StartRight: Getting the right classification and treatment from diagnosis in adults with diabetes.

SHORT STUDY TITLE : StartRight

PROTOCOL VERSION NUMBER AND DATE

Version 4.0 28 AUGUST 2018

RESEARCH REFERENCE NUMBERS

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Sponsors Number: 1703396

Funder Number: CS-2015-15-018 (NIHR)
17/0005624 (DUK)

This protocol has regard for the HRA guidance.

SIGNATURE PAGE

The undersigned confirm that the following protocol has been agreed and accepted and that the Chief Investigator agrees to conduct the study in compliance with the approved protocol and will adhere to the principles of GCP, the Sponsor's SOPs, and other regulatory requirements as amended.

I agree to ensure that the confidential information contained in this document will not be used for any other purpose other than the evaluation or conduct of the clinical investigation without the prior written consent of the Sponsor

I also confirm that I will make the findings of the study publically available through publication or other dissemination tools without any unnecessary delay and that an honest accurate and transparent account of the study will be given; and that any discrepancies from the study as planned in this protocol will be explained.

For and on behalf of the Study Sponsor:

Signature: Date:
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Name (please print):

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Position:
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Chief Investigator:

Signature: Date:
..../...../.....

Name: (please print):

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Statistician:

Signature: Date:
..../...../.....

Name: (please print):

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Position:
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STUDY SUMMARY

StudyTitle	STARTRIGHT: Getting the right classification and treatment from diagnosis in adults with diabetes
Internal ref. no. (or short title)	CRF243 (StartRight Study)
Study Design	Prospective observational
Study Participants	Adults who have been diagnosed with diabetes (excluding gestational or secondary diabetes) for ≤ 1 year at the time of study recruitment. Aged ≥ 18 at the time of diabetes diagnosis.
Planned Sample Size	Main cohort: 1000 Additional late onset diabetes cohort: 800
Follow up duration	3 years
Planned Study Period	6 years
Study Summary	This study aims to achieve more accurate early classification of diabetes and identification of which patients will rapidly require insulin treatment. We will recruit 1200 participants who have been diagnosed with diabetes in the last year and were aged between 18 and 50 years at the time of diagnosis. We will recruit an additional cohort of 800 participants diagnosed after age 50. We will record clinical features and biomarkers that may help us to determine diabetes type at diagnosis and follow participants for 3 years to assess the development of severe insulin deficiency (measured using C-peptide) and insulin requirement. We will assess utility of clinical features and additional biomarkers in identifying patients with rapid progression to insulin requirement. Findings will be integrated into a freely available clinical prediction model.

FUNDING AND SUPPORT IN KIND

FUNDER(S)	**FINANCIAL AND NON FINANCIAL SUPPORT GIVEN**
National Institute for Health Research (NIHR Clinician Scientist award CS-2015-15-018): Integrating clinical features and biomarkers to improve diabetes classification and treatment in young adults. (≤ age 50 cohort) Diabetes UK (17/0005624) Combining Clinical Features and biomarkers to identify patients with Type 1 diabetes in later life (> age 50 cohort)	Financial: NIHR £866,655, Diabetes UK £366,766
NIHR Exeter Clinical Research Facility and NIHR Clinical Research Network	Support Infrastructure
Research & Development Directorate, Royal Devon & Exeter NHS Foundation Trust	Study Sponsorship and Research Governance Support

ROLES & RESPONSIBILITIES:

SPONSOR:
The sponsor has no role in the study design or data analysis, interpretation or manuscript writing.
The sponsor will review and approve the study protocol and supporting documents.
The sponsor takes overall responsibility for the study site monitoring that is carried out by the CI team.

STEERING GROUP:

An independent steering committee will be appointed, made up of researchers, physicians and patients, to provide independent guidance and ensure that study progress and the conduct of the study is appropriate.
The steering committee will meet 6 monthly in the first year of the study, and annually thereafter, with the CI and members of the study team.

STUDY MANAGEMENT:
Day to day management of the study will be undertaken by the CI and study coordinator with the support of the NIHR Exeter Clinical Research Facility.

STUDY PROCEDURE SUMMARY FLOW CHART

NEWLY DIAGNOSED DIABETES PATIENTS IDENTIFIED IN PRIMARY OR SECONDARY CARE & SCREENED FOR ELIGIBILITY

- Diagnosed with Diabetes in past 12 months
- Aged 18 or over at the time of diabetes diagnosis

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BASELINE RESEARCH VISIT & CONSENT (approx 30-45mins)

- Blood & urine samples, clinical characteristics, treatment details, demographics & questionnaire
- Notes review by recruiting nurse to collect/confirm diagnosis details.
- Samples analysed for c-peptide & islet autoantibodies (GAD, IA2 & ZNT8), DNA, serum and plasma bio banked
- Clinician & participant informed of autoantibody results

↓

Follow Up 1 Year Post Recruitment

- Contact (by phone,email, post or in person) to collect details of treatment change & hypoglycaemia
- Patient sends urine or blood sample by post to Exeter for UCPCR or serum C-peptide..
- Research team collects HbA1c results from GP practice or hospital records.

