-Kajander M, Moltchanova E, Libman I, LaPorte R, Tuomilehto J. Incidence of childhood type 1 diabetes worldwide. Diabetes Mondiale (DiaMond) Project Group. *Diabetes Care*. 2000;23(10):1516-1526. doi:10.2337/diacare.23.10.1516
29. Karvonen M, Pitkäniemi M, Pitkäniemi J, Kohtamäki K, Tajima N, Tuomilehto J. Sex difference in the incidence of insulin-dependent diabetes mellitus: an analysis of the recent epidemiological data. World Health Organization DIAMOND Project Group. *Diabetes Metab Rev*. Dec 1997;13(4):275-91. doi:10.1002/(sici)1099-0895(199712)13:4<275::aid-dmr197>3.0.co;2-v
30. Nyström L, Dahlquist G, Ostman J, et al. Risk of developing insulin-dependent diabetes mellitus (IDDM) before 35 years of age: indications of climatological determinants for age at onset. *Int J Epidemiol*. Apr 1992;21(2):352-8. doi:10.1093/ije/21.2.352
31. Type 1 Diabetes Genetics Consortium (T1DGC): ImmunoChip Study. 2023.
32. StartRight. StartRight. https://www.diabetesgenes.org/current-research/startright/
33. Allen LA, Cannings-John RL, Evans A, et al. Pregnancy in teenagers diagnosed with type 1 diabetes mellitus in childhood: a national population-based e-cohort study. *Diabetologia*. 2020/04/01 2020;63(4):799-810. doi:10.1007/s00125-019-05063-w
34. Howson JMM, Cooper JD, Smyth DJ, et al. Evidence of Gene-Gene Interaction and Age-at-Diagnosis Effects in Type 1 Diabetes. *Diabetes*. 2012;61(11):3012-3017. doi:10.2337/db11-1694
35. Valdes AM, Erlich HA, Carlson J, Varney M, Moonsamy PV, Noble JA. Use of class I and class II HLA loci for predicting age at onset of type 1 diabetes in multiple populations. *Diabetologia*. 2012/09/01 2012;55(9):2394-2401. doi:10.1007/s00125-012-2608-z
36. Wallace C, Smyth DJ, Maisuria-Armer M, Walker NM, Todd JA, Clayton DG. The imprinted DLK1-MEG3 gene region on chromosome 14q32.2 alters susceptibility to type 1 diabetes. *Nat Genet*. Jan 2010;42(1):68-71. doi:10.1038/ng.493
37. Blunk I, Thomsen H, Reinsch N, et al. Genomic imprinting analyses identify maternal effects as a cause of phenotypic variability in type 1 diabetes and rheumatoid arthritis. *Sci Rep*. Jul 14 2020;10(1):11562. doi:10.1038/s41598-020-68212-x
38. Bonifacio E, Pflüger M, Marienfeld S, Winkler C, Hummel M, Ziegler AG. Maternal type 1 diabetes reduces the risk of islet autoantibodies: relationships with birthweight and maternal HbA(1c). *Diabetologia*. Jul 2008;51(7):1245-52. doi:10.1007/s00125-008-1022-z
39. Paronen J KM, Savilahti E. Effect of cow's milk exposure and maternal type 1 diabetes on cellular and humoral immunization to dietary insulin in infants at genetic risk for type 1 diabetes. *Diabetes*. 2000;49:1657-1665.
40. Koczwara K, Bonifacio E, Ziegler AG. Transmission of maternal islet antibodies and risk of autoimmune diabetes in offspring of mothers with type 1 diabetes. *Diabetes*. Jan 2004;53(1):1-4. doi:10.2337/diabetes.53.1.1