↓

Follow Up 2 Year Post Recruitment

- Contact (by phone.email, post or in person) to collect details of treatment change & hypoglycaemia.
- Patient sends urine or blood sample by post to Exeter for UCPCR or serum C-peptide
- Research team collects HbA1c results from GP practice or hospital records.

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FINAL VISIT - 3 YEARS POST RECRUITMENT

Repeat blood & urine samples, clinical characteristics, treatment details & questionnaire data collected at Visit 1.

STUDY PROTOCOL

StartRight: Getting the right classification and treatment from diagnosis in adults with diabetes.

1 Lay summary

The treatment of Type 1 and Type 2 diabetes is very different. People with Type 1 diabetes rapidly stop making their own insulin, so need insulin injections from diagnosis. People with Type 2 diabetes can keep making their own insulin but it may not work as well as it should, so they can be treated with diet or tablets. While they may eventually need insulin treatment it is usually not until many years after diagnosis.

It is often difficult for doctors to tell which kind of diabetes a person has. Because of this, sometimes people are given the wrong diagnosis. This can have a huge impact as it means they could receive the wrong treatment. A person incorrectly diagnosed with Type 1 diabetes will be prescribed unnecessary insulin injections and miss out on other helpful therapies. A person incorrectly diagnosed with Type 2 diabetes may develop severely high glucose and become unwell with a condition called Diabetic Ketoacidosis if they do not receive insulin treatment.

This study aims to improve this situation by helping doctors more accurately tell the type of diabetes a person has when they are first diagnosed. We will recruit 1000 participants who have recently been diagnosed with diabetes between the ages of 18 and 50. We will recruit an additional 800 recently diagnosed participants who developed diabetes after age 50, half of whom will be receiving insulin therapy. We will record clinical features and measure blood tests that may help us determine diabetes type at diagnosis and follow participants for 3 years to see whether they stop producing their own insulin and need insulin treatment, which confirms Type 1 diabetes. We will assess whether clinical features and blood tests can help us tell if a patient needs rapid insulin treatment and should be initially treated as Type 1 or Type 2 diabetes.

We will combine results from this study and existing previous studies to produce a calculator, called a clinical probability model that will allow doctors and patients to combine information from clinical features and (where necessary) blood tests to accurately diagnose what type of diabetes a person has and therefore give the correct treatment. This will be freely available to doctors and patients as a website calculator and smartphone app.

2 BACKGROUND AND RATIONALE

Correct diagnosis is needed for appropriate management of Type 1 and Type 2 diabetes

Evidence based guidelines for the treatment of Type 1 and Type 2 diabetes are very different (1-4). These differences predominantly relate to the rapid development of severe insulin deficiency in Type 1 diabetes (3, 5). This means patients with type 1 diabetes need early insulin treatment, are at risk of life threatening ketoacidosis without insulin treatment and have poor response to non-insulin therapy(3). They require accurate insulin delivery (e.g. multiple injections, carbohydrate counting, pumps) due to the very high glycaemic variability associated with absolute insulin deficiency (5, 6). In contrast patients with Type 2 diabetes continue to produce substantial endogenous insulin even decades after

diagnosis (7). Glycaemia may therefore be initially managed with lifestyle change and non-insulin therapies, should insulin be required simple insulin regimens are combined with other agents (4, 8). Misdiagnosis will mean a patient receives inappropriate treatment, leading to unnecessary insulin therapy or, where Type 1 diabetes is misdiagnosed as Type 2, poor glycaemic control, potential ketoacidosis and earlier development of diabetes complications (9, 10).

Misclassification of diabetes is common and increasingly difficult in young adults

BMI and age of diagnosis are usually used to differentiate diabetes subtypes however with increasing obesity Type 1 patients may be obese and Type 2 is seen in children and very young adults (11-13). Diagnosis on clinical features is therefore often difficult and initial misclassification is frequent (7-25%), particularly in young adults, an age group where incident Type 1 and Type 2 diabetes are both common (9, 14-16). In a recent study using similar definitions to this project, 26% of those treated with insulin from diagnosis and 23% of Type 2 diabetes diagnosed before age 35 were misclassified (17). . Initial diagnosis is rarely revisited therefore misdiagnosis will have long term implications. A joint report by the Royal College of General Practitioners and NHS diabetes in 2011 highlighted the problems of misclassification of diabetes including a lack of clinical data and its importance from a clinician and patient perspective (9).