41. Naserke HE, Bonifacio E, Ziegler AG. Prevalence, characteristics and diabetes risk associated with transient maternally acquired islet antibodies and persistent islet antibodies in offspring of parents with type 1 diabetes. *J Clin Endocrinol Metab*. Oct 2001;86(10):4826-33. doi:10.1210/jcem.86.10.7931
42. Miodovnik M, Lavin JP, Knowles HC, Holroyde J, Stys SJ. Spontaneous abortion among insulin-dependent diabetic women. *Am J Obstet Gynecol*. Oct 15 1984;150(4):372-6. doi:10.1016/s0002-9378(84)80141-6
43. Inkster ME, Fahey TP, Donnan PT, Leese GP, Mires GJ, Murphy DJ. Poor glycated haemoglobin control and adverse pregnancy outcomes in type 1 and type 2 diabetes mellitus: systematic review of observational studies. *BMC Pregnancy Childbirth*. Oct 30 2006;6:30. doi:10.1186/1471-2393-6-30
44. Groen B, Links TP, van den Berg PP, de Vos P, Faas MM. The role of autoimmunity in women with type 1 diabetes and adverse pregnancy outcome: A missing link. *Immunobiology*. Mar 2019;224(2):334-338. doi:10.1016/j.imbio.2019.02.003
45. Stene LC GE. The prenatal environment and type 1 diabetes. *Diabetologia*. 10 May 2013 2013;56:1888-1897. doi:10.1007/s00125-013-2929-6
46. Koczwara K BE, Ziegler AG. Transmission of maternal islet antibodies and risk of autoimmune diabetes in offspring of mothers with type 1 diabetes. *Diabetes* 2004;53:1-4.
47. Stefan M, Zhang W, Concepcion E, Yi Z, Tomer Y. DNA methylation profiles in type 1 diabetes twins point to strong epigenetic effects on etiology. *Journal of autoimmunity*. 2014;50:33-37.
48. Johnson RK, Vanderlinden LA, Dong F, et al. Longitudinal DNA methylation differences precede type 1 diabetes. *Sci Rep*. Feb 28 2020;10(1):3721. doi:10.1038/s41598-020-60758-0
49. Knorr S, Skakkebæk A, Just J, et al. Epigenetic and transcriptomic alterations in offspring born to women with type 1 diabetes (the EPICOM study). *BMC Med*. Sep 23 2022;20(1):338. doi:10.1186/s12916-022-02514-x
50. Hidayat K, Zou SY, Shi BM. The influence of maternal body mass index, maternal diabetes mellitus, and maternal smoking during pregnancy on the risk of childhood-onset type 1 diabetes mellitus in the offspring: Systematic review and meta-analysis of observational studies. *Obes Rev*. Aug 2019;20(8):1106-1120. doi:10.1111/obr.12858
51. Sims EK, Geyer S, Johnson SB, et al. Who Is Enrolling? The Path to Monitoring in Type 1 Diabetes TrialNet's Pathway to Prevention. *Diabetes Care*. Dec 2019;42(12):2228-2236. doi:10.2337/dc19-0593

Acknowledgements

Personal thanks: The authors are immensely grateful to the participants, clinicians and research groups who have contributed data to the each of the studies, without whom this study would not have been possible. The authors also wish to thank Dr Adrian Sayers, University of Bristol, for providing additional statistical advice and assistance.

Funding and Assistance: This research was funded by the Wellcome Trust (Ref: 225446/Z/22/Z). The study funders were not involved in the design of the study the collection, analysis, and interpretation of data; writing the report; nor its preparation for publication. For the purpose of Open Access, the author has applied a CC BY public copyright license to any Author Accepted Manuscript version arising from this submission.

The BOX Study Group is comprised of the study team based at the University of Bristol including Mrs Isabel Wilson, Mrs Rachel Aitken, Ms Ilana Kelland and Mrs Clare Megson; Local consultants in the BOX region are Drs Chitrabhanu Ballav, Atanu Dutta, and Michelle Russell-Taylor, Bucks Healthcare Trust, UK; Dr Rachel Besser, Oxford University Hospitals Trust UK, UK; Drs James Bursell and Shanthi Chandran, Milton Keynes University Hospital, UK, Dr Sejal Patel, Wexham Park Hospital, UK; Drs Anne Smith and Manohara Kenchaiah, Northampton General

Hospital, UK; Dr Gomathi Margabanthu, Kettering General Hospital, UK; Drs Foteini Kavvoura and Chandan Yaliwal, Royal Berkshire Hospital, UK.

The BDD Study is funded by The Swedish Child Diabetes Foundation. BDD is comprised of a national study group, Ass.Prof. Annelie Carlsson, Prof. Helena Elding Larsson and Prof Åke Lernmark, Department of Clinical Sciences, Lund University, Ass.Prof. Auste Pundziute Lyckå, and Ass.Prof. Gun Forsander, Department of Paediatrics, Institute for Clinical Science, Sahlgrenska Academy, University of Gothenburg, MD Anna EK, Ass.Prof. Martina Persson, Department of Medicine, Clinical Epidemiology, KI, Prof. Claude Marcus, KI, Prof Johnny Ludvigsson and Ass. Prof. Karin Åkesson, Department of Biomedical and Clinical Science, Linköping University, Linköping, Sweden. BMA Qefsere Brahim and Marlena Maziarz have worked with the BDD-data set.

The Type 1 Diabetes TrialNet Study Group is a clinical trials network currently funded by the National Institutes of Health (NIH) through the National Institute of Diabetes and Digestive and Kidney Diseases, the National Institute of Allergy and Infectious Diseases, and The Eunice Kennedy Shriver National Institute of Child Health and Human Development, through the cooperative agreements U01 DK061010, U01 DK061034, U01 DK061042, U01 DK061058, U01 DK085461, U01 DK085465, U01 DK085466, U01 DK085476, U01 DK085499, U01 DK085509, U01 DK103180, U01 DK103153, U01 DK103266, U01 DK103282, U01 DK106984, U01 DK106994, U01 DK107013, U01 DK107014, U01 DK106993, UC4 DK117009, and the JDRF. NIH NIDDK

R01DK121843 funded MJR, SOG, SSR and RAO, as well as genotyping, imputation, and genetic score calculations for TrialNet samples.

The Type 1 Diabetes Genetics Consortium, is a collaborative clinical study sponsored by the National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK), National Institute of Allergy and Infectious Diseases (NIAID), National Human Genome Research Institute (NHGRI), National Institute of Child Health and Human Development (NICHD), and Juvenile Diabetes Research Foundation International (JDRF) and supported by U01 DK062418.