Half of older patients receiving insulin from diagnosis have been misdiagnosed

Most patients developing diabetes after age 50 will have type 2 diabetes, therefore most patients (97%) of those treated as type 2 diabetes will be correctly classified. However we have recently shown that 49.5% of those treated as Type 1 diabetes (with insulin from diagnosis) after age 50 have been wrongly classified, as they continue to produce large amounts of their own insulin many years after diagnosis, confirming Type 2 diabetes (Jones et al unpublished, with misclassification 56% in a separate multicentre cohort (17)). This means that >6000 new patients annually in the UK in this age group are misclassified as having T1D, and consequently unnecessarily treated with insulin (18).

Robust evidence and guidelines for clinical classification of diabetes are needed

We have systematically reviewed evidence on diabetes classification and found only 11 papers assessed clinical features against robustly defined diabetes subtypes with only 2 published in the last decade (19). No papers assessed young adult onset diabetes and only one assessed the utility of combining clinical features (20). While developments in assay technology have meant biomarkers such as C-peptide and autoantibodies are more robust and inexpensive there is a lack of evidence to support or refute their use, with the evidence quality for islet autoantibody testing to aid diabetes classification rated as low on expert committee review (21). Large prospective studies of mixed populations are needed to support or refute the use of these tests in clinical care.

Novel genetic biomarkers may improve diabetes classification

The Exeter group have recently developed an inexpensive genetic risk score, based on 34 variants predisposing to Type 1 diabetes, which can discriminate Type 1 and 2 diabetes with AUC ROC 0.88 in cross-sectional datasets (22, 23). The score is independent and additive to autoantibody status and clinical features.

A validated clinical probability model for diabetes classification has the potential to improve clinical care

In many areas of clinical care decision making has been improved by using probability models ('calculators') that give appropriate weight to a multitude of patients features in a way that an individual clinician cannot (24). This approach has improved decision making in a wide range of conditions, for example physicians following NICE guidance for cardiovascular disease or osteoporosis will be familiar with the QRISK, GRACE and FRAX risk models. In response to widespread misdiagnosis of monogenic diabetes (25) the Exeter group has developed a probability model for common types of monogenic diabetes ('MODY') that outperforms standard clinical criteria (C statistic >0.94) (26). The resulting online calculator (http://www.diabetesgenes.org/content/mody-probability-calculator) has received 11900 visits in less than 2 years and is used prior to the majority of referrals for diagnostic genetic testing at the UK MODY referrals laboratory. It has recently been incorporated into an iphone app which aims to bring together current evidence and expert opinion to aid differential diagnosis of diabetes subtypes (Diabetes Diagnostics app). We are developing a model integrating clinical features to diagnosis diabetes subtype (Jones Shields unpublished) using cross sectional datasets which has not been prospectively tested. Simultaneous with the early stages of this study this model will be further developed using cross sectional cohorts in Exeter and Oxford.

3. OBJECTIVES AND OUTCOME MEASURES/ENDPOINTS

Primary objective

To establish diagnostic performance of biomarkers including islet autoantibodies, C-peptide and a genetic risk score in identifying patients with rapid requirement for insulin, alone and in combination with clinical features

Secondary objectives

To prospectively validate a clinical prediction model developed from cross sectional datasets in predicting rapid insulin requirement in young onset diabetes.

To integrate discriminative and additive biomarkers into the clinical prediction model.

To establish a bio resource for future biomarkers discovery and assessment.

Primary endpoint/outcome

Diabetes type defined by insulin requirement at 3 years:

Type 1 diabetes = Progression to insulin treatment and severe insulin deficiency (post meal plasma C-peptide <0.6nmol/L) at 3 years

Type 2 diabetes = Lack of requirement for insulin at 3 years (HbA1c <90mmol/mol without insulin treatment or post meal C-peptide ≥ 0.6nmol/L if insulin treated)

Rationale for primary outcome

A stimulated C-peptide less than 0.6nmol/L reflects severe insulin deficiency and is consistent with Type 1 diabetes outside the 'honeymoon' period and requirement for insulin treatment. A stimulated C-peptide over 0.6nmol/L represents substantial endogenous insulin secretion and is consistent with type 2 diabetes, non-insulin requirement and response to oral therapy, as reviewed in Jones 2013(27). Equivalent thresholds for fasting C-peptide and post meal urine C-peptide creatinine ratio have been derived from a dataset of 120 participants with insulin treated diabetes as previously reported (26). We have included early insulin treatment alongside measured insulin secretion as should an individual develop severe insulin deficiency at a late stage but have achieved glycaemic control without insulin for >3 years after diagnosis we would consider their initial treatment as Type 2 diabetes appropriate.

Where urine and blood C-peptide are discordant, classification will be based on the highest value. A cut off of 0.6nmol/mol will be used for classification based on urine C-peptide creatinine ratio (26).

Secondary endpoints/outcomes

Stimulated C-peptide <0.2nmol/L at 3 years ('absolute insulin deficiency') (27, 28)

C-peptide rate of change (UCPCR and plasma)

HbA1c (mean and at 3 years)

Weight change (baseline to 3 years)

Self-reported hypoglycaemia & hypoglycaemic awareness (29, 30)

Wellbeing and resilience (SF12 and CD-RISC questionnaire) (30, 31)

Ketoacidosis (self-reported and confirmed from medical notes)

This research will produce a robustly validated clinical prediction model that will for the first time integrate validated clinical features and biomarkers to allow clinicians to accurately assess classification and need for insulin treatment at diabetes diagnosis. The model will be freely available to clinicians and patients through a website and smartphone App.

4. STUDY DESIGN

Prospective observational study. We will recruit a prospective cohort of 1200 adults diagnosed with diabetes within the previous 1 year and aged between 18 and 50 at the time of diagnosis.

We will also recruit an additional cohort of 800 participants diagnosed with diabetes in the last year aged >50 at diabetes diagnosis, with recruitment stratified by insulin treatment (400 insulin treated, 400 non insulin treated).

At baseline (visit 1) blood & urine samples will be collected for biomarker assessment and biobanking, and clinical features recorded. We will follow participants for 3 years with annual remote contact (phone, email or letter) and postal urine or capillary blood sample for C-peptide. A further face to face visit with blood & urine sampling will be performed at 3 years.

We will assess the utility of adding biomarkers to clinical features in diagnosing diabetes subtype (defined by insulin requirement at 3 years), and where utility is shown, integrate these into a probability model.

Please also refer to the study summary flow chart on page 7 for a summary of study procedures.

4.1 STUDY SETTING

NHS primary care, secondary care, or community setting.

This is a multicentre study and participants will be recruited through up to 60 recruiting sites in the UK.

Recruiting sites will need to confirm the following capabilities in order to deliver the study:

- Ability to recruit (possibly with the assistance of PICs) at least 30 participants before 31 December 2019
- Sufficient research nurse time to undertake the recruitment and all follow-up procedures as detailed in the study design for the full duration of the study.
- Research nurse(s) must be trained and competent in phlebotomy.
- Ability to spin blood samples (in accordance with SOPs provided by the CI) and despatch on day of collection to the lead site in Exeter , UK.
  Note: Samples transferred to the Royal Devon and Exeter Hospital blood sciences laboratory for analysis will be labelled with participant identifiable data (Name, date of birth & NHS number) in accordance with the requirements for clinical sample analysis. Once initial analysis is complete, the remaining samples will be anonymised and assigned an individual study code prior to storage.
- Undertake to transfer scanned (PDF) copies of data collection forms, consent forms & biochemistry results (via secure nhs.net email) to the CI site within 5 working days of the study visit.

- Full contact details for both patients and clinicians (GP or secondary care) will have to be included in the information transferred to the lead site, to facilitate feedback of antibody results after visit 1 and C-Peptide result at 3 years.

- Undertake to transfer scanned (PDF) copies of recruitment logs and site specific delegation logs to the CI site on a monthly basis to facilitate remote site monitoring requirements.

4.2 ASSESSMENT AND MANAGEMENT OF RISK

Concurrent medication and diabetes management:

This is an observational study therefore all decisions on patient management will continue to be taken by participant's usual clinicians. All diabetes medications will be unaltered.

Blood sampling:

Blood samples will be taken which may result in slight pain and bruising. Samples will be taken by staff trained and experienced in venepuncture procedures.

Patient burden:

Participation involves 2 blood samples (3 years apart) and 2 additional remote contacts (phone, email or letter according to participant preference) over the 3 year period. Participants will also collect a urine sample or capillary blood sample at home on 4 occasions.

Concomitant studies:

Recruitment to this study does not preclude recruitment to concurrent studies related or unrelated to diabetes

Reporting adverse events

This study is categorised as:

• Type A = No higher than the risk of standard medical care

This is a low risk study and it is not anticipated that participants involved in this project will be subject to adverse or serious adverse events (AEs/SAEs).
AEs/SAEs that occur as a result of study participation (e.g. venepuncture during research visit) should be reported to the study sponsor and CI, with 24 hours of notification, in accordance with the SAE/AE SOP and report forms provided to recruiting sites in their study site file. These study related AEs/SAEs should also be reported locally in accordance with individual Trust protocols.

AEs/SAEs that are not a result of study participation should be dealt with at each recruiting site in accordance with their local Trust protocols.