The StartRight study was funded by Diabetes UK (17/0005624) and the National Institute for Health and Care Research (NIHR; CS-2015-15-018). Genotyping for generation of the T1D GRS was supported by the European Foundation for the Study of Diabetes (2016 Rising Star Fellowship). The study was supported by the NIHR Exeter Biomedical Research Centre and NIHR Exeter Clinical Research Facility. . The StartRight Study Group is led & coordinated by the study team in Exeter, comprising Prof Angus Jones, Mrs Anita Hill, Prof Timothy McDonald, Prof Andrew Hattersley, Prof Beverley Shields, Mr Robert Bolt, Mr Peter Tippett and Dr Lucy Gates. Full details of the local recruiting sites are listed at https://www.diabetesgenes.org/current-research/startright/startright-recruiting-sites/

Author contribution statements:

LAA, PNT, KMG, CMD, RAO developed the original idea for this work. LAA wrote the statistical analysis plan, which was reviewed and approved by all authors. WAH and RAO genotyped BOX and BDD samples and DPF generated their T1D-GRS2 scores. CLW and GLM conducted laboratory work, prepared samples, and provided data from the BOX study. LAA carried out statistical analyses as per the pre specified plan. PNT reviewed the data and statistical analyses and assisted in the development of figures. All authors discussed the results and their significance. LAA wrote the manuscript. All authors reviewed the initial draft, provided critical feedback, and contributed to the final manuscript. PNT, CMD, KMG, RAO provided overall supervision of the project.

LAA is the guarantor of this work and, as such, had full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis. PNT reviewed all data and statistical analyses.

Table 1: Summary of the studies from which participants were included

Descriptions of the individual studies contributing to our analyses are provided.

Figure 1: Meta-analysis of odds of having a mother with type 1 diabetes compared with the odds of having a father with type 1 diabetes amongst individuals with type 1 diabetes.

The number and proportion (%) of individuals with affected mothers and fathers are shown for each individual study. Unadjusted odds ratios are shown for the odds of individuals diagnosed with type 1 diabetes having an affected mother versus father. The odds ratios for each individual study are shown, as well as the overall odds ratio as derived from a random effects meta-analysis. p<0·0001. *Abbreviations: 95%CI; 95% confidence intervals, T1D; Type 1 diabetes*

Figure 2: Meta-analyses of odds of having a mother with type 1 diabetes compared with a father with type 1 diabetes in individuals with type 1 diabetes, divided according to age at diagnosis

2A: Individuals diagnosed with type 1 diabetes ≤18years of age

The number and proportion (%) of individuals with affected mothers and fathers are shown for individuals diagnosed with type 1 diabetes at or before the age of 18 years. Unadjusted odds ratios are shown for the odds of these individuals having an affected mother versus father. The odds ratios for each individual study are shown, as well as the overall odds ratio as derived from a random effects meta-analysis. p<0·0001. StartRight contributed no mothers and just one father to this analysis, and therefore was dropped from the analysis. As expected, the overall

result was the same regardless of whether StartRight was included (OR 0.55, 95% CI 0.47, 0.64 when included).

2B: Individuals diagnosed with type 1 diabetes >18 years of age

The number and proportion (%) of individuals with affected mothers and fathers are shown for individuals diagnosed with type 1 diabetes after age 18 years. Unadjusted odds ratios are shown for the odds of these individuals having an affected mother vs father. The odds ratios for each individual study are shown, as well as the overall odds ratio as derived from a random effects meta-analysis. p=0.01.

Abbreviations: 95%CI; 95% confidence intervals, T1D; Type 1 diabetes

Missing data regarding age at diagnosis: BOX n=1, BDD n=74, TrialNet PTP n=0, T1DGC n=11, StartRight n=0 (0.7% of all cases in study affected by missing data).

Figure 3: Comparison of age at onset of type 1 diabetes between individuals with mothers and fathers with type 1 diabetes

Hedges's g standardised mean difference in age at diagnosis (in years) between individuals with type 1 diabetes and an affected mother compared with father. The results are shown for the

individual studies, and an overall estimate of effect as derived from random effects meta-analysis. Complete case analysis was performed. p=0·45

Abbreviations: 95%CI; 95% confidence intervals, T1D; Type 1 diabetes

Missing data regarding age at onset amongst individuals with parents with type 1 diabetes: BOX n=0, BDD n=8, TrialNet PTP n=0, T1DGC n=2, StartRight n=0 (total n=10, 0.8%).

Figure 4: Meta-analyses of odds of having a mother with type 1 diabetes compared with a father with type 1 diabetes in individuals with type 1 diabetes, divided into subgroups according to whether parental diagnosis was made prior to or after the birth of the offspring

4A: Parental diagnosis prior to the birth of the offspring

The number and proportion (%) of individuals with affected mothers and fathers are shown for when analysis is confined to individuals with parents diagnosed before offspring birth. Unadjusted odds ratios shown for the odds of individuals with type 1 diabetes having an affected mother vs father. The odds ratios for each individual study are shown, as well as the overall odds ratio as derived from a random effects meta-analysis. These data were only available from BOX and TrialNet PTP. p<0·001. *Abbreviations: 95%CI; 95% confidence intervals, T1D; Type 1 diabetes*

4B: Parental diagnosis after the birth of the offspring

The number and proportion (as % of those at risk i.e. excluding those with a mother/father diagnosed prior to their birth as appropriate) of individuals with affected mothers and fathers are shown for when analysis is confined to individuals with parents diagnosed after offspring birth. Unadjusted odds ratios are shown for the odds of individuals with type 1 diabetes having an affected father vs mother. The odds ratios for each individual study are shown, as well as the overall odds ratio as derived from a random effects meta-analysis. These data were only available from BOX and TrialNet PTP. p=0·87. *Abbreviations: 95%CI; 95% confidence intervals, T1D; Type 1 diabetes*

Missing data regarding timing of parental diagnosis: BOX fathers n=0, mothers n=3, TrialNet PTP fathers n=7, mothers n=8).

Table 2: Comparison of proportion of individuals with an affected mother versus father, divided into subgroups according to whether parental diagnosis was made prior to or after the birth of the offspring

Data regarding timing of parental diagnosis were only available in BOX and TrialNet PTP. For these studies, the number and proportion (%) of individuals with an affected mother versus father are shown according to whether the parents were diagnosed before or after offspring

birth. Data regarding the timing of diabetes diagnosis were not available for 3 mothers in BOX, and for 8 mothers and 7 fathers in TrialNet PTP.

Figure 5: Comparison of Type 1 diabetes genetic risk score (T1D-GRS2) between individuals with mothers and fathers with type 1 diabetes

Hedges's g standardised mean difference in type 1 diabetes genetic risk score (T1D-GRS2) for individuals with an affected mother compared with an affected father. The results are shown for the individual studies, alongside an overall estimate of effect as derived from random effects meta-analysis. Complete case analysis was performed. p=0·25.

Abbreviations: 95%CI; 95% confidence intervals, T1D; Type 1 diabetes

Missing data regarding T1D-GRS2 amongst individuals with parents with type 1 diabetes: BOX n=134, BDD n=0, TrialNet PTP n=0, T1DGC n=0, StartRight n=2 (total n=136, 11%).

1 Figures and Tables

2 Table 1: Summary of the studies from which participants were included

3

Study	Number of participants meeting eligibility criteria for inclusion in this study	Age at diagnosis of type 1 diabetes	Recruitment Area	Recruitment period	Type 1 diabetes at enrolment?
BOX[18,19]	3,040	<21 years Median 10 years (IQR 6, 13 years)	Oxfordshire Health Authority Region, UK	Since 1985	Yes (newly diagnosed). Diagnosis of type 1 diabetes based on WHO criteria and clinical requirement for insulin treatment.
BDD[20]	3,896	<18 years Median 10 years (IQR 6, 14 years)	Sweden (nationwide)	Since 2005	Yes (newly diagnosed) since 2005. Since 2011 estimated to have captured over 95% of new diagnoses of type 1 diabetes under age 18. To establish diabetes classification, the study included analyses of HLA-DQ genotypes, autoantibodies and levels of c-peptide at first recruitment.

TrialNet PTP[22,51]	1,316	0-55 years Median 12 years (IQR 8, 16 years)	International, multi-centre Participating countries include the United States of America, Canada, United Kingdom, Germany, Italy, Sweden, Finland, Australia and New Zealand	Since 2000	No. Recruitment of first-degree relatives of individuals with type 1 diabetes found to be positive for at least one type 1 diabetes associated autoantibody. Progression to type 1 diabetes (on the basis of the results of oral glucose tolerance test (OGTT), HbA1c level and antibody testing results) was recorded.
T1DGC 'Families Dataset' [21].	2,662	0-32 years Median 7 years (IQR 4, 12 years)	International, multi centre. Data collected across 4 regional networks: Asia-Pacific (participating countries Australia, India, Malaysia, New Zealand, Philippines, Singapore, Thailand) Europe (Austria, Belgium, Cameroon, Czech Republic, Denmark, Estonia, Finland, Germany, Greece, Hungary, Israel, Italy, Latvia, Lithuania, Netherlands, Poland, Portugal, Romania, Russia, Slovenia, Spain, Sweden, Switzerland, Turkey) North America (57 data collection sites in the United States of America) United Kingdom Network (48 data collection sites in the United Kingdom)	2004-2009	Yes. Recruitment of predominantly affected sibling pairs and a smaller group of parent-child trios to generate a "families dataset" within the T1DGC.

StartRight[26,32].	561	17-88 years Median 35 years (IQR 27, 49 years)	55 UK sites	2015-2020	Recruited participants with newly diagnosed diabetes (<12 months' duration). Type of diabetes recorded and validated using autoantibody testing and clinical information.

1

2 Descriptions of the individual studies contributing to our analyses are provided.

3

Figure 1: Meta-analysis of odds of having a mother with type 1 diabetes compared with the odds of having a father with type 1 diabetes amongst individuals with type 1 diabetes.