4.3 ELIGIBILITY CRITERIA

4.3.1 Inclusion criteria

- Adults diagnosed with diabetes within the previous 12 months.
- Aged ≥18 and ≤50 at the time of diabetes diagnosis or (additional late onset diabetes cohort) aged >50 at the time of diabetes diagnosis*.

- Able and willing to provide informed consent.

* Clinical diagnosis of diabetes mellitus. Biochemical diagnosis confirmed from healthcare records post recruitment.

4.3.2 Exclusion criteria

- Gestational diabetes
- Known secondary diabetes (diabetes considered likely due to medication, cystic fibrosis, pancreatitis, pancreatic cancer, pancreatic surgery, hemochromatosis or Cushing's syndrome).

5 STUDY PROCEDURES

Please refer to the flow chart on page 7 for a summary of the study procedures.

The study procedures described below will be conducted in accordance with detailed SOPs provided to all sites by the CI.

5.1 Identification & Recruitment

5.1.1 Patient Identification & Eligibility Screening

Eligible participants will be identified in the following ways:

- At routine clinical care appointment (primary and secondary care): Clinicians will introduce the study to eligible patients and obtain verbal consent for the research team to contact the participant to discuss the study in more detail.
- Regular screening of primary and secondary care records: Clinicians will carry out periodic searches of their patient lists to identify eligible patients and invite those patients to take part in the study by letter, email, telephone or in person at the patient's next appointment.
- Retinal Screening: Invitation letter or copy of the study poster (advertisement) sent to new retinal screening participants.
- Self-referral: Study posters (advertisements) will be displayed in primary care, secondary care or community settings. The study may also be promoted on research websites & appropriate social media (e.g. diabetes or research specific Facebook pages) using wording from ethically approved study documents (ie patient information sheet or study poster)
- Identification from existing research databases where participants have given formal written consent for contact about further research.

This is a multicentre study and identification methods will vary by centre, for example it is likely some sites will recruit from only secondary or primary care

5.1.2 Targeted recruitment of participants aged over 50 by insulin treatment status

To ensure sufficient prevalence of the primary study outcome of Type 1 diabetes in those over age 50 recruitment of this age group will target equal numbers (400 each) of participants receiving insulin treatment at the time of recruitment (high prevalence Type 1), and those not receiving insulin treatment (low prevalence Type 1). This will be managed using separate recruitment targets for these subgroups, based on prior recruitment rate, with recruitment to the non insulin treated cohort limited to a subset of study sites.

5.1.3 Screening

A member of the research team will contact referred/identified patients directly to discuss the project in more detail and confirm eligibility. A copy of the project specific participant information sheet (PIS) will be provided. In all cases, potential participants will be given written information and the opportunity to discuss the project with one of the research team prior to recruitment.

5.1.4 Consent Visit (Visit 1) – Duration <1 hour

To facilitate recruitment participants will be offered home and/or evening visits where this can be supported by the recruiting site.

Following written informed consent demographic and clinical features will be recorded, including weight, height, waist/hip ratio & blood pressure. Data will be collected relating to the patient's diabetes, diagnosis and other relevant medical condition and will include a hypoglycaemia questionnaire. Non-fasting (within 1-5 hours of a meal) blood sample will be collected at all sites for

- Serum C-peptide and other routine biochemistry
- Islet autoantibodies (GAD, IA2, ZnT8)
- DNA extraction (T1D genetic risk score typing, biobanking)
- Plasma and serum samples for storage for future analysis

At selected sites, blood sample will also be collected for additional tests including immune function analysis

Total blood sample volume to be collected at visit 1 will be $\leq$ 90ml (volume in sites not undertaking immune function studies $\leq$40ml).

Samples will be transferred to Exeter Clinical Laboratory (Royal Devon and Exeter NHS Foundation Trust) directly by post or frozen for subsequent batched transfer.

Participants will be given sterile collection kits and prepaid packaging for home post meal urine or capillary blood collection. The sample will be posted directly to Exeter Clinical laboratory and will be analysed for Urinary C-Peptide Creatinine Ratio (UCPCR) or serum C-peptide

Samples will be collected and processed in accordance with detailed SOPs.

Islet autoantibody results (with clinical interpretation) will be reported to clinicians and participants within 4 weeks of visit 1.

With specific consent additional data will be collected from clinical and laboratory records including diagnosis weight, HbA1c, FBC, glucose and lipid profile, and confirmation of ketoacidosis.

Participants will be asked to complete an optional 25 question questionnaire assessing psychological wellbeing and resilience (31, 32).

At some sites, participants will be offered the option to gift samples taken during their routine diabetes care to be used to aid the on-going collection of research data (e.g. biochemical analysis for indicators of long-term insulin production).