The number and proportion (%) of individuals with affected mothers and fathers are shown for each individual study. Unadjusted odds ratios are shown for the odds of individuals diagnosed with type 1 diabetes having an affected mother versus father. The odds ratios for each individual study are shown, as well as the overall odds ratio as derived from a random effects meta-analysis. p<0·0001. *Abbreviations: 95%CI; 95% confidence intervals, T1D; Type 1 diabetes*

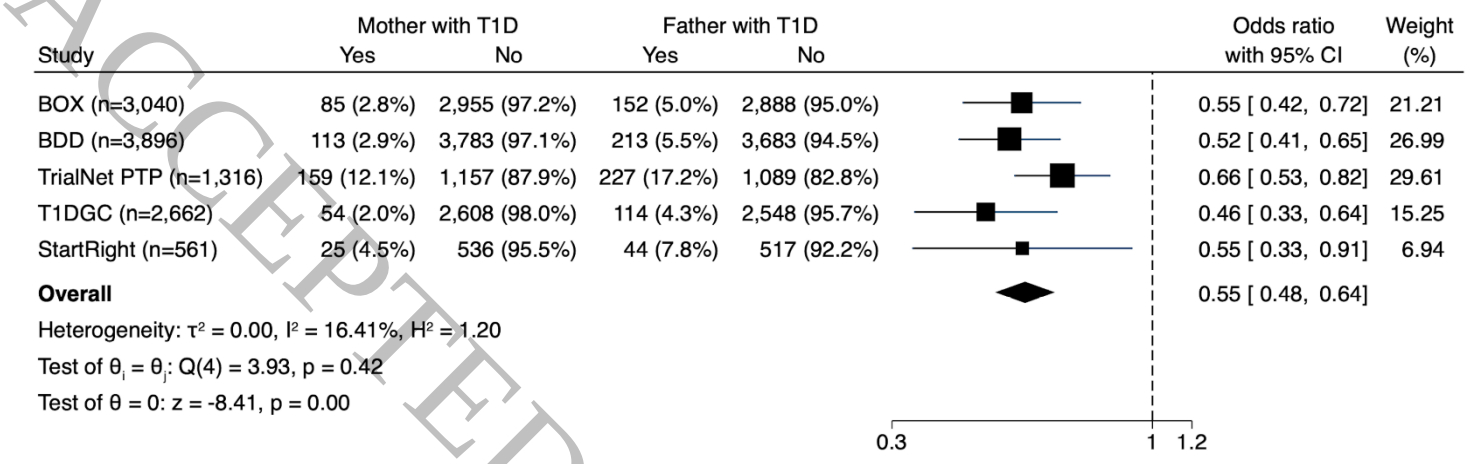


Figure 2: Meta-analyses of odds of having a mother with type 1 diabetes compared with a father with type 1 diabetes in individuals with type 1 diabetes, divided according to age at diagnosis

2A: Individuals diagnosed with type 1 diabetes ≤18years of age

The number and proportion (%) of individuals with affected mothers and fathers are shown for individuals diagnosed with type 1 diabetes at or before the age of 18 years. Unadjusted odds ratios are shown for the odds of these individuals having an affected mother versus father. The odds ratios for each individual study are shown, as well as the overall odds ratio as derived from a random effects meta-analysis. p<0·0001. StartRight contributed no mothers and just one father to this analysis, and therefore was dropped from the analysis. As expected, the overall result was the same regardless of whether StartRight was included (OR 0.55, 95% CI 0.47, 0.64).

2B: Individuals diagnosed with type 1 diabetes >18 years of age

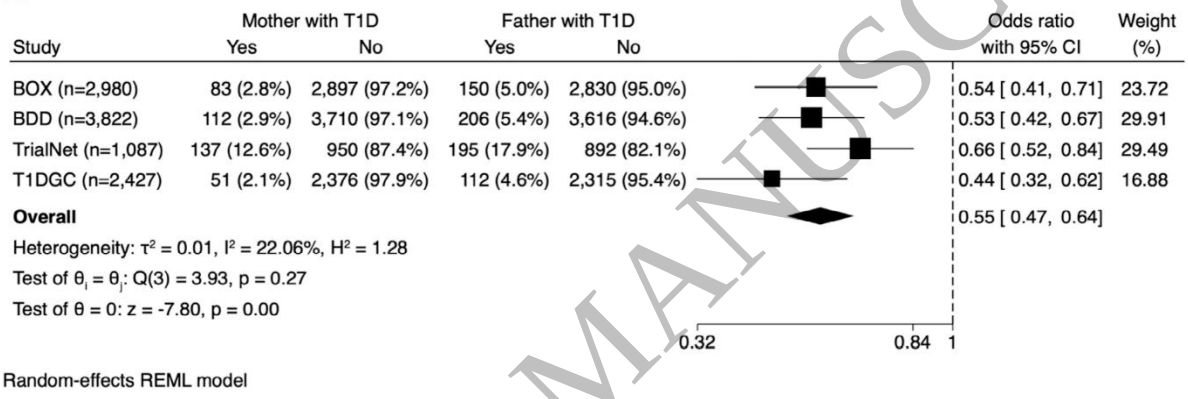
The number and proportion (%) of individuals with affected mothers and fathers are shown for individuals diagnosed with type 1 diabetes after age 18 years. Unadjusted odds ratios are shown for the odds of these individuals having an affected mother vs father. The odds ratios for

each individual study are shown, as well as the overall odds ratio as derived from a random effects meta-analysis. p=0.01.

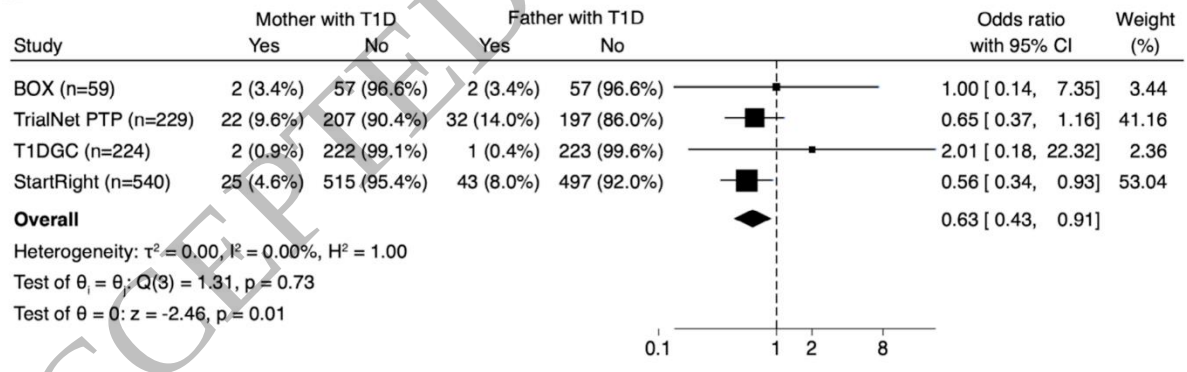
Abbreviations: 95%CI; 95% confidence intervals, T1D; Type 1 diabetes

Missing data regarding age at diagnosis: BOX n=1, BDD n=74, TrialNet PTP n=0, T1DGC n=11, StartRight n=0 (0.7% of all cases in study affected by missing data).