5.1.5 Follow-Up - Visits 2 & 3

Follow up by telephone,email, post or in person at 1 year (10-16 months) and 2 years (22-28 months)
Duration: <15 minutes

At approximately 1 year and 2 years post recruitment, participants will be contacted to record concurrent treatment, hypoglycaemia and health service utilisation.

Participants will be asked to collect a home post meal urine or capillary blood sample and post to the Exeter Clinical Laboratory for Urinary C-Peptide /Creatinine Ratio (UCPCR) or serum C-peptide

HbA1c results will be obtained from participants GP practice or laboratory records.

5.1.6 Follow Up - Visit 4

Follow up visit at 3 years (34-40 months)
Approximate duration: 30 minutes

Participants will be invited to attend a further face to face non-fasting visit.

Weight, blood pressure, concurrent treatment, hypoglycaemia and health service utilisation will be recorded.

Participants will be asked to complete a further, optional, wellbeing/resilience questionnaire.

A non-fasting (ideally within 1-5 hours of a meal) blood sample will be collected at all sites for

- Serum C-peptide and other routine biochemistry
- Plasma and serum samples for storage for future analysis

At selected sites, blood sample will also be collected for additional tests including immune function analysis.

Total blood sample volume to be collected at visit 4 will be $\leq$ 85ml (sites without immune function tests $\leq$35ml)

5.1.7 Clinical Reporting

With specific consent from the participant, results of assays of clinical relevance (including routine clinical bloods analysed at local site, baseline autoantibodies and C-peptide assessment at 3 years) will be copied to the participant's GP or secondary care clinician with information on test interpretation.

Baseline islet autoantibody results and 3 year C-peptide will be sent to participants, with patient appropriate information on interpretation, from the central research team at the lead site.

6. WITHDRAWAL & END OF STUDY

6.1 Withdrawal criteria

Subjects will be informed that they are free to withdraw from the study at any time up until the samples and data are coded but not anonymised. When samples are fully anonymised, the participants will still be able to withdraw but their samples and associated data will be retained for use in analysis. If a patient permanently withdraws from the study, or is lost to follow-up, the reason will be recorded.

Criteria for discontinuing participation in the study are:

- Participant withdrawal of consent.
- Investigator's discretion that it is in the best interest of the participant to withdraw.
- Termination of the study by the sponsor or funding body.

6.2 End of Study

We will recruit participants over 30-36 months, with 36 months participant follow up (total duration of study = 66-72 months. Individual participant involvement = 36 months). The study will end when the last participant has completed the study. The study may be discontinued at the sponsor's or CI's discretion.

7. SAMPLE PROCESSING

The sample processing procedures described below will be conducted in accordance with detailed SOPs provided to all sites by the CI.

7.1 Sample Transfer & Initial Analysis

Following sample collection, initial analysis may be undertaken locally or centrally, depending on the best logistical method that optimises sample analysis accuracy (this may differ for different biomarkers, some being more stable than others).

At each study visit blood and/or urine samples will be transferred to the Royal Devon & Exeter NHS Foundation Trust Blood Sciences Laboratory for central analysis and biobanking according to detailed sample handling SOPs.

Samples transferred to the Exeter blood sciences laboratory for analysis will be labelled with participant identifiable data (Name, date of birth & NHS number) in accordance with the requirements for clinical sample analysis. Once initial analysis is complete, the remaining samples will be anonymised and assigned an individual study code prior to storage.

DNA will be extracted by the Royal Devon and Exeter Hospital Clinical Genetics Laboratory who will store extracted DNA pending analysis. Blood samples for DNA extraction will be labelled with the study name and unique study code but not patient identifiable data.

7.2 Sample Storage Procedures

All saved serum, plasma and DNA samples will be stored under a unique ID code, with the file linking the code to personal identifiable information held securely by the CI and appropriate local PI and accessible only to personnel with training in data protection who require this information to perform their duties.

7.3 Long Term Storage & Future Analysis

Enduring consent will be sought to store the samples (serum, plasma and DNA) in the Peninsula Research Bank (PRB) following completion of analysis for this study. All future analysis of samples and/or data will be conducted with the specific agreement of the PRB steering committee.

8. DATA COLLECTION AND RECORDING

8.1 Data Collection Form

Data will be obtained and recorded on a paper data collection form (DCF) with all forms returned to the NIHR Exeter Clinical Research Facility and entered onto a specially designed database. Data collected on the DCFs and subsequently entered onto the database will be reviewed for discrepancies, missing data and queries.

8.2 Source Data

CVs, delegations logs, DCFs and questionnaires will be deemed source data.

8.3 Data Monitoring

The coordinating research team will undertake regular audits of raw data for both accuracy and completeness and research data will be cleaned before analysis is undertaken. The CI reserves the right to close recruiting sites who fail to meet data and sample quality standards on a regular basis.