2A

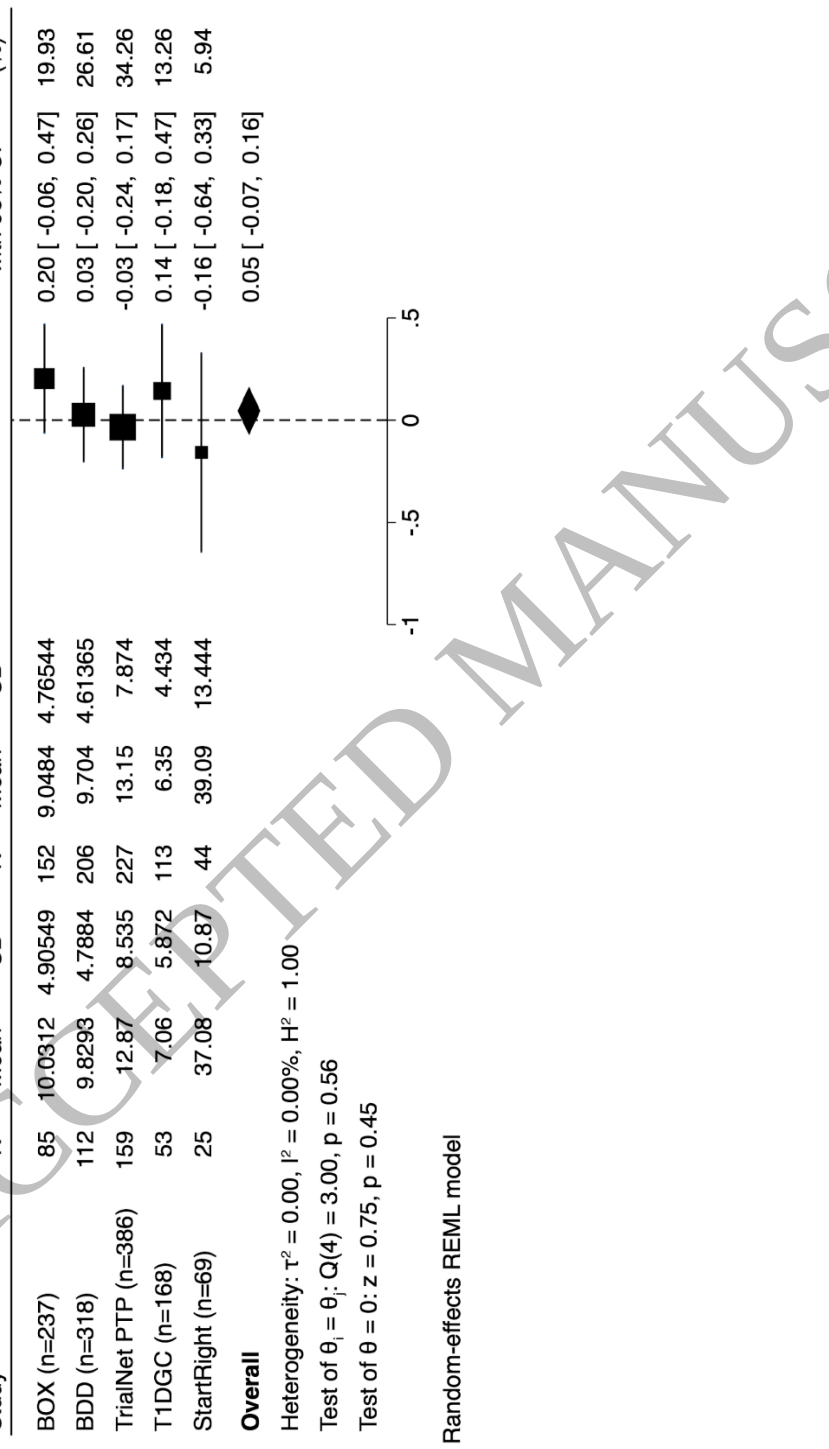


2B



1 Figure 3: Comparison of age at onset of type 1 diabetes between individuals with mothers and

2 fathers with type 1 diabetes



3

Hedges's g standardised mean difference in age at diagnosis (in years) between individuals with type 1 diabetes and an affected mother compared with father. The results are shown for the individual studies, and an overall estimate of effect as derived from random effects meta-analysis. Complete case analysis was performed. p=0·45

Abbreviations: 95%CI; 95% confidence intervals, T1D; Type 1 diabetes

Missing data regarding age at onset amongst individuals with parents with type 1 diabetes: BOX n=0, BDD n=8, TrialNet PTP n=0, T1DGC n=2, StartRight n=0 (total n=10, 0.8%).

Figure 4: Meta-analyses of odds of having a mother with type 1 diabetes compared with a father with type 1 diabetes in individuals with type 1 diabetes, divided into subgroups according to whether parental diagnosis was made prior to or after the birth of the offspring

4A: Parental diagnosis prior to the birth of the offspring

The number and proportion (%) of individuals with affected mothers and fathers are shown for when analysis is confined to individuals with parents diagnosed before offspring birth. Unadjusted odds ratios shown for the odds of individuals with type 1 diabetes having an affected mother vs father. The odds ratios for each individual study are shown, as well as the overall odds ratio as derived from a random effects meta-analysis. These data were only available from BOX and TrialNet PTP. p<0·001. *Abbreviations: 95%CI; 95% confidence intervals, T1D; Type 1 diabetes*

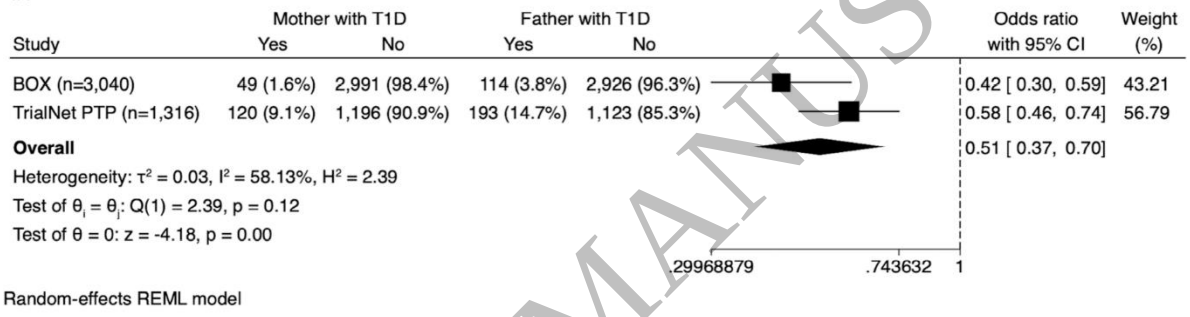
4B: Parental diagnosis after the birth of the offspring

The number and proportion (as % of those at risk i.e. excluding those with a mother/father diagnosed prior to their birth as appropriate) of individuals with affected mothers and fathers are shown for when analysis is confined to individuals with parents diagnosed after offspring birth. Unadjusted odds ratios are shown for the odds of individuals with type 1 diabetes having

an affected father vs mother. The odds ratios for each individual study are shown, as well as the overall odds ratio as derived from a random effects meta-analysis. These data were only available from BOX and TrialNet PTP. p=0·87. *Abbreviations: 95%CI; 95% confidence intervals, T1D; Type 1 diabetes*

Missing data regarding timing of parental diagnosis: BOX fathers n=0, mothers n=3, TrialNet PTP fathers n=7, mothers n=8).