8.4 Data Confidentiality

All participant data will be held in a link-anonymised format, with personal identifiable data only accessible to personnel with training in data protection who require this information to perform their duties. Participants' research and sample data will be identified by unique study ID numbers and all data will be held on password-protected computers. All paper copies of study data will be stored under ID number and kept in locked offices within the research facilities. Researchers involved in data and sample analysis will not have access to personal identifiable data.

8.5 Data Storage and Archiving

All consent and data collection forms will be held in the relevant local research centre, with access to personal identifiable data restricted to personnel with training in data protection who require this information to perform their duties. Where consent and data collection forms are completed at primary/secondary care or home locations, these forms will be sent to the research centre for monitoring and data entry. The coordinating research team will undertake regular audits of raw data for both accuracy and completeness and research data will be cleaned before analysis is undertaken.

At the end of the study, archiving at each recruitment site will be carried out in accordance with local policy.

At the end of the study, following analysis, anonymised data (together with samples) will be transferred to the Peninsula Research Bank for long term storage and management.

9 STATISTICS AND DATA ANALYSIS

9.1 Sample Size

Diagnosis <= age 50 cohort

Assuming conservative estimates that 25% of participants will have Type 1 diabetes (18) and 80% of participants will have the primary outcome measure a sample size of 1000 (<50 cohort) will give confidence intervals of 85-94% and 87-92% around a biomarker sensitivity/specificity of 90% for Type 1 diabetes (33).

Diagnosis >age 50 cohort

We expect 26% of participants diagnosed after age 50 will have Type 1 diabetes using the study definition, based on analysis of study primary outcome in population based cohort (DARE study – prevalence T1D as defined by primary outcome where insulin treated within 12 months of diagnosis 48%, prevalence where non-insulin treated at diagnosis 3.9%). Assuming the primary outcome is available in 90% of participants (higher retention expected in older participants) this will give confidence intervals of 85-94% and 87-92% around a biomarker sensitivity/specificity of 90% for Type 1 diabetes.

9.2 Feasibility

With >200 new cases of diabetes per 100,000 population annually in the <=age 50 age group this sample size represents <6% of expected incidence at each centre for the recruitment period. In the >age 50 cohort incidence of diabetes treated with insulin from diagnosis is 149/100000, and the recruitment target is 5% of expected incidence at recruiting sites. For non insulin treated participants diagnosed with diabetes after age 50 the recruitment target represents <1% of expected incidence at recruiting sites. A 18 month window for recruitment means we should be able to invite a large proportion of eligible participants through regular screening of practice records or hospital records or writing to participants identified through the retinal screening service (all newly diagnosed patients are referred for retinal screening). A pilot study for this project is currently running at the Exeter site and 27 participants have been recruited by identification through routine care and secondary care records in a period of 4 months at this one recruiting site.

9.3 Statistical analysis

Analysis will be pre-registered with clinicaltrials.gov. Both analysis and reporting will conform to STARD and TRIPOD (34, 35). We will assess the discriminative ability of the clinical probability model for early insulin requirement and perform updating if necessary. We will identify discriminatory biomarkers (alone and in combination with clinical features) using logistic regression and generate updated models integrating both clinical features and discriminatory biomarkers (26). Where appropriate covariates will be log transformed, or if necessary recoded into categories. Independent discriminators will be combined using backward stepwise approaches to enter explanatory variables (using predefined candidate co-variates as below), with the combination of variables assessed by the Deviance statistic. Performance of the resulting models will be assessed using Receiver Operating Characteristic (ROC) analysis. Incremental benefit of additional characteristics in nested models will

be assessed using Net Reclassification Improvement and internal validation using bootstrapping and assess goodness of fit. Analysis will be performed separately for participants diagnosed before and after age 50. Model predictions for the >age 50 cohort will be adjusted to reflect population incidence of Type 1 diabetes in this age group using data from Clinical Practice Research Datalink.

Candidate clinical features and biomarkers which will be assessed and where appropriate integrated into prediction models include:

Model development on cross sectional data	Additional features assessed in prospective study only
Age at diagnosis BMI Waist hip ratio Ethnicity Gender Family history diabetes (1st degree relative) Diagnosis HbA1c, glucose, lipid profile	Symptoms at diagnosis (thirst, polyuria, weight loss (patient reported)) Ketosis (hospital diagnosis DKA, hospital/GP ketone level (urine dip/ capillary) where tested) Acanthosis Personal and family history of autoimmune disease (thyroid, IBD, coeliac, pernicious anaemia, vitiligo, Addison's, rheumatoid arthritis) Hypertension C-peptide & islet autoantibodies (GAD, IA2, ZnT8), Type 1 diabetes genetic risk score (T1DGRS), CRP

10. Study Management:

The study will be managed by the NIHR Exeter Clinical Research Facility providing infrastructure support and facilitate recruitment and sample collection. Study management will be overseen by a steering group which will include external academic and lay members.