4A



4B

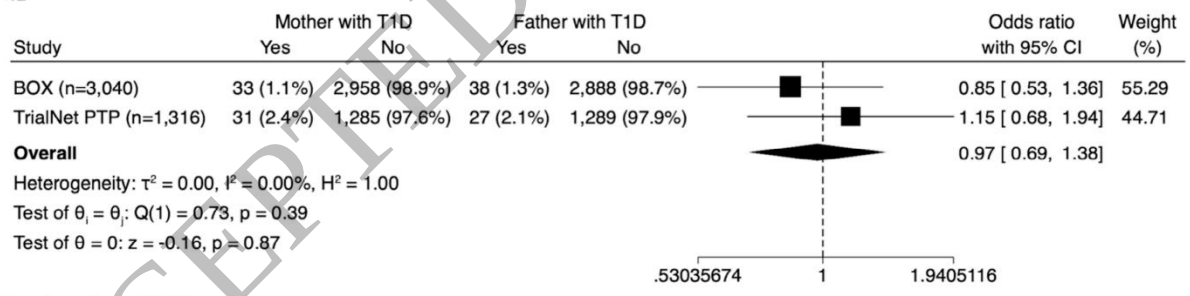


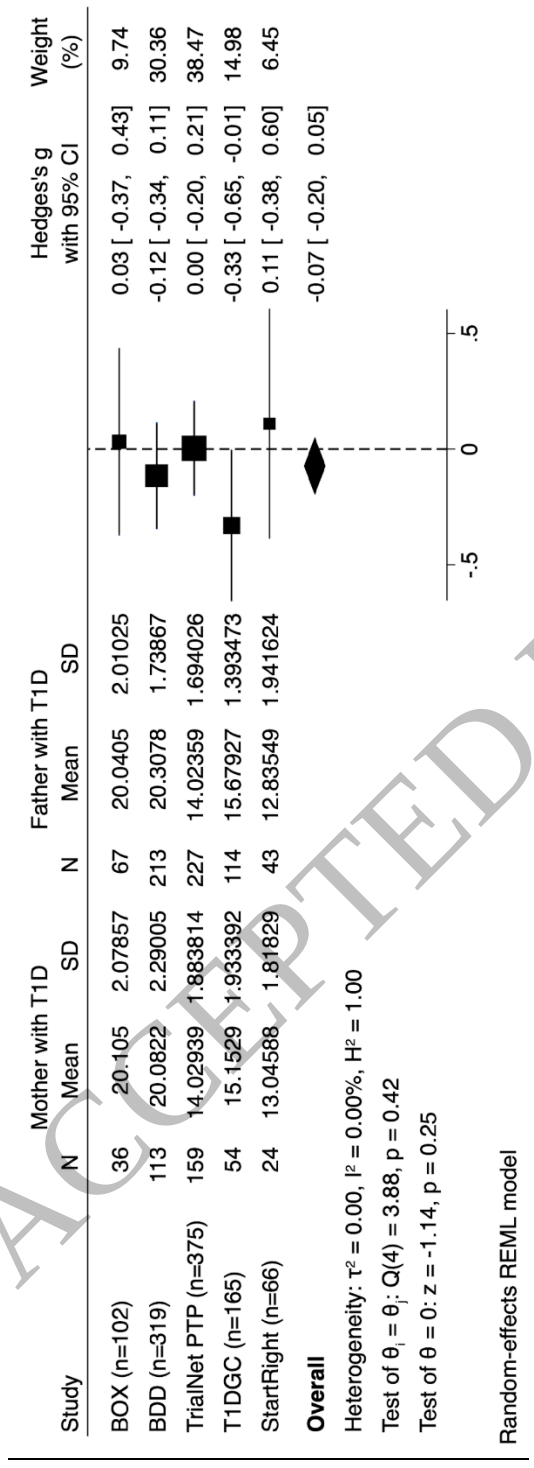
Table 2: Comparison of proportion of individuals with an affected mother versus father, divided into subgroups according to whether parental diagnosis was made prior to or after the birth of the offspring

Study	Number (%) of individuals with mother diagnosed prior to birth	Number (%) of individuals with father diagnosed prior to their birth	Odds ratio (95% CI)	p value	Number (%) of individuals with mother diagnosed after their birth	Number (%) of individuals with father diagnosed after their birth	Odds ratio (95% CI)	p value
BOX (n=3040)	49 (1·6%)	114 (3·8%)	0.42 (0.30, 0.59)	<0·0001	33 (1·1%)	38 (1·3%)	0.85 (0.53, 1.36)	0·49
TrialNet PTP (n=1316)	120 (9.1%)	193 (14.7%)	0.58 (0.46, 0.74)	0·0003	31 (2.4%)	27 (2.1%)	1.15 (0.68, 1.94)	0·14
Pooled estimate as derived from random effects meta-analysis			0.51 (0.37, 0.70)	0·001			0.97 (0.69, 1.38)	0·12

Data regarding timing of parental diagnosis were only available in BOX and TrialNet PTP. For these studies, the number and proportion (%) of individuals with an affected mother versus father are shown according to whether the parents were diagnosed before or after offspring birth. Data regarding the timing of diabetes diagnosis were not available for 3 mothers in BOX, and for 8 mothers and 7 fathers in TrialNet PTP.

1 Figure 5: Comparison of Type 1 diabetes genetic risk score (T1D-GRS2) between individuals with

2 mothers and fathers with type 1 diabetes



3

Hedges's g standardised mean difference in type 1 diabetes genetic risk score (T1D-GRS2) for individuals with an affected mother compared with an affected father. The results are shown for the individual studies, alongside an overall estimate of effect as derived from random effects meta-analysis. Complete case analysis was performed. p=0·25.

Abbreviations: 95%CI; 95% confidence intervals, T1D; Type 1 diabetes

Missing data regarding T1D-GRS2 amongst individuals with parents with type 1 diabetes: BOX n=134, BDD n=0, TrialNet PTP n=0, T1DGC n=0, StartRight n=2 (total n=136, 11%).