11. Monitoring & Indemnity

Monitoring of this study will ensure compliance with Good Clinical Practice. The Investigator(s) will permit monitoring, audits, REC review, and regulatory inspections by providing the Sponsor(s), Regulators and REC direct access to source data and other documents (eg patients' data collection forms, consent forms etc. Periodic remote monitoring will be carried out by the CI study team and the sponsor. Consent will be taken to permit access to data by external staff responsible for monitoring and auditing the project as a requirement of participating in the project.

Remote monitoring of recruitment activities at all sites will be carried out periodically by the study management team (on behalf of the CI & Sponsor) by exploring data, consent forms, delegation logs & recruitment logs copied to the CI in accordance with the study SOPs.

To facilitate this remote monitoring, and ensure timely and efficient transfer of data to the central study database, recruiting sites will be required to

- Transfer scanned (PDF) copies of completed data collection forms, consent forms & biochemistry results (via secure nhs.net email) to the CI site within 5 working days of the study visit.
- Transfer scanned (PDF) copies of recruitment logs & site specific delegation logs to the CI site on a monthly basis.

Monitoring reports will be stored in the study site file and copied to the Sponsor.

NHS Indemnity will apply.

12. Importance and potential benefit

Diabetes affects 6% of the adult UK population, accounts for 10% of NHS expenditure and is rapidly increasing in prevalence. Misclassification and subsequent suboptimal management are a widespread problem as reported by the Royal College of General Practitioners and NHS Diabetes (9), with an incorrect classification at diagnosis rarely revisited. This research therefore addresses a fundamental problem for one of the most common (and costly) conditions managed by the NHS, and has the potential to improve care for large numbers of patients. This project aims to directly improve patient care by for the first time integrating clinical features and biomarkers to allow clinicians to accurately assess classification and need for early insulin treatment at diabetes diagnosis. This will improve patient experience, outcomes and NHS costs by reducing inappropriate insulin treatment (where Type 2 diabetes is misdiagnosed as Type 1), and improving early glycaemic control and healthcare utilisation (where Type 1 diabetes is misdiagnosed as Type 2).

13 Public and Patient Involvement

The study team has access to the user representative group of the NIHR Exeter Clinical Research Facility (ECRF). In keeping with the NHS Patient Carer and Public Involvement (PCPI) strategy the ECRF invites user representatives to contribute to the development of various projects within its portfolio. These individuals have agreed to maintain contact and regular meetings have been established at which researchers discuss the development of current projects within the ECRF.

The ECRF user group have been actively involved in the study design and wording of the patient-facing documents for this study and the feasibility (pilot) study currently recruiting in Exeter.

Study progress will be overseen by a steering group which will include external lay and patient members.

14. Dissemination/implementation of research

Results will be written up and submitted for publication in a peer-reviewed journal. Abstracts will be submitted to national and international conferences. Results will be presented to clinical colleagues at regular in-house meetings. Written information in the form of a letter outlining the key findings of the study will be sent to all participants.

The output of this research will be immediately accessible post publication to clinicians and patients through a website calculator (www.diabetesgenes.org) and smartphone app (Diabetes Diagnostics).

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16. SCHEDULE OF AMENDMENTS

Amendment No.	Protocol version no.	Date issued	Author(s) of changes	Details of changes made
SA01	2.0	13jan17	Dr Angus Jones	Extend recruitment to patients diagnosed over the age of 50 who are insulin treated and recruit an additional 400 patients to this older cohort of patients.
SA02	3.0	19 Sept 17	Dr Angus Jones	Extend recruitment of the cohort diagnosed at >50 years to include 400 non-insulin treated patients to be recruited at selected sites. Remove the word 'young' from the full study title to reflect the inclusion of the older cohort and avoid any misunderstanding of inclusion criteria. Extend the recruitment period to 30th September 2018 and increase the number of recruiting sites.

				Funder details updated to include additional grant from Diabetes UK. Update the AE/SAE reporting procedures in line with Sponsor requirements and agreement. Update the means of contact for follow-up visits to include by post or in person. Promotion of the study on appropriate website and social media.
SA03	4.0	28 Aug 18	Dr Angus Jones	To extend the recruitment period to 31 December 2019. To modify recruitment criteria for cohort 1 to continue to recruit a further 200 participants receiving insulin treatment diagnosed after age 30 (total cohort 1 participants 1200), and cease recruiting other participants in this cohort. To include the option of capillary blood collection for C-peptide analysis at follow-up in accordance with participant preference